

 $2^{\text{nd}}$  International Congress on Engineering and Life Sciences



ABSTRACT AND PROOCEDING BOOK

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# **R&D** Necessities of Superconductivity, from Applied Laboratory to Industrialization and University-Industry Cooperation Leading to a Sustainable Scientific and Technological Development

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**Abstract:** Superconductivity was discovered in 1911 by Kamerlingh Onnes during dc electrical resistance measurements by falling to zero at 4.2 K, and it is surely one of the most important discoveries of the 20th century. Since its discovery more than a century past, and there has been noteworthy scientific and technological developments. In this presentation, a historical development of superconductivity and milestones achieved will be mentioned to note the well-established applications in addition to futuristic potential applications. Especially, since the discovery of MgB2 by Jun Akimitsu in 2001, current status of MgB2 and an assessment of its potential applications in medical, transportation, and energy sectors will be provided. In this sense, in the field of superconductivity are to be discussed with comparison including the deployment of industry 4.0 program. Besides, within the concept of Turkey's 2023 vision, potential industrialization with novel opportunities and suggestions are to be mentioned.

Keywords: Superconductivity, MgB2, Technological Applications, Value-Added Products, Industry 4.0.



# **Cryogenic Issues with the Engineering Applications of Superconductors**

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**Abstract:** Cryogenics is well known as an enabling technology for the operation and applications of superconductors. Cryogenics started via air separation engineering in 19th century, and then Dutch Scientist Kamerlingh Onnes liquefied helium for the first time in 1908. This remarkable achievement has lead to the discovery of superconductivity in mercury cooled by liquid helium in 1911, by manifesting zero resistivity at 4.2 K and below. After many developments until 1960's, real applications were not available. By the discovery of A15 type II superconductivity, wire making developments were first established, and they have been cooled via Liquid He for the magnet science and technology leading to other Superconducting Magnetic Energy Storage, Superconducting Fault Current Limiter, MRI and NMR magnet applications, Propulsion Motor for the Navy Applications in addition to Particle Accelerator and Fusion Magnets. Recent developments in Cryogenics technology will be overviewed with some historical milestones.

**Keywords:** Superconductivity, Technological Advances in Cryogenics, Cryogenic Properties, Cryo-cooler Technology, Value-Added Products.



### The Effect of Smoking on Balanced Nutrition: Smoking Cessation Method

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Abstract: Nutrition is the consumption of required food for human growth and healthy. Nutrition should not be perceived as just food consumption. Balanced Nutrition is intake of necessary nutrients and energy for the growth, renewal and operation of the body. Smoking has many damages. The negative effects of smoking are also available on diet and eating habits. Carbon monoxide in cigarette has a negative effect on balanced nutrition. Smoking negatively impacts absorption of calcium and vitamin C. Smoking causes osteoporosis. It also increases the need of vitamin C. Smoking causes deficiency of vitamin A in the human body. Cigarette users have unhealthy nutrition. Because they do not consume enough fruits and vegetables. Smoking affects the taste of foods. Smoking causes a decrease in the consumption of high energy foods. It was observed that general liquid general liquid intake such as water, milk and fruit juices was quite low in smokers. adversities have been observed in some periods in body functions due to insufficient liquid intake. smoking is the biggest obstacle to a balanced nutrition due to numerous negativities. Smoking is an important psychosocial problem. Smoking habits are very common in Turkey and World. There are many methods for smoking cessation. One of these methods is behavioral procedure. This method aims to strengthen positively and includes self-directed procedures. This procedure is usually managed by leaders, pioneers or therapists. In this method, Self-control is very important. The awareness of the self-control is given consideration. Smoking cessation is done immediately. Support for smoking cessation is provided.

Keywords: Effect of Smoking, Balanced Nutrition, Smoking Cessation Method, Behavioral Procedure, Self-Control



### **Biomass Production and Carbon Sequestration Potential of Shrub Willow Plantations**

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**Abstract:** Shrub willow (Salix spp.) is a fast growing plant species, which are widely distributed in temperate regions, including Turkey. This species has been widely utilized in biomass and carbon sequestration projects in Northeastern and Midwestern US as well as in Europe. One of the uncertainties in the shrub willow biomass and carbon sequestration projects is the greenhouse gas contribution of the production system. This study investigated the biomass production and carbon sequestration potentials of 5-, 12-, 14- and 20-year old shrub willow plantations in Tully, New York, USA. To determine the above- and belowground biomass production, the aboveground biomass components were harvested and roots were excavated at 15-cm depth interval to 45-cm depth. The soil respiration rates, which is one of the main sources of uncertainties in carbon balance estimates, were also quantified using an automated soil CO2 flux machinery (LI-8100A). Results showed an increased biomass production with age, ranging from 6.81- to 20.99-Mg ha-1, which are equivalent to 12.48-Mg ha-1 CO2 equivalent (CO2e) and 38.48-Mg ha-1 CO2e, respectively. The average soil respiration rates (Rs) ranged from 27.2- to 35.5-Mg CO2 ha-1 yr-1. The soil respiration rates (Rs) also indicated continuous production, even when the soil is totally frozen, with the following proportions across the different seasons: 7 % in winter, 20% in spring, 46 % summer, and 27 % in fall. The Rs rates, however, are offset by the C sequestration in the above and belowground biomass components, suggesting that in a fully vegetated and undisturbed soil surface, the CO2 emissions from soils are totally recycled within the canopy of the growing vegetation.

Keywords: above- and belowground biomass, soil respiration rates, CO2 emissions, carbon balance, carbon sequestration.



### **Bamboo for Sustainable Livelihoods of Rural Communities**

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**Abstract:** Bamboo is a fast growing plant, renewable resource which is available in abundance in many parts of the world. Due to its properties, versatilities and high biomass production, it offers a great potential to meet an increasing demand for raw materials. In the tropics and other regions of the world, bamboo has been used as an alternative raw material in view of the scarcity and slow growing of timber from natural forests. This paper examines the important roles of bamboo resources utilization from a global perspective in helping to sustain the livelihoods of communities. It also assesses how several countries are working to find ways to utilize bamboo resources for sustainable development and to raise living standard of the people. With its multiple uses and high value for a range of products, bamboo can be utilized for value-added products, builds a variety of income and thus generates employment opportunities for the communities. For the low-income rural communities with little access to modern technology and investment capital, bamboo utilization offers significant advantages to improve their socio-economics status and hence contributes to poverty alleviation.

Keywords: Bamboo, livelihoods, communities.



# For a Liveable Future Work and Fight for a Land Degradation Neutral World

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**Abstract:** Land degradation is a complex phenomenon occurs in all land types under combined effects of many natural and anthropogenic factors. Since the future of natural resource sustainability will be drawn by the equilibrium between the speeds of land degradation and land restoration, the main target should be to reduce land degradation based environmental and socioeconomic vulnerability of people living on degraded lands. Turkey is one of the high-risk countries facing with severe land degradation and desertification problems because of climate changes and the increase in the pressure on land due to population increase and to meet demands for natural resources. The main goal of this paper is to define main types of land/soil degradation problems in Turkey, to drive attention on how to achieve land degradation neutrality in the country scale and to outline Turkey's efforts on combating land degradation and desertification.

Keywords: land degradation, land degradation neutrality, combating desertification, Desertification Model of Turkey.

#### **1. INTRODUCTION**

Land degradation processes limit ecosystem productivity more importantly in arid and semiarid regions. Arid and semiarid areas of Turkey is about 1/3 of the country's surface area and are highly fragile against land degradation and desertification.

Land degradation is a complex phenomenon occurs in all land types under combined effects of many natural and anthropogenic factors. In the UNCCD 2018-2030 Strategic Framework document, signed at the 13th Conference of the Parties (COP 13) held in Ordos, China on 6-16 September 2017, it was stated that desertification, land degradation and drought lead to economic, social and environmental problems such as poverty, health problems, food safety risk, biodiversity loss, water scarcity, decline in resistance to climate change and forced migration. It was also stated that these problems cause serious difficulties for the sustainable development of all countries, especially in the affected countries. The future of natural resource sustainability will be drawn by the equilibrium between the speeds of land degradation and land restoration (https://www.unccd.int). Therefore, it is important to assess, monitor and manage degraded lands in a way to neutralize land degradation.

Turkey is one of the high-risk countries facing with severe land degradation and desertification problems because of climate changes and the increase in the pressure on land due to population increase and to meet demands for natural resources.

### 2. WHAT IS LAND DEGRADATION NEUTRALITY (LDN)?

At the COP 12 held in Ankara, Turkey, in October 2015, the UNCCD Country Parties reached a breakthrough agreement on Land Degradation Neutrality (LDN) for better managing the land and restoring the land that have already degraded (UNCCD, 2016, 2017).

The 2030 Agenda for Sustainable Development, adopted by all United Nations Member States in 2015, provides a shared blueprint for peace and prosperity for people and the planet, now and into the future. The 15th goal of the 2030 Agenda for Sustainable Development is "Life on Land" and the Target 15.3 says, "By 2030, combat desertification, restore degraded land and soil, including land affected by desertification, drought and floods, and strive to achieve a land degradation-neutral world (UNCCD, 2016, 2017).

Land Degradation Neutrality (LDN) has been defined by the Parties to the Convention at the 12th session of the Conference of the Parties to the UNCCD (COP12) as: A state whereby the amount and quality of land resources, necessary

to support ecosystem functions and services and enhance food security, remains stable or increases within specified temporal and spatial scales and ecosystems (UNCCD, 2016, 2017). National governments have agreed to global and regional targets and commitments to halt and reverse land degradation and restore degraded land. Turkey one of the 122 countries which have already signed this agreement (UNCCD https://www.unccd.int/actions/ldn-target-setting-programme).

LDN is a simple idea and a powerful tool. It means securing enough healthy and productive resources by avoiding degradation whenever possible, better managing the land, and restoring the land we have already degraded. It is about achieving a balance between three processes: use/degradation, rehabilitation/restoration and sustainable land management (UNCCD, 2016, 2017).

#### 3. WHAT ARE THE MAIN LAND DEGRADATION TYPES IN TURKEY?

Turkey is one of the high-risk countries facing with severe land degradation and desertification problems because of climate changes and the increase in the pressure on land due to population increase and to meet demands for natural resources. Lands of Turkey suffer severely from degradation processes specifically by soil erosion, compaction, acidification, salinization, alkalization, organic matter loss, fertility decline and contamination. Moreover, surface sealing recently became the most threatened degradation factor because of increasing pressures by industrial developments, population growth and urbanization. Here are some causes and/or driving forces for soil/land degradation problems in Turkey:

- ✓ complexity in climatic conditions and high variability in distribution patterns of precipitation,
- ✓ unfertile soils with low organic matter (85% of the country's soil has low P and 90% has OM <3%),
- ✓ Shallow soil depth (only 15% of country's land has soil deeper than 90 cm),
- $\checkmark$  broken and improper topography (less than 20% of land with a slope gradient <6%),
- ✓ misuse of lands (use of agricultural land for other purposes and/or use of pasturelands or forestlands for cultivation),
- ✓ still running with conventional tillage systems, monoculture agricultural practices and fallow application
- ✓ burning plant residue
- ✓ limited application of no-till and minimum tillage practices
- ✓ soil tillage in steep lands (>20%),
- ✓ occupation of agricultural lands (land sealing)
- ✓ Land conversion from pasture & forest lands to farmland

#### 4. IS TURKEY SUCCESSFUL IN COMBATTING LAND DEGRADATION AND DESERTIFICATION?

In combatting desertification and land degradation Turkey has already prepared the National Strategy and Action Plan to Combat Desertification (2015-2023) and defined the planned actions and activities by different institutions in order to reach the 4 Strategic Objectives; (1) improving affected and likely to be affected population's living conditions, (2) improving affected and likely to be affected ecosystem conditions, (3) ensuring national and global benefit in conserving biological diversity and fighting climate change as well as in combating desertification and (4) mobilizing necessary resources to support the implementation of the UNCCD Convention by means of establishing an efficient partnership between national and international actors; in that regard, Turkey leading the bilateral, regional and global cooperation, sharing experiences and information with other parties (General Directorate of Combating Desertification and Erosion, 2015).

In Turkey, different Ministries, government agencies, institutions and units, universities and non-governmental organizations put great efforts to prevent desertification and land degradation at regional and national levels. Under this scope there are many and very important and successful completed or ongoing studies on natural resource conservation, sustainable land and natural resource management, adaptation to climate change and increasing ecosystem resistance. One of these projects is the development of Turkey's Land Degradation and Desertification Project and Risk Map (TÇM). The TÇM was developed in 2015 under the Project for the Development of the Basin Monitoring and Evaluation System (HIDS), which is implemented in cooperation with ÇEM and TÜBİTAK-BİLGEM. The validation and calibration studies of the TCM has already been completed. Using the Turkey's Desertification Model, it is possible to determine, map and monitor land degradation and desertification risk at watershed scale as well as to define priorities in the investment

projects and implementation activities integrated into the national development and government program on protecting natural resources of our country, minimizing occurrence of desertification, preventing rural migration and minimizing rural poverty (TÜBİTAK BİLGEM-YTE. 2017).

#### **5. CONCLUSION**

Making effective land use and management decisions have great influence on natural resource conservation. Soil's productivity and agricultural production strongly depend on the degree of soil degradation. Therefore, use of natural soil resources without leading soil degradation is extremely important for sustainability.

Strategies on developing innovative perspectives and techniques in preventing desertification and land degradation will play a significant role in sharing Turkey's efforts and success in combating desertification and land degradation on national and international platforms.

Turkey is a leading country on combating land degradation problems with its great knowledge, experience and technology. Turkey has completed or still running on many research and implementation projects on natural resource conservation, sustainable land and natural resource management, climate-friendly and best agricultural practices and increasing ecosystem resistance for preventing land degradation.

#### REFERENCES

- Çölleşme ile Mücadele Genel Müdürlüğü. 2017. Çölleşme ile Mücadele İlerleme Raporu 2016. T.C. Orman ve Su İşleri Bakanlığı. Çölleşme ile Mücadele Genel Müdürlüğü. Ankara.
- General Directorate of Combating Desertification and Erosion. 2015. National Strategy and Action Plan to Combat Desertification (2015-2023). Republic of Turkey, Ministry of Forestry and Water Affairs.
- TÜBİTAK BİLGEM-YTE. 2017. Havza İzleme ve Değerlendirme Sistemi (HIDS): Havza İzleme ve Değerlendirme Sistemi'nin Geliştirilmesi Projesi, Türkiye Çölleşme Modeli Raporu. Proje Kodu: G555000. TÜBİTAK Bilişim ve Bilgi Güvenliği İleri Teknolojiler Araştırma Merkezi (BİLGEM), Yazılım Teknolojileri Enstitüsü (YTE), Ankara.
- UNCCD. 2016. Achieving Land Degradation Neutrality at the country level Building blocks for LDN target setting. The Global Mechanism of the UNCCD. ISBN 978-92-95110-31-1 (e-copy).

UNCCD. 2017. The Convention. United Nations Convention to Combat Desertification. Official web page. http://www2.unccd.int/.

UNCCD. https://www.unccd.int/actions/ldn-target-setting-programme



### Local and Regional Endemic Plants of Aşağıkepen Village (Eskişehir-Sivrihisar)

#### Yağmur Noyan BAHADIR\*, Hayri DUMAN

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Abstract: This study was made on the purpose of introducing some local and regional endemic species (Scutellaria yildirimlii Çiçek & Yaprak-Lamiaceae, Aethionema dumanii M.vural & N. Adıgüzel-Brassicaceae, Salsola tucica Yıld.-Amanranthaceae, Sideritis gulendamiae H. Duman & Karavel.-Lamiaceae, Alyssum niveum Dudley-Brassicaceae, Achillea ketenoglui H. Duman-Asteraceae, Glaucium secmenii Yıld.-Papaveraceae), which are collected from during the field researchs in Aşağıkepen village as part of m. sc. thesis, which called "Aşağı Kepen-Ertuğrul-Kurtşeyh-Buzluca (Sivrihisar-Eskişehir) Köyleri Arası Florası". Plant samples were collected between 2017-2018 from the field researchs that made with especially considered flowering and fruiting times. Collectted samples were pressed and made dry according to herbarium rules, identified from relevant resources and recorded, and protected in GAZI Herbarium. Aşağıkepen Village and its environment is located in Upper Sakarya District, and in B3 square for Davis's Grid System Map. Area has marly and gypsum-bearing soil type. Gypsum (also known as gypsum rock) is white coloured chemical sediment rock. Gypsum rock is a mineral, whose chemical compound is calcium sulfate and encountered in sediment masses. Gypsum is generally found with marn, clay and sometimes with limy layers. Gypsum accumulation is rearly found in pure from. Although Turkey has a lower gypsum rate like %0.50, endemism rate is very high in these places. Despite the fact that gypsum-bearing areas were arisen after the ebb, endemic plants that found in these places are discernable in arid and subarid areas. These areas are generally not convenient for agriculture and animal breeding, therefore agriculture and animal breeding couldn't be done, and it makes natural protection for endemic taxa. According to the informations that we gave in the upper sentences, it can be understood that plants in marly and gypsum-bearing soils are special, and endemism rate is high in these places. We emphasise the importance of proctecting these natural areas, and local and regional endemic plants that special for these places in our study.

Keywords: Eskişehir, Sivrihisar, Aşağı Kepen, gypsum, flora

#### **1. INTRODUCTION**

Aşağıkepen Village and its environment is located in Upper Sakarya District, and in B3 square for Davis's Grid System Map. Area has marly and gypsum-bearing soil type. Gypsum (also known as gypsum rock) is white coloured chemical sediment rock. Gypsum rock is a mineral, whose chemical compound is calcium sulfate and encountered in sediment masses. Gypsum is generally found with marn, clay and sometimes with limy layers. Gypsum accumulation is rearly found in pure from. Although Turkey has a lower gypsum rate like %0.50, endemism rate is very high in these places. Despite the fact that gypsum-bearing areas were arisen after the ebb, endemic plants that found in these places are discernable in arid and subarid areas. These areas are generally not convenient for agriculture and animal breeding, therefore agriculture and animal breeding couldn't be done, and it makes natural protection for endemic taxa.

According to the informations that we gave in the upper sentences, it can be understood that plants in marly and gypsumbearing soils are special, and endemism rate is high in these places. We emphasise the importance of proctecting these natural areas, and local and regional endemic plants that special for these places in our study.

#### 2. MATERIAL AND METHOD

Plant samples were collected between 2017-2018 from the field researchs that made with especially considered flowering and fruiting times. Collectted samples were pressed and made dry according to herbarium rules, identified from relevant resources and recorded, and protected in GAZI Herbarium.

#### **3. RESULTS AND DISCUSSION**

#### Results

Most of the local and regional endemics like *Glaucium secmenii* Yıld., *Alyssum niveum* T. R. Dudley, *Aethionema dumanii* Vural & Adıgüzel, *Scutellaria yildirimlii* Çiçek & Yaprak, *Achillea ketenoglui* H. Duman, *Acantholimon riyadguelii* Yıldırım, *Sideritis gulendamiae* H. Duman & Karavel., *Cephalaria aytachii* Göktürk & Sümbül and *Salsola turcica* Yıld. that identified in last years from gypsum- bearing and marly habitats, which are located around the Aşağıkepen village, were confirmed. From these plants, type locality of *Glaucium seçmenii* Yıld., *Scutellaria yildirimlii* Çiçek & Yaprak, *Acantholimon riyadguelii* Yıldırım, *Sideritis gulendamiae* H. Duman & Karavel., *Cephalaria aytachii* Göktürk & Sümbül and *Salsola turcica* Yıld. is environment of Aşağıkepen village.

#### Discussion

Aşağıkepen village, which has gypsum-bearing and marly habitat type, is very important in terms of biodiversity. Because of this, loss of habitat for the sake of gaining grazing and agriculture territory will not be allowed, and special areas like this habitats must be in protected status.

#### 4. CONCLUSION

Gypsum-bearing and marly habitats are very important and special for endemic plant diversity.

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#### REFERENCES

- Aytaç, Z., Koruklu, T., Kaptaner İğci, B. (2017). Acantholimon gemicianum (Plumbaginaceae), a New Species from Turkey. Annales Botanici Fennici, 54(1-3), 83-88.
- Çakır, E. G., (2018). İliç-Kemah (Erzincan) Jips Florası. Doktora Tezi, Gazi Üniversitesi Fen Bilimleri Enstitüsü, Ankara, 1-188.
- Çiçek, M. and Yaprak, A. E. (2013). Scutellaria yildirimlii (Lamiaceae), a new species from Turkey. Phytotaxa, 132. (1): 53-58.

Davis, P. H., "Flora of Turkey and East Aegean Islands", Edinburgh Unv. Press., Edinburgh, Vol.1-9 (1965-1985).

- Davis, P. H., Mill, R. R. and Tan. K. (Editörler) (1988) Flora of Turkey and East Aegean Islands, Vol. 10, Edinburgh, Edinburgh Unv. Press., 1-303.
- Duman, H., Aytaç, Z., Ekici, M., Karavelioğulları, A.F., Dönmez, A., Duran, A., (1995). Three new species (Labiatae) from Turkey. Flora Mediterranea, 5, 221-228.
- Göktürk, R. S. and Sümbül, H. (2003). Cephalaria aytachii (Dipsacaceae), a New Species from Central Anatolia, Turkey. Annales Botanici Fennici 40: 123-127.
- Güner, A. (Editör). (2014). Resimli Türkiye Florası cilt 1. İstanbul: Nezahat Gökyiğit Botanik Bahçesi Yayınları, 1-512.
- Güner, A. (Editör). (2018). Resimli Türkiye Florası cilt 2. İstanbul: Nezahat Gökyiğit Botanik Bahçesi Yayınları, 1-800.
- Güner, A., Aslan, S., Ekim, T., Vural, M. Ve Babaç, M. T. (Editörler). (2012). Türkiye Bitkileri Listesi (Damarlı Bitkiler). İstanbul: Nezahat Gökyiğit Botanik Bahçesi ve Flora Araştırmaları Derneği Yayını, 1-887.
- Güner, A., Özhatay, N., Ekim, T. and Başer, K. H. C. (Editörler). (2000). Flora of Turkey and the Aegean islands, Vol. 11, Edinburgh, Edinburgh Unv. Press.
- Yıldırımlı, Ş. (2010). Some new taxa, records and taxonomic treatments from Turkey. OT Sistematik Botanik Dergisi, 17(2), 1-114.
- Yıldırımlı, Ş. (2012). The heaven of gysophilous phytodiversity of Turkey: Kepen, Sivrihisar, Eskişehir, Turkey, 13 taxa as new. OT Sistematik Botanik Dergisi, 19(2), 1-52.



# Type 2 Diabetes Mellitus and Microbiota

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**Abstract:** The aim of this study was to explain the relationship of Type 2 Diabetes Mellitus (T2DM) with the microbiota and to determine whether the probiotic supplements could improve the metabolic profiles by modifying the intestinal microbiota in T2DM.

Worldwide, obesity prevalence is increasing day by day. The prevalence of T2DM, which is one of the diseases associated with obesity, increases accordingly. The relationship between intestines and nutrition play a role in most of the vital homoeostatic functions of the human body. Recently, the role of the intestinal microbiome in T2DM pathogenesis has been investigated. There is clear evidence that the intestinal microbiota affects the host through various mechanisms.

Increased evidence suggests that various mechanisms associated with bowel barrier disorders cause the onset of lowgrade inflammation, which characterizes metabolic disorders. Furthermore, the accumulation of intestinal bacterial inflammatory molecules in the intestine is thought to accelerate inflammation in T2DM.

T2DM, one of the most obesity-related disorders, is associated with abnormal energy metabolism and low-level chronic inflammation in adipose tissues. Increased weight gain seems to be one of the factors triggering this low-grade inflammation. It has been suggested that changing microbiota in obesity causes chronic low-grade inflammation, pathogenesis of insulin resistance and increased metabolic endotoxin secretion leading to the onset of T2DM.

Abnormal metabolic profile in diabetics, proinflammatory factors, and oxidative stress; it causes the increased risk of cardiovascular disease, diabetic complications, and hypertension. In this regard, it has been reported that the microbiota improves the metabolic profile with the immunomodulatory and anti-inflammatory effects of probiotics.

Keywords: Type 2 diabetes mellitus, obesity, intestinal microbiota, probiotic, prebiotic, sinbiotic.



# Effect of İnsecticide on Pollen Germination in in-vivo Conditions On Pear (*Pyrus communis*) Plant

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**Abstract:** Tree conditions, chemical applications and environmental factors are very important in pear cultivation. Pear is a stable and high yield product although; production due to intense cultural application due to some diseases and pests it has a lower share of 18% compared to apple. In pears, there are both flower and leaf places in one bud. Thus, when the flowers are opened, there is foliation on the tree. Reproduction of flowering plants depends on delivery of the gamet to the egg, which occurs through a long, polarized projection of a pollen cell, called the pollen tube. The pollen tube grows at its tip, and this growth is very fast extended lengths. Deltamethrin is a synthetic insecticide paralyzes the nervous system give a quick knockdown effect. In this study, the effects on pollen germination of Deltamethrin, which is used for insect control in Pyrus plants, are investigated in in vivo conditions.

30 cm length of flowering branches of the pear tree was cut. Branches of the tree were treated with different doses of Deltamethrin. 24 hours laterThe flowers collected were lightly tapped with a brush so that they shed pollen was collected on a medium. The pollen of Prunus varieties was cultivation in 2% agar, 15% sucrose medium. After 4 hours' incubation, germination and development of pollen tubules were observed in light microscope.

The chemicals such as deltamethrin that are used affect not only the environmenthall problems but also the reproductive mechanisms that occur in plants. Depending on the presence of deltamethrin, the germination of the pollen tube was affected at different levels. Pure deltamethrin was stopped pollen germination completely. In order to increase fruit production in agriculture, such chemical applications should be carreful done in plant flowers; pollen germination at seasonal time should be considered.

### **Keywords:**



### The Taxonomy and Biology of Order Trichoptera (Insecta)

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**Abstract:** The order Trichoptera is one of the densest order on account of aquatic larvae in ecosystems. It is the only order that lives in freshwater and is in subjection to this habitat in larvae and pupae periods among the insect orders that undergo holometabolous metamorphosis. Most of the Trichoptera larvae live in each zone from the spring regions of the rivers to the potomal zones where they are poured into the sea. There are also Trichoptera species that live in lakes besides rivers and these consist of %20 of total species numbers. The adults are terrestrial and usually fly over riverside and coastal plants. 49 families, 616 genus and 14548 species of them are known over the world in total. The members of the Trichoptera order are one of the most important bioindicator models among the aquatic insect groups. As sedimantation, industrial pollution, mine and agriculture wastes, sewerage system wastes and acid rains gather on the surface of water, this living beings are damaged by this pollutants. The larvae of different species are used to evaluate the water quality in the various pollution level. For this reason, the Trichoptera orders ise seen as a biologic following device which is used to define the water quality. Especially in developed countries such as North America, Europa and Australia, this method is preferred. In this study, a compile is made about the life cycle of Trichoptera orders which is one of the most important benthic macroinvertabrates and used commonly in environmental quality evolations, following the freshwater ecosystems. The comments and findings on the larvae, pupae and adult stages of Trichoptera order are given by compiling.

Keywords: Trichoptera, biology, larvae, pupae, adult

#### **1. INTRODUCTION**

The Trichoptera order (Caddisfly) are holometabol insects. Larval stage aquatic adult stage terrestrial Trichoptera's developmental stages; egg, larvae, pupae and adults take place in four stages (Thorp and Rogers, 2011) (Figure 1). Trichoptera is closely related to Lepidoptera (butterfly and moth), which have different shapes of scales covering their wings to a certain extent. It is considered as a sister order because of its similarity to moths (Monson, 1994).



Figure 1. Trichoptera life cycle

Trichopteras are very important benthic macroinvertebrates in terms of feeding dynamics and energy flow in lentic and lotic waters (Resh and Rosenberg, 1984). These organisms are severely damaged by biotic and abiotic pollutants. Because deposition, environmental pollution, pesticides, household wastes, anthropogenic effects and acid rains accumulate on the surface of the water, especially Trichoptera larvae adversely affect (Resh and Unzicker 1975; Resh, 1993; Dohet, 2002; Brown et al., 2007). For this reason, the Trichoptera orders assumes the role of biomonizer in the quality determination of lentic and lotic waters. In some developed countries (America, Europe and Australia) this method is used (Pauls et al., 2008).

In research, the oldest Trichoptera fossils have been found in the upper Triassic. The earliest aquatic stage date of the Trichoptera team coincides with Late Jurassic time. Trichoptera differences increased in Jurassic and lower and middle Jurassic remains belong to several extinct families. The main line of distinction in the history of Trichoptera development is seen in late Jurassic and Cretaceous (Holzenthal et al., 2007). 78 genera and 488 species belonging to 7 families are known as fossils (Moor and Ivanov, 2008).

The first studies on Trichoptera were found in Systema Naturae, Linnaeus (1758). Here, 18 "Neuroptera beetles" have been identified in the genus Phyrganea, but 4 of them were included in the Trichoptera order, along with 7 species identified by Linnaeus. Trichoptera as the order name was first used by Kirby. As a result of recent researches, Trichoptera is known to be represented by 49 families, 6184 genera and 14584 species worldwide (Morse, 2017). There are also 12 fossil families, 125 fossil genera and 685 fossil taxa. It is estimated that the world Trichoptera fauna consists of at least 50000 species (Schmid, 1984). The Trichoptera orders in Turkey 22 families and 500 (461 species, 39 sub type) is represented by the aquatic insect orders by type of group (Darılmaz and Salur, 2015; Küçükbasmacı and Kıyak, 2017; Sipahiler, 2016, 217, 2017b, 2018, 2018b). Of these, 217 species group taxa and 2 genera are endemic to Turkey. Turkey has been a lot of research on the Trichoptera. However, these studies were mostly on adults and not much studies on larvae. Because of the importance of their role in Trichoptera stream habitats, many studies contain information on their biology. According to the literature studies, the first records of Trichoptera orders of our country were found in the study of Sipahiler and Malicky in 1987. In this study, *Tinodes manni* McLachlan, 1878 species were collected from Bursa in 1851, 2  $\sqrt[3]{}}$  specimens were collected and stored in the Vienna "Naturhistorisches Museum" information was given (Sipahiler and Malicky, 1987).

### 2. ADULT

Adult Trichoptera live on land and resemble small butterflies as they navigate coastal vegetation (Photo 1). They are often found in large numbers in lakeside or stream habitats, although they are rarely encountered by collectors. Both pairs of wings and body are covered with feathers or hairs. Trichoptera adults are usually brown, gray, yellow, beige or other showy shades or combinations of these colors. This coloration is probably an adaptation that allows them to hide during the day in vegetation. However, a number of species are yellow, red, orange, green, silver, blue or sometimes iridescent. Bright colours are available on the wing (Rosenberg and Resh, 1984) and chest covered with feathers and scales. In both cases, the different patterns of different colored hairs on the wings allow for the identification of species, as is often the case for more colorful Lepidoptera (Butterfly) relatives (Holzenthal et al., 2007).



Photograph 1. Philopotamus montanus

The most important distinguishing feature of adult Trichopteras is the presence of different hairs, a form of hair on both wings. The head and chest also carry a different number of hairs. The presence of hairs instead of scales on the wings, the absence of a wrapped hose of an adult butterfly and the mouth structure of the chewy form is separated from the butterflies.

Adult Trichopteras body length varies in some species may be up to 2-2.5 cm (Thorp and Rogers, 2011). In some species, the body length (Hydroptilidae and some Glossosomatidae species) 1.5 mm-3 mm, Phyrganeidae family can reach up to 4.5 cm (Holzenthal et al., 2007). During rest in Trichoptera, the wings are kept in the form of a roof over the abdomen (Thorp and Rogers, 2011). Most species are nocturnal, often flying around light, while in daylight they are hidden (Imes, 2000).

The head structure and patterning of the adult Trichoptera, the details in the abdomen and dorsal, the wing structure (vascularization and wing length), legs and genital structures are important characters used to identify species. All these components of their morphology, but in particular the characteristic structures of the antennas and palpus, the presence or absence of dot eyes (osellus), the number of tibia spines, and the vascularization of wings are used to describe various families and genera. The male genitalia used to identify genera and species in the 9th and 10th abdominal segments of adult males stands out (Ivanov, 2005; Morse, 1975; Nielsen, 1957; Ross and Unzicker, 1977; Schmid, 1970, 1979, 1989). In the 9th abdominal segment of the male, there is tergum and sternum that boil to a sclerotized ring; pleural scleritis is absent (8th segment is the last typical abdominal segment with a prominent tergum and sternum and membranous pleural regions). The 10th segment is much shorter than the 9th segment and its shape is highly variable. It can be completely membranized, but is typically found dorsally and laterally sclerotized, concave, reduced, membranized or ventral (Holzenthal et al., 2007).

There is no real ovipositivity in females. But instead, the terminal abdominal segments (segments 8-10 or 11) are either elongated into a protruding oviscapt to place eggs on the substrate, or formed into a shorter apparatus for forming and retaining egg mass. The female genitals (Nielsen, 1980) are very simple compared to men, but internal structures that hold the top of the phallus during copulation and accept male ejaculate are often complex and species-specific.

The wing provides movement of adults, but also includes a number of adult morphological features. The front wings are longer than the rear wings, but the rear wings are wider than the front wings. There are secondary sexual characters on male wings. Wing vein formation is primitive (Holzenthal et al., 2007) (Photo 2).



Photograph 2. Adult Trichoptera wing structure, (a) front wing, (b) rear wing

Adult mouth pieces are not well developed. Mandibules are absent, very dull or not functional. However, at least they act as biting organs. However, the maxillary and labial palpes are prominent and consist of 5 and 3 parts, respectively. In males of some species, maxillary palpus may cause segmentation reduction and secondary sexual changes or both (Holzenthal et al., 2007). The most important feature of the mouth parts of adult Trichoptera is haustellum (Crichton, 1957; 1991; 1993), a unique structure for the order Trichoptera. The Haustellum consists of the fusion of the labium (prelabium) and the apex of the hypopharynx to form a short proboscis. The haustellum is of membrane structure at the top and has channels consisting of thin rows of hairs that help to soak up water or sugary liquids.

In adults, compound eyes are well developed. However, in a few groups, the compound eyes of males are significantly larger than females. Three simple eyes or ocelli are found in several families (Photo 3).



Photograph 3. Adult Hydropsyche spp. compound eye

In most species, the antennae are filamentous in shape and are as long as the body length, but in some families they can also be several times longer than the body (Photo 4). Scape is the widest part of the antenna and is much larger than that of males of many species and has a modified hair, scales and glandular structure (Holzenthal et al., 2007).



Photograph 4. Adult Trichoptera antenna structure

Prothorax is small and underdeveloped compared to wing-bearing pterothorax (consisting of mesothorax and metathorax). Each of the mesothorox and metatorax contains taxonomically important coxal and pleural scleritis, wart-bearing scleritis and internal apodems (Tindall, 1965). The head, especially in males, may have additional structures such as patches of modified scales or bristles. In particular, the head contains tentorium, which varies in shape and complexity between various families (Neboiss, 1991b). The number of segments of maxillar palpuses used in the diagnosis of species ranges from 1-5 gender (Malicky, 2004). Labial palpi is 3-segment (Holzenthal et al., 2007).

The legs are long and thin, the front legs are the shortest of the legs. However, some females have tibia and tarsi on their middle legs to allow them to lay eggs by swimming below the water surface. The number of tibia spines in the legs varies according to the species. The tibia spines, which are thorn-shaped, are an important taxonomic character (Baszio and Richter, 2002), and their numbers are important in the determination of species. The tibia spines in the legs are 0-3 (1 preapic, 2 apical) in the anterior leg and 1-4 (2 preapic, 2 apical) in the other legs (Schmid, 1998).

Studies have shown that some species of Trichopteras are univoltine, but in warmer temperate climates, they occur in two generations (bivoltine) of species during the year. It is also known that the emergence of adult Trichopteras in the higher mountain regions can take up to 2 years (Thorp and Rogers, 2011). Although the life cycle of adult Trichopteras is short (two weeks), there are species that survive for up to a month (Ross, 1944). Mating in adults occurs on the ground, near vegetation, while flying. The incubation period of the eggs is about two or three weeks (Gould, 2012).

### 3. LARVAE

Trichoptera larvae are found in fresh water, cold and fast flowing streams. Scientists interested in environmental research are focusing on Trichoptera larvae to assess the water quality of water resources, as Trichoptera larvae are very sensitive to many pollutants (Grinager, 2009).

Trichoptera larvae usually have a thin flexible structure and the larvae have very short antennae and simple eyes. The larvae of the larvae, which have a different head, chest and abdomen structure than adults, are completely well developed and sclerotized. The larval eyes consist of several stemmata (Holzenthal et al., 2007).

Unlike adults, the larval mouth parts are well developed and consist of a small labrum, a pair of well-developed mandible, a pair of short, compact maxillary and a labium. Maxilla and labium form a composite structure. The larvae usually have short maxillary palpus and labial palpus (Photo 5).



Photograph 5. Mouth parts of Hydropsyche bulbifera

Each thoracic segment of the larvae is different and carries a double segmented leg (Photo 6). These legs are almost the same length or the hind legs are longer than the front legs (Holzenthal et al., 2007).



Photograph 6. Hydropsyche instabilis (lateral view), thoracic segments and legs

In Hydropsychidae and Hydroptilidae, mesothorax and metatorax are significantly sclerotized, but in other families these segments are completely membranized or less sclerotized (Photo 7).



**Photograph 7.** a) *Hydropsyche bulbifera*, sclerotized prothorax, mesothorax and metatorax, b) *Philopotamus montanus*, sclerotized protorax, membranized mesothorax and metatorax

Trichoptera larvae have five instar stages, but some species are known to have six or seven instar stages, though rare (Holzenthal et al., 2007). Larvae perform their respiration by diffusion from the body surface or with abdominal gills in the abdomen (Thorp and Rogers, 2011). The fringed feathers on the legs help to collect food in water in some families (Brachycentridae), while in some families (Leptoceridae) it helps to swim. Larvae of Trichoptera; leaves, algae, benthic macroinvertebrates can be fed by hunting or collecting organic debris. The development of larvae is influenced by the nutritional quality provided by forests along the banks of the river (Fiance, 1978; Malley, 1980; Rodgers, 1984). Domestic larvae often use plants to feed, while larvae make their nets against water flow by feeding algae, bacteria or debris from the flow (Brown, 2008). In the free-living larvae, feeding takes place completely by hunting other creatures (Borror et al., 1989).

Larvae have the ability to make a silky web obtained from salivary glands (Mackay and Wiggins, 1979; Wiggins, 2004). Trichoptera larvae produced by this network and invertebrate animals between the spider web as an alternative to a structure that can produce a network (Michalak et al., 2005). In some groups, silk is secreted from the top of the labrum, which is a thin, protruding structure. This silk secretion from a pair of labial glands reaches a flawless structure in Amphiesmenoptera, which includes teams of Trichoptera (Caddisfly) and Lepidoptera (moths and butterflies) (Akai et al., 2003).

These networks, taught by trichopter larvae, are fascinating structures for entomologists. Trichoptera is one of the most abundant insect species in lotic waters, and these silky nets produced by larvae are largely involved in providing this diversity (Mackay and Wiggins, 1979). This network consists of polymerized protein fibers known as silk (Sehnal and Akai, 1990; Craig, 1997). This silk secretion produced by Trichoptera larvae is an enormous morphological adaptation that enables larvae to survive in almost any freshwater ecosystem (Wiggins, 1996; 2004). Many Trichoptera larvae build these silky nets in fast flowing rivers. Each species has its own net mesh shape and diameter. The location of the network depends on the environmental conditions (Michalak, 2005). Whatever the conditions of hydrology, the networks are often mesh-shaped (photo 8) and highly intriguing in patterns (Michalak, 2005).



Photograph 8. Trichoptera network construction; a) aquarium original, b) Daday brook (Kastamonu) original

The network is not the only function of the salivary glands. Without this web produced by the larvae, the larvae are unable to produce the house. Many Trichoptera larvae form portable households of various forms and shapes using currents and sediment materials, sand grains, plant waste (Cianficconi and Moretti, 1991 b; Moretti and Cianficconi, 1992) (Giller and Malmqvist, 1998; Flory and Milner, 1999; Lepneva, 1998; Lepneva; Tomaszewski, 1981; Nepomnyashchikh, 1992). Trichoptera larvae carefully select material choices for domestication using well-defined larval behaviors (Hansell, 1968; Stuart and Currie, 2002).

The larvae of Trichoptera also use this network to produce pupae. This produced web is an essential component in pupal construction. Every Trichoptera larva in the fifth instar phase forms pupae using this network. Because no larvae that can not pass the pupal stage and can not make pupal stage can not pass to the next stage of adulthood (Michalak, 2005).

### 4. PUPAE

In Trichoptera pupae, antennas, legs and developing wings are exarct types separate from the body. Mandibules usually apically cross each other and extend forward. The antennas extend along the chest and abdomen towards the abdomen and sideways. In species with a long antenna, the antennas curl towards the end of the abdomen (Photo 9). Chest unchanged, but thoracic legs usually have swimming hairs. There are remains of larval gills on the abdomen. Abdomen ends with a pair of anal sections that help identify different families with the number, arrangement and morphology of the hook plates and mandible morphology (Holzenthal et al., 2007).



#### Photograph 9. Hydropsyche spp. Pupae

The majority of pet insect pupae are experts in swimming; they use their legs in a shovel-like manner rather than all their bodies for swimming (Wallace, 2003). It is known that some Trichopter larvae pass into the pupal phase to overcome these difficult conditions in severe winter conditions or when environmental conditions become difficult. This lasts for two to three weeks. Adults who are ready to get out of pupal case use mandibules to get out of pupal sheath (Holzenthal et al., 2007).

#### REFERENCES

- Akai, H., Hakim, R. S., Kristensen, N. P., 2003. Exocrine glands: saliva and silk. In: Kristensen NP (ed) Lepidoptera: moths and butterflies 2. Handbuch der Zoologie [Handbook of Zoology] IV. Walther de Gruyter, Berlin, Vol 36, pp 377–388
- Baszio, S., Richter, G., 2002.Ultrastructural characters of adult leg armature in Trichoptera (Insecta) and their potential for use in phylogenetic analysis. *Aquatic Insects*, 24, 1–20.
- Bohle, H.W., 1983. Drift-catching and feeding behaviour of the larvae of *Drusus discolor* (Trichoptera: Limnephilidae). *Archiv für Hydrobiologie*, 97, 455–470.
- Borror, D. J., Triplehorn, C. A., and Johnson, N. F., 1989. An Introduction to the Study of Insects. 6th edition, Philadelphia, Saunders College Publishing.
- Brown, L. E., Hannah, D. M., and Milner, A. M., 2007. Vulnerability of alpine stream biodiversity to shrinking glaciers and snowpacks. *Global Change Biology*, 13 (5), 958-966.
- Cianficconi, F., Moretti, G. P., 1991b. Biologia, morfologia e geonemia di *Catagapetus nigrans* McL. *Mem Soc Ent Ital Genova* 70 (2): 225–238.
- Craig, C. L., 1997. Evolution of arthropod silks. Annu Rev Entomol 42:231-267
- Crichton, M. I., 1957. The structure and function of the mouth parts of adult caddis flies (Trichoptera). Transactions of the Royal Entomological Society of London, Series B, Biological Science, 241, 45–91.
- Crichton, M. I., 1991. A scanning electron microscope study of the mouth parts of adult Phryganea grandis (L.). In: Tomaszewski, C. (Ed.) Proceedings of the 6th International Symposium on Trichoptera. Adam Mickiewicz University Press, Poznan, Poland, pp. 329–333.
- Crichton, M. I., 1993. A scanning electron microscope study of the mouth parts of some adult Limnephilidae (Trichoptera). In: Otto, C. (Ed.) Proceedings of the 7th International Symposium on Trichoptera. Backhuys Publishers, Leiden, The Netherlands, pp. 45–48.
- Darılmaz, M. C., and Salur, A., 2015. Annotated Catalogue of the Turkish Caddisflies (Insecta: Trichoptera). Munis Entomology & Zoology, 10 (Suppl.), 521-734
- De Moor, F. C., and Ivanov, V. D., 2008. Hydrobiologia. 595:393-407 DOI 10.1007/s10750-007-9113-2
- Dohet, A., 2002. Are caddisflies an ideal group for the biological assessment of water quality in streams? Nova Supplementa Entomologica (Proceedings of the 10th International Symposium on Trichoptera), 15, 507–520.
- Fiance, S. B., 1978. Effect of pH on the biology and distribution of Ephemerella funeralis (Ephemeroptera). Oikos, 31, 332-339.
- Flory, E. A., and Milner, A. M., 1999. Influence of riparian vegetation on invertebrate assemblages in a recently formed stream in Glacier Bay National Park, Alaska. J. N. Am. Benthol. Soc, 18, 261-273.
- Giller, P. S., and Malmqvist, B., 1998. The biology of stream and rivers. Oxford, 296p.
- Gould, J. L., and Gould, C. G., 2012. Animal architects: building and the evolution of intelligence. New York, NY: Basic Books.

Grinager, J., 2009. Caddisfly Larvae Visual System: Response to Light. Madison, WI: University of Wisconsin.

- Hansell, M. H., 1968. The house building behaviour of the caddisfly larva Silo pallipes Fabricius: II. Description and analysis of the selection of small particles. Anim Behav. 16:562–577.
- Holzenthal, R. W., Blahnik, R. J., Prather, A. L., Kjer, K. M., 2007. Order Trichoptera Kirby, 1813 (Insecta), Caddisflies, Zootaxa, 1668, 639-698.
- Imes, R., 2000. "Beginner's Guide To Entomology". Cancellor Press. London, 151
- Ivanov, V. D., 2005. Ground plan and basic evolutionary trends of male terminal segments in Trichoptera. In: Tanida, K. & Rossiter, A. (Eds.) Proceedings of the 11th International Symposium on Trichoptera. Tokai University Press, Kanagawa, Japan, pp. 207– 218.
- Küçükbasmacı, İ., and Kıyak, S., 2017. A study on the Caddisfly fauna (Insecta: Trichoptera) of Kastamonu and a new species record for Turkey. Munis Entomology & Zoology, 12 (2), 486-499
- Lepneva, S. G., 1971. Fauna of The USSR. Trichoptera 1, Larvae and Pupae of Annulipalpia. Translation from 1964 Russian edition. Jerusalem, Israel Program For Scientific Translations, 1-638.
- Mackay, R. J., and Wiggins, G. B., 1979. Ecological diversity in Trichoptera. Annual Review of Entomology, 24, 185-208.
- Malicky, H., 2004. Atlas of European Trichoptera (second ed.). Dordrecht, Netherlands: Springer.
- Malley, D. F., 1980. Decreased survival and calcium uptake by the crayfish Orconectes virilis in Low pH. Can. J. Fish. Aquat. Sci., 37, 364-372.
- Michalak, M., Tszydel, M., Bilska, J., and Krucińska, I., 2005. FIBRES and TEXTILES in Eastern Europe January / December Vol. 13, No. 6 (54)
- Monson, M. P., 1994. The Caddisflies (Insecta: Trichoptera) of the Lake Itascaregion, Minnesota, And A Preliminary Assessment of The Conservation Status of Minnesota Trichoptera, M. Sc. Thesis, Submitted to the Faculty of the Graduate School of the University of Minnesota. Minesota.
- Moretti, G. P., and Cianficconi, F., 1992. Un caso di colonizzazione ipogea nelle Alpi Orientali sostenuta da Philopotamus ludificatus McL. Mem Soc Ent Ital Genova 71 (2): 637–648.
- Morse, J. C., 1975. A phylogeny and revision of the caddisfly genus Ceraclea (Trichoptera, Leptoceridae). Contributions of the American Entomological Institute, 11, 1–97.
- Morse, J., 2017. Trichoptera World Checklist [Online]. [Accessed 12 January 2017].
- Neboiss, A., 1991b. Comparative study of tentorial structures in caddis-flies (Trichoptera). In: Tomaszewski, C. (Ed.) Proceedings of the 6th International Symposium on Trichoptera. Adam Mickiewicz University Press, Poznan, Poland, pp. 283–290.
- Nepomnyashchikh, V. A., 1992. The Control of Selection of Particles for Case Building by Larvae of Chaetopteryx Vilosa Fabr. (Trichoptera, Limnephilidae). In: Proc. 7th Int. Symp. On Trichoptera, (Ed.): C. Otto, Bachhuys Pub., Umea, 263-265
- Nielsen, A., 1957. A comparative study of the genital segments and their appendages in male Trichoptera. Biologiske Skrifter Danske Videnskabernes Selskab, 8, 1–159.
- Nielsen, A., 1980. A comparative study of the genital segments and the genital chamber in female Trichoptera. Biologiske Skrifter Danske Videnskabernes Selskab, 23, 1–200.
- Pauls, S. U., Graf, W., Haase, P., Lumbsch, H. T., Waringer, J., 2008. Grazers, Shredders and Filtering Carnivores The evolution of Feeding Ecology in Drusinae (Trichoptera: Limnephilidae): Insights from a Molecular Phylogen, Molecular Phylogenetics and Evolution, 46, 776-791.
- Resh, V. H., and Rosenberg, D. M., 1984. The ecology of aquatic insects. New York: Praeger Publishers.
- Resh, V. H., and Unzicker, J. D., 1975. Water quality monitoring and aquatic organisms: the importance of species identification. Journal Water Pollution Control Federation, Washington, 47, 9-19.
- Rodgers, D. W., 1984. Ambient pH and calcium concentration as modifiers of growth and calcium dynamics of brook trout, Salvelinus fontinalis. Can. J. Fish. Aquat. Sci., 41, 1774-1780.
- Ross, H. H., 1944. The caddisflies or Trichoptera of Illinois. Bulletin of the Illinois Natural History Survey, 23, 1-326.
- Ross, H. H., and Unzicker, J. D., 1977. The relationships of the genera of American Hydropsychinae as indicated by phallic structures (Trichoptera, Hydropsychidae). Journal of the Georgia Entomological Society, 12, 298–312.
- Schmid, F., 1970. Le genre Rhyacophila et la famille des Rhyacophilidae (Trichoptera). Memoires de la Société Entomologique du Canada, 66, 1–230.

- Schmid, F., 1979. On some new trends in trichopterology. Bulletin of the Entomological Society of Canada, 11, 48-57.
- Schmid, F., 1984. Essai d'evaluation de la faune mondiale des Trichopteres. In: MORSE, J. (ed.) Proceeding of the 4th International Symposium on Trichoptera. The Netherland: Dr. W. Junk Publisher.
- Schmid, F., 1989. Les hydrobiosides (Trichoptera, Annulipalpia). Bulletin de l'Institute Royal des Sciences Naturelles de Belgique, Entomologie, 59, Supplement, 1–154.
- Sehnal, F., Akai, H., 1990. Insects silk glands: their types, development and function, and effects of environmental factors and morphogenetic hormones on them. Int J Insect Morphol Embryol 19:79–132
- Sipahiler, F., 2017a. "Four new species of the genus Kelgena Mey from Turkey (Trichoptera: Limnephilidae, Chaetopterygini)". Nova Acta Científica Compostelana (Bioloxía), 24, 13-20.
- Sipahiler, F., 2017b. "Descriptions of two new species of the genus Psilopteryx Stein from Turkey (Trichoptera, Limnephilidae, Chaetopterygini)" Nova Acta Científica Compostelana (Bioloxía), 24, 69-73.
- Sipahiler, F., 2018a. "Three new species of caddisflies (Trichoptera Hydroptilidae, Leptoceridae) from Turkey and faunistic list for the Seyhan and Ceyhan rivers". Nova Acta Científica Compostelana (Bioloxía), 25, 37-43.
- Sipahiler, F., 2018b. "Studies on the males of the genus Philopotamus Leach in Turkey (Trichoptera, Philopotamidae)". Nova Acta Científica Compostelana (Bioloxía), 25, 55-70.
- Sipahiler, F., Malicky H., 1987. "Die Köcherfliegen der Türkei (Trichoptera)". Entomofauna, 8: 77-165
- Stuart, A. E., Currie, D. C., 2002. Behaviour is not reliably inferred from end-product structure in caddisflies. Ethology. 108:837-856.
- Thorp, J. H., and Rogers, D. C., 2011. Caddisflies: Insect Order Trichoptera. Field Guide to Freshwater Invertebrates of North America, Chap, 25.
- Tindall, A. R., 1965. The functional morphology of the thorax of Limnephilus marmoratus Curtis (Trichoptera: Limnephilidae). Transactions of the Royal Entomological Society of London, 117, 127–166.
- Tomaszewski, C., 1981. The Principles of Case Building Behavior in Trichoptera. In: Proc. 3rd Int. Symp. on Trichoptera, ed. G.P. Moretti, Junk, The Hague, 365-373.
- Wallace, I., 2003. The Beginner's Guide to Caddis (Order Trichoptera). Bulletin of the Amateur Entomologist Society, 62, 15-26.
- Wiggins, G. B., 1996. Larvae of the North American Caddisfly Genera (Trichoptera). University of Toronto Press, Toronto, 457 pp.

Wiggins, G. B., 2004. Caddisflies: the underwater architects. University of Toronto Press, Toronto, 292 pp.


# Effects of Some Plant Extracts on Multi-Drug Resistant Acinetobacter baumannii Isolates

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Abstract: Multi-drug resistance (MDR) A. baumannii is among the most difficult antimicrobial resistant bacteria for control and treatment. The emergence of multidrug resistant isolates has limited the treatment methods in the clinic. Many new methods such as investigating the effect of natural products obtained from plants to combat antibiotic resistance are being improved. In particular, recent studies have focused on investigating the effects of plant extracts on resistant bacteria. Therefore, the aim of this study is to investigate the minimum inhibition concentrations of Zingiber officinale, Vaccinium myrtillus, Hypericum perforatum, Lycium barbarum, Aquilaria agallocha, Nigella sativa extracts aganist to multi-drug resistant A. baumannii isolates. Methanol was used as a solvent in the preparation of plant extracts. 6 different strains were selected according to the resistance genes they carried among MDR isolates. (AB38, AB43, AB45, AB46, AB59, AB69). MIC values of plant extracts were determined by liquid microdilution method. Experiments were performed in triplicate using 96 well-plate. As a result of the study; Aquilaria agallocha and Vaccinium myrtillus extracts did not inhibit growth of any strains in tested concentrations. The MIC values of the Nigella sativa extract against all strains were determined as 20 mg/ml. Lycium barbarum extract has a MIC of 35 mg/ml against AB38, AB43, AB46, AB59 strains. Also, MIC values of this exract aganist to AB45 and AB69 were found to be 17.5 mg / ml. MIC value of Hypericum perforatum extract was 10 mg / ml against AB38, AB43, AB45 isolates, but it did not inhibit growth of other strains in tested concentrations. MIC values of Zingiber officinale extract were determined as 30 mg/ml against AB38 and AB45 strains, and 15 mg/ml against AB46 and AB59 strains. In this study, the antimicrobial activity of the methanol extract of selected plants on MDR A. baumannii isolates were investigated. In the following studies, it is planned to investigate the antimicrobial activity of the same plants extracted in different solvents. In order to combat antibiotic resistance, the screening of antimicrobial activity of plants are of great importance.

Keywords: A.baumannii, MDR, MIC. plant extracts, resistance.



# Effect of *Teucrium orientale L*. Extract on the Mesenchymal Stem Cell Proliferation

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**Abstract:** *Teucrium* family members have many biological activities such as anti-oxidant, inflammatory, antiproliferative, antipyretic, etc. however, no evidence about their regeneration capability of stem cells. Our previous researches, which made first in the literature was showed that its anti-cancer activity on the HCT116 and Hep3B carcinoma cell lines. *Teacrium orintale* L. that is member of *Teacrium* family also used in the treatment of haemorrhoids between local people in the Samsun province in Turkey. Therefore, it seems that this plant has a therapeutic effect on the gastrointestinal system and tissue regeneration. So that, the aim of this study was evaluation of the proliferative effect of the T.orientale L. on rat mesenchymal stem cells (MSC) in relation to therapeutic features on the haemorrhoids.

Keywords: T.orientale L., MSC, Soxhlet extraction, HCT116, haemorrhoids.

#### 1. INTRODUCTION

*Teucrium orientale* L. (TOL) is a perennial herbaceous plant belongs to the family Lamiaceae. *Teucrium* species have many biological activities such as anti-oxidant, inflammatory, anti-proliferative, antipyretic, etc. (Bağcı et al., 2010). *Teucrium orientale* L. plant belonging to this family is used as a haemorrhoids healing agent in Samsun province today (Emin et. al., 2018a). Haemorrhoid is a degenerative disease which is today developing treatments (Alabaz,2007; Cirocco, 2007). Medicinal herbs from past to present are used today in the treatment of many diseases. When the traditional use of these medicinal plants is investigated, it is seen that it is mostly used in the treatment of haemorrhoid disease (Çalışkan et al., 2017; Tetik et al., 2013; Gürhan and Ezer, 2004; Altundağ and Öztürk, 2011).

*T. orientale* L. is known as "Kirve Otu" in Anatolia and this plant is usually growing on meadows and rocky slopes (Çakır et al., 2006). As a result of the research done *T. orientale* L. plant is known to preparation tea from flowers and participate in the food as spices in Anatolia (Korkmaz and Alpaslan, 2014). Also Cakir et al in their study, *T. orientale* L. plant antioxidant activity of the plant is evaluated and isolated from the acetone extract of Luteolin7-O-glucoside and Luteolin7-O-rutinosid compounds have a strong antioxidant and DPPH radical cleaning activity (Çakır et al., 2006).

Also because of our studies, *T. orientale* L. extract showed 75% success in the treatment of hemorrhoids. However, it is known that many mixtures currently used have limited activity to improve haemorrhoid. Nevertheless, this extract obtained from the plant *T. orientale* L. seems to have a high success rate alone to improve haemorrhoids. In addition, the recovery status of patients using this plant extract varies depending on factors such as the duration of use of the extract, regular use, age and the degree of the disease. However, it can be seen that the sagging of the skin, which is one of the symptoms of haemorrhoids, improves more easily in young individuals. This can be explained by the fact that tissue regeneration is much faster at younger ages (Emin et al., 2018a).

Our previous researches, which made first in the literature was showed that its anti-cancer activity on the HCT116 and Hep3B carcinoma cell lines (Emin et al., 2018b; Özerkan et al., 2018). It is thought that plant extract may have beneficial effects on stem cell regeneration because of both the strong antioxidant properties of *T. orientale* L. plant and the improvement of tissue on the haemorrhoids. Therefore, goal of the study was investigated of the proliferative effect of the *T.orientale* L. on rat mesenchymal stem cells (MSC) in relation to therapeutic features on the haemorrhoids and carcinoma cells.

#### 2. MATERIALS AND METHODS

#### **Preparation of Extract**

*Teucrium orientale L.* samples were collected after flowering and left to dry in a cool and aired place without contact with the sun. The dried plant parts were made into small pieces by dissection. Plant was extracted by using Soxhlet method in the distillate water. Stock solution of the extract was diluted different concentrations for investigation of proliferative concentration (PC) values.

#### **Cell Isolation and Culture**

MSCs were isolated from the femur bone marrow of young Wistar rats according the ethical rules (permission of the KUHADYEK: 19.02.2018/7654). The MSCs obtained were seeded in T75-culture dishes and culturing with the  $\alpha$ -MEM medium consist of Fetal bovine serum (FBS) and penicilin-streptomycin. Then they were placed in the incubator at 37°C, 5% CO<sub>2</sub> and the medium was changed every 3-4 days. MSCs obtained by trypsinization during sub-culturing were seeded in 96-multi well plates as 10<sup>4</sup> cells/well concentration. The plant extract diluted at different concentrations was added into each well according the tabel-1 for the investigation of proliferative concentration (PC) values.

Table 1. Extract concentrations were used for the analysis.

| Sample                | 1   | 2   | 3   | 4    | 5    | 6    | 7    | 8    | control |
|-----------------------|-----|-----|-----|------|------|------|------|------|---------|
| Concentration (µg/ml) | 2.5 | 5.0 | 7.5 | 10.0 | 12.5 | 15.0 | 17.5 | 20.0 | 0.0     |

PC values were calculated from dose-response curve. As a cross control, HCT116 cells were also tested with the plant extract for determination of the optimum PC values. MTT colorimetric-assay test kit based on cell-viability was used to observe cytotoxic/proliferative effect of the plant extract. Cell morphologies were also evaluated by inverted microscope.

#### MTT Cell Viability Analysis

In order to evaluate cell viability, MTT assay was performed for all groups on days 3 and 7 after cell transplantation. Briefly, culture medium was removed and cells were incubated with MTT reactive for 4 hours at 37 °C. At the end of the period, formazan crystals were solved with 1% HCl in 2-propanol solution (MTT solvent), and then absorbance values were read using by microplate reader at a wavelength of 570 nm.

#### 3. RESULTS AND DISCUSSION

As a result of MTT analysis, 7.5  $\mu$ g/ml concentration was found the most effective on both cancer cell death and MSC proliferation. Cell viability of HCT116 was observed 41% on 3<sup>rd</sup> day and 20% on 7<sup>th</sup> day.





It was determined that MSC numbers increased with using plant extract, and most proliferative dose response is 17.5  $\mu$ g/ml concentration. Comparison with the control group, MSC showed 156% proliferation activity at end of the 7<sup>th</sup> day. According the cross control experiments, optimum *T. orientale L.* concentration for MSC proliferation were 7.5  $\mu$ g/ml concentration that increase the proliferation 125% on 3<sup>rd</sup> day and 153% on 7<sup>th</sup> day.



Figure 2. Cell viability and proliferation rate of MSC measured by MTT analysis.

According to inverted microscopic images, HCT116 cells treated with extracts had higher rates of cancer cell death compared to negative control. Opposite of that, there was no significant differences in the cell morphology between the MSC that was treated and untreated with the extract. At the same time, it was found that MSCs treated with extract had higher proliferation than the untreated cells.

#### 4. CONCLUSION

The highest proliferative concentration for the MSC was found to be  $17.5 \,\mu$ g/ml. However, considering the most effective dose of T.orientale L. for the rate of killing unhealthy cells at the lowest concentration, the PC value for MSC was found to be 7.5  $\mu$ g/ml. It was determined that T.orientale L. promotes the proliferation of healthy cells simultaneously while killing unhealthy (carcinogenic) cells. Therefore, it is thought that T.orientale L. can be used in regenerative treatment.

As a conclusion, T.orientale L. extract actives MSC proliferation without affecting cell structure. Despite all this, we can suggest that T.orientale L. may be a clinically benefit, because of it has no toxic effect on healthy MSC cells, contrary to that exhibited cytotoxic effect on colorectal cancer cells.

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#### REFERENCES

- Alabaz, Ö., 2007. Etiyoloji ve Fizyopatoloji. In: (Baykan A., Füzün M., Zorluoğlu A.). Hemoroit Hastalığı ve Tedavisi. İstanbul: Avrupa Tıp Kitapçılık. 1: 27-29. ISBN: 9978-9756257-27-2.
- Altundağ, E., Öztürk, M., 2011. Ethnomedicinal Studies on the Plant Resources of East Anatolia, Turkey. Procedia Social and Behavioral Sciences. 19: 754-756. DOI: 10.1016/j.sbspro.2011.05.195.
- Bağcı, E., Yazgın, A., Hayta, Ş., Çakılcıoğlu, U., 2010. Composition of the Essential Oil of Teucrium Chamaedrys L. (Lamiaceae) from Turkey. Journal of Medicinal Plants Research. 4(23): 2587-2588. DOI: 10.5897/ JMPR10.823. ISSN: 1996-0875.
- Çakır, A., Mavi, A., Kazaz, C., Yıldırım, A., Küfrevioğlu, O. İ., 2006. Antioxidant Activities of the Extracts and Components of Teucrium orientale L. var orientale. Turkish Journal of Chemistry. 30: 483-494.
- Çalışkan, U. K., Aka, C., Öz M. G., 2017. Plants Used in Anatolian Traditional Medicine for the Treatment of Hemorrhoid. Records of Natural Products. 11(3): 238-247. ISSN: 130761-67.
- Cirocco, W. C., 2007. Why Are Hemorrhoids Symptomatic? The Pathophysiology and Etiology of Hemorrhoids. Seminars in Colon and Rektal Surgery. 18: 152-159. DOI: 10.1053/j.scrs.2007.07.004.
- Emin, N., Nural, K., Güzel, A., 2018a. Investigation of The Effectiveness of Teucrium Orientale L. Plant in Hemorrhoid Treatment. Turkish Journal of Agriculture - Food Science and Technology. 6(3):326-332.
- Emin, N., Özerkan, D., Nural ,K., Kuruca, D. S., 2018b. Comparative Evaluation of the Effect of Teucrium Orientale L. Extract on the Cell Proliferation by Using HCT116 and HUVEC Cells. In: International Congress On Engineering And Life Science, 26-29 April, Kastamonu.
- Gürhan, G., Ezer, N., 2004. Halk Arasında Hemoroit Tedavisinde Kullanılan Bitkiler-I. Hacettepe Üniversitesi Eczacılık Fakültesi Dergisi. 24(1): 41-45.

- Korkmaz, M., Alpaslan, Z., 2014. Ergan Dağı (Erzincan-Türkiye)' nın Etnobotanik Özellikleri. Bağbahçe Bilim Dergisi. 1(3): 14. Available from: http://edergi.ngbb.org.tr/ (03.02.2015). E-ISSN:2148-4015.
- Özerkan, D., Emin, N., Nural ,K., Kuruca, D. S., 2018. Hepatotoxic Effect of Teucrium Orientale L. on Hep3B Cells. In: II. International Eurasian Agricultural and Natural Sciences Congress, 11-15 September, Bakü, Azerbaycan.
- Tetik, F., Civelek, Ş., Çakılcıoğlu, U., 2013. Traditional Uses of Some Medicinal Plants in Malatya (Turkey). Journal of Ethnopharmacology. 146: 335-339.



# Is Fructose Harmful?

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**Abstract:** In order to identify the effects of especially the starch based sugar and all other recently introduced partially modified nutrients on the human health, it is very important to compile and assess up to date information based on controlled experiments.

There is a bidirectional communication between the gastrointestinal system (GIS) and brain, and this communication effectively regulates the appetite and the metabolism.

This communication is secreted through vagus nerve, spinal afferent neurons, cytokines, GIS hormones and signal molecules released from the microbiata. These signal molecules are orexigenic and anorexigenic. The variety of these signals combined with different type and amount of food combinations result in quite complicated interactions.

The arcuat nucleus of the hypotalamus is the center that regulates the energy metabolism. The increase of the AMP level in the arcuat nucleus activates the AMP-dependent kinases which in turn activates/inactivates the key enzymes which play the most critial roles in the regulation of metabolism.

With the diets prepared using the data based on information of how the nutrients are individually metabolized in the human body, the risks of obesity, dyslipidemia, diabetes and related diseases will be vastly reduced.

The risks of obesity, dyslipidemia, diabetes and related diseases could be vastly reduced if the diets are prepared using the current knowledge of how the nutrients are individually metabolized in the human body.

Keywords: Carbohydrate, metabolism, signaling, nutrition, obesity.



# Extracts of *Rhus coriaria L.* and *Pistacia terebinthus L.* grown in Turkey: Determination of Antimicrobial Activity and Chemical Contents

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Abstract: In this study, the antimicrobial activities of the ethanolic extracts of *Rhus coriaria* L. (Sumac/Sumak) and *Pistacia terebinthus* L. (Turpentine tree/Menengiç), used in the folk-medicine, were tested against gram-negative and gram positive bacteria as well as yeast-like fungi by the agar diffusion method. 19 microorganisms namely, Enterobacter aerogenes ATCC 13048, Salmonella infantis, Listeria monocytogenes ATCC 7644, Klebsiella pneumoniae, Pseudomonas aeruginosa DSMZ 50071, Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis ATCC 29212, Listeria innocua, Salmonella enteritidis ATCC 13075, Enterococcus durans, Salmonella typhimurium SL 1344, Candida ablicans ATCC 10231, Enterococcus faecium ATCC 25923, Staphylococcus aureus, Staphylococcus epidermidis DSMZ 20044, Bacillus subtilis DSMZ 1971, Escherichia coli ATCC 25922 and Serratia marcescens were used. Gas-Chromatography Mass Specrometry (GC-MS) analysis of their ethanol extracts were carried out in Shimadzu GCMS-QP-2010 system. The extracts of sumac and turpentine tree showed antibacterial activity against tested microorganisms at different levels.

Keywords: Rhus coriaria, Sumac, Pistacia terebinthus, Turpentine tree, Disc Diffusion, GC-MS.

#### **1. INTRODUCTION**

Medical Aromatic Plants (MAPs) are important in the economic, cultural and social aspects of the communities around the world. MAPs can be defined as plants that provide drugs to protect human health, prevent and treat diseases. MAP can grow in some aquatic ecosystems and continental climates in the world. The characteristics and uses of different sources in the world include a wide variety of species that are part of traditional medical systems in local communities. Health and wealth from Medicinal Aromatic Plants, Elaine Marshall, Rural Infrastructure and Agro-Industries Division Food and Agriculture Organization of the United Nations Rome 2011.

The Mediterranean diet is especially ample in spices. *Rhus coriaria* L. known as Sumac is one sample, which is largely used in Turkey and the Middle East. The fruits are red colored and include one seed. Its Dried and ground leaves contain high tannin content, used as tanning agent. Previous phytochemical studies of this plant have been reported to contain flavon, tannin, anthocyanin and organic acid. After all, it is typically the fruit of the plant consumed as a spice after drying and milling. It is utilized for treatment of dyspepsia, anorexia, catharsis, hemorrhagia and hyperglycemia in traditional medicine (Wetherilt and Pala, 1994).

*P. terebinthus* is a non-evergreen tree and is member of Anacardiaceae family. In the Middle East and Mediterranean territory, it is a small tree growing up to 10 m. The extracts of this plant have anti-inflammation and antioxidant flavonoid, omega-3 fatty acids, tannin and other beneficial compounds. Being a folk medicine, it is using for the remedy of liver and kidney and also drinks coffee. It can be used in the production of vegetable oils because of its high amount of oleic acid. Different parts of the *P. terebinthus* tree have been reported to possess many biological activities as they have been widely used in phytopharmacy from ancient times (Orhan et all., 2012)

This paper is concerned with antimicrobial activity of ethanol (65%) extracts of *Rhus coriaria* L. and *Pistacia terebinthus* L. against 19 microorganisms by using the disk diffusion method, and volatile components of the Sumac and Turpentine tree were determined by GC-MS analysis (gas chromatography-mass spectroscopy).

#### 2. MATERIALS AND METHODS

#### **Plant Samples**

*Rhus coriaria* L. and *Pistacia terebinthus* L. were obtained from Pinarbaşi/Kastamonu/ Turkey, in July at 2018. Plant materials were dried at room temperature and on draft for three weeks. All the plants was identified by Assist. Prof. Dr. Kerim GÜNEY, Department of Forest Engineering, Faculty of Forestry, Kastamonu University.

#### **Extraction method**

The plants were washed thoroughly 2-3 times with water, and then they were air dried under shade. Afterwards, the dried plant materials were ground in a mixer, the powder was kept in the amber glass bottle. About 10-30 g ground plant samples were extracted with 250 mL of ethanol (%65) in a Soxhlet apparatus by continuous heat extraction for 24 hours. All extract solutions were filtered through Whatman No.1 paper. Then, filtrates were evaporated with a rotary evaporator. The filtrates were freeze dried and stored in refrigerator at about 4°C after sealed with paraffin for further studies.

#### **Determination of Antimicrobial Activities**

#### **Preparation of Extract Stock**

Extract stocks to test the antimicrobial activity were prepared by dissolving 1 mg of extract in each 3 mL of ethanol for disk diffusion test.

#### Strains

In order to analyse the antimicrobial activity of plants extracts, 19 microorganisms namely, *Enterobacter aerogenes* ATCC 13048, *Salmonella infantis*, *Listeria monocytogenes* ATCC 7644, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* DSMZ 50071, *Pseudomonas fluorescens*, *Salmonella kentucky*, *Enterococcus faecalis* ATCC 29212, *Listeria innocua*, *Salmonella enteritidis* ATCC 13075, *Enterococcus durans*, *Salmonella typhimurium* SL 1344, *Candida ablicans* ATCC 10231, *Enterococcus faecium* ATCC 25923, *Staphylococcus aureus*, *Staphylococcus epidermidis* DSMZ 20044, *Bacillus subtilis* DSMZ 1971, *Escherichia coli* ATCC 25922 and *Serratia marcescens* were used.

#### **Preparation of Innocula**

All strains were incubated according to their requirements as it was previously mentioned by Altuner and Canlı (2012) and Canlı et al (2015).

For the inocula, morphologically similar colonies of each organism were transferred into physiological saline (Canlı et al, 2016a and b, Onbasli, 2013) and to adjust equal the number of the colonies in the solution, 0.5 McFarland standard was used (Hammer et al., 1999; Altuner et al., 2012a and b). Thus standard inocula is adjusted to contain approximately 108 cfu/mL for bacteria and 107 cfu/mL for C. albicans (Canlı et al, 2016c and d).

#### **Disk Diffusion Test**

The disk diffusion test was applied as it was mentioned in the previous studies (Andrews, 2003; Canlı et al, 2014). Petri dishes containing Mueller Hinton Agar were used for disk diffusion test (Ilhan et al., 2006; Canlı et al, 2016e). 20, 40 and 80  $\mu$ L of extracts were loaded on empty sterile antibiotic disks (SAD). Disks were kept at 40°C for 24 h in aseptic conditions (Altuner et al., 2010). Microorganism suspensions were inoculated on the surfaces of the Mueller Hinton plates and left in aseptic conditions for 2-3 minutes before applying disks as described in the previous studies (Altuner and Akata, 2010). Inhibition zones were defined in mm by the method mentioned by Altuner et al. (2014).

#### Controls

Empty SAD was used as negative controls for disk diffusion test, where broth medium inoculated with each microorganism was used to control microorganisms.

#### GC-MS (Gas chromatography-mass spectroscopy) Analysis

GC-MS analysis was performed using Shimadzu GCMS QP 2010 ULTRA series device. Samples were passed through RTX-5MS capillary column (30 m x 0.25 mm x 0.25  $\mu$ m). Helium gas was used as carrier gas. The injection temperature was maintained at 250° C. GC-MS analysis was performed according to the procedure in the literature. The oven is heated to 40° C and is allowed to stand at this temperature for 3 minutes. Then the temperature is increased by 4 degrees per minute to 240° C and waited for 10 minutes. Finally, the temperature is increased by 4 degrees per minute to 260° C and kept for 65 minutes and is completed to a total of 78 minutes. The injection temperature was maintained at 250° C and

the injection volume was determined as 1  $\mu$ l. The intermittent temperature is 250° C and the ion boiling temperature is 200°C. It was treated with hexane.

#### 3. RESULTS AND DISCUSSION

The GC-MS results of the extracts are displayed in Table 1, and the antimicrobial activity test results are displayed in Table 2.

GC- MS analysis was carried out ethanol (%65) extracts of *Rhus coriaria L*. and *Pistacia terebinthus L*. The GC/MS analysis of the ethanol (%65) extracts of *Rhus coriaria L*. displayed that there were 19 compounds with 9-Octadecenoic Acid, Methyl Ester, (E)- (11.99%), Phenol, 3-Pentadecyl- (7.90%), 9,12-Octadecadienoic Acid (Z,Z)-, Methyl Ester (7.47%) and Phytol, Acetate (7.68%) as the most plenty ones.

The GC/MS analysis of the ethanol (%65) extracts of *Pistacia terebinthus L*. demonstrated that there were 16 compounds with Methyl elaidate (20.23%), Methyl Palmitate (13.05%) and Lupan-3-ol, acetate (12.91%) as the most plenty ones (Table 1).

|      | Rhus coriari | a L.  |                              | Pistacia tere | binthus L. |  |
|------|--------------|-------|------------------------------|---------------|------------|--|
| Peak | R. Time      | Area% | Compenent Name               | R. Time       | Area%      | Compenent Name   |
| 1    | 3.708        | 2.14  | 2,2-<br>dimethoxybutane      | 41.704        | 13.05      | Methyl Palmitate   |
| 2    | 4.307        | 1.35  | Ethyl n-propyl<br>ketone     | 43.406        | 1.19       | Ethyl palmitate  |
| 3    | 4.415        | 1.77  | 2-Hexanone (CAS)             | 45.840        | 8.15       | Methyl linoleate   |
| 4    | 41.726       | 6.62  | Methyl Palmitate             | 45.978        | 20.23      | Methyl elaidate  |
| 5    | 41.991       | 2.02  | Cembrene                     | 46.108        | 2.19       | 11-Octadecenoic<br>acid, methyl ester                            |
| 6    | 45.854       | 7.47  | Methyl Linoleate             | 46.585        | 1.22       | Methyl stearate  |
| 7    | 45.989       | 11.99 | Methyl elaidate              | 47.399        | 1.15       | Linoleic acid ethyl ester  |
| 8    | 46.105       | 1.15  |                              | 47.525        | 2.00       | Ethyl Oleate   |
| 9    | 46.600       | 1.96  | Methyl stearate              | 50.421        | 2.82       | Hexadecanoic acid,<br>1-(hydroxymethyl)-<br>1,2-ethanediyl ester |
| 10   | 47.410       | 1.75  | Linoleic acid ethyl<br>ester | 54.158        | 1.54       | Linoleoyl chloride   |
| 11   | 50.433       | 1.09  | Glyceryl 1,3-<br>Distearate  | 54.252        | 5.38       | Dı-(9-<br>Octadecenoyl)-<br>Glycerol                             |
| 12   | 54.289       | 3.73  | 13-Octadecenal,<br>(Z)-      | 54.375        | 1.57       | -  |
| 13   | 54.350       | 2.61  |                              | 54.495        | 3.00       | Cyclohexanecarbox<br>ylic acid,<br>heptadecyl ester              |
| 14   | 54.580       | 6.18  | Phenol, 3-<br>pentadecyl-    | 54.700        | 1.26       | Oxalic acid, 3,5-<br>difluorophenyl<br>tetradecyl ester          |
| 15   | 54.763       | 7.90  | Phenol, 3-<br>pentadecyl-    | 65.861        | 2.83       | Squalene   |
| 16   | 59.275       | 6.27  | Stearyl Benzoate             | 68.510        | 12.91      | Lupan-3-ol, acetate  |
| 17   | 70.595       | 1.40  |                              | 76.715        | 1.12       |  |
| 18   | 72.220       | 1.64  |                              | 76.840        | 1.00       |  |
| 19   | 73.582       | 7.68  | Phytol, acetate              | 77.416        | 2.84       |  |
| 20   | 75.373       | 2.51  | Betulin                      |               |            |  |

Table 1. Rhus coriaria L. and Pistacia terebinthus L. GC-MS aroma analysis results

Disk diffusion test results showed that *Rhus coriaria L.* is active against *P. fluorescens, S. kentucky, E. faecalis, L. innocua, S. enteritidis, E. durans, C. ablicans, S. ayreus, S. epidermidis, B. subtilis, E. coli and S. marcescens with inhibition zones between 7-13 mm, where Pistacia terebinthus L. is active against K. pneumoniae, P. fluorescens, E. faecalis, S. enteritidis, C. ablicans and E. faecium with inhibition zones between 7-9 mm.* 

|                  | Rhus coriaria L. |         | Pistacia terebinthu |       | thus L. |       |
|------------------|------------------|---------|---------------------|-------|---------|-------|
|                  | 20               | 40      | 80                  | 20 µL | 40      | 80 µL |
| E. aerogenes     | μL<br>-          | μL<br>- | μL<br>-             | _     | μL<br>- | _     |
| S. infantis      |                  | -       |                     | -     | -       | -     |
| L. monocytogenes | -                | -       | -                   | -     | -       | -     |
| K. pneumoniae    | -                | -       | -                   | 8     | 8       | 8     |
| P. aeruginosa    | -                | -       | -                   | -     | -       | -     |
| P. fluorescens   | -                | 7       | 8                   | -     | -       | 7     |
| S. kentucky      | -                | 9       | 10                  | -     | -       | -     |
| E. faecalis      | -                | 8       | 8                   | -     | -       | 9     |
| L. innocua       | -                | -       | 9                   | -     | -       | -     |
| S. enteritidis   | -                | -       | 8                   | -     | 7       | 7     |
| E. durans        | 9                | 9       | 10                  | -     | -       | -     |
| S. typhimurium   | -                | -       | -                   | -     | -       | -     |
| C. ablicans      | 9                | 11      | 13                  | -     | 8       | 9     |
| E. faecium       | -                | -       | -                   | 7     | 7       | 7     |
| S. aureus        | 8                | 10      | 13                  | -     | -       | -     |
| S. epidermidis   | -                | 7       | 8                   | -     | -       | -     |
| B. subtilis      | -                | 7       | 8                   | -     | -       | -     |
| E. coli          | 7                | 7       | 8                   | -     | -       | -     |
| S. marcescens    | -                | 8       | 9                   | -     | -       | -     |
|                  |                  |         |                     |       |         |       |

Table 2. Disk Diffusion Test Results of extracts at the Rhus coriaria L. and Pistacia terebinthus L. concentration

#### 4. CONCLUSION

In this study, the antimicrobial activity of the extract of *Rhus coriaria* L. and *Pistacia terebinthus* L.was determined. The extracts showed activity against almost 14 strains were resistant among the Gram-negative bacterial and fungi strains and tested. However, the phytochemical component of ethanol extract of *Rhus coriaria* L. and *Pistacia terebinthus* L. exposed by GC-MS analysis shows its cost in phytopharmaceuticals, cosmetic and food industries.

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#### REFERENCES

- Altuner, E.M. 2011. Investigation of antimicrobial activity of Punica granatum L. fruit peel ash used for protective against skin infections as folk remedies especially after male circumcision. African Journal of Microbiology Research. 5(20):3339-3342.
- Altuner, E.M. and I. Akata. 2010. Antimicrobial activity of some macrofungi extracts. Sakarya Üniversitesi Fen Bilimleri Enstitüsü Dergisi. 14(1):45-49.
- Altuner, E.M. and K. Canlı. 2012. In vitro antimicrobial screening of Hypnum andoi A.J.E. Sm. Kastamonu U. J. For. Fac. 12:97-101.
- Altuner, E.M., I. Akata and K. Canlı. 2012a. In vitro antimicrobial screening of Bovista nigrescens (Pers.). Kastamonu U. J. For. Fac. 12:90-96.
- Altuner, E.M., I. Akata and K. Canlı. 2012b. In vitro antimicrobial screening of Cerena unicolor (Bull.) Murrill (Polyporaceae Fr. Ex Corda). Fresen. Environ. Bullet. 21:3704-3710.
- Altuner, E.M., K. Canlı and I. Akata. 2014. Antimicrobial screening of Calliergonella cuspidata, Dicranum polysetum and Hypnum cupressiforme. Journal of Pure and Applied Microbiology. 8(1):539-545.

- Andrews, J.M. 2003. BSAC standardized disc susceptibility testing method (version 6). Journal of Antimicrobial Chemotherapy. 60:20-41.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016d. In vitro antimicrobial screening of Aquilaria agallocha roots. African Journal of Traditional, Complementary and Alternative Medicines. 13(5):178-181.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016e. In vitro antimicrobial activity screening of Rheum rhabarbarum roots. International Journal of Pharmaceutical Sciences Invention, 5(2):1-4.
- Canlı, K., B. Çetin, E.M. Altuner, Y. Türkmen, U. Üzek and H. Dursun. 2014. In vitro antimicrobial screening of Hedwigia ciliata var. leucophaea and determination of the ethanol extract composition by gas chromatography/mass spectrometry (GC/MS). Journal of Pure and Applied Microbiology. 8(4):2987-2998.
- Canlı, K., E.M. Altuner and I. Akata. 2015. Antimicrobial screening of Mnium stellare. Bangladesh Journal of Pharmacology. 10:321-325.
- Canlı, K., E.M. Altuner, I. Akata, Y. Türkmen and U. Üzek. 2016a. In vitro antimicrobial screening of Lycoperdon lividium and determination of the ethanol extract composition by gas chromatography/mass spectrometry. Bangladesh Journal of Pharmacology. 11(2):389-394.
- Canlı, K., I. Akata and E.M. Altuner. 2016c. In vitro antimicrobial activity screening of Xylaria hypoxylon. African Journal of Traditional, Complementary and Alternative Medicines. 13(4):42-46.
- Hammer, K.A., C.F. Carson and T.V. Riley. 1999. Antimicrobial activity of essential oils and other plant extracts. Journal of Applied Microbiology. 86:985-990.
- Ilhan, S., F. Savaroğlu, F. Çolak, C.F. Iscen and F.Z. Erdemgil. 2006. Antimicrobial activity of Palustriella commutata (Hedw.) Ochyra extracts (Bryophyta). Turk. J. Biol, 30:149-152.
- Kartal, M. (2004). Avrupa birliği ülkelerinde tıbbi bitkisel ürünlerin ruhsatlandırılması, Ankara Üniversitesi Eczacılık Fakültesi Farmakognozi Anabilim Dalı.
- Nychas, G. J. E. (1995). Natural antimicrobials from plants. In: Gould, G. W. (Ed.), New Methods of Food Preservation. (58pp) London, Blackie: Academic Profesional.
- Onbasli, D., G. Yuvali Celik, E.M. Altuner, B. Altinsoy and B. Aslim. 2013. In vitro antimicrobial, antioxidant, and antibiofilm activities of Bryum capillare, a bryophyte sample. Current Opinion in Biotechnology, 24 (Supplement 1): 113.
- Orhan, I.E.; Senol, F.S.; Gulpinar, A.R.; Sekeroglu, N.; Kartal, M.; Sener, B. Neuroprotective potential of some terebinth co ee brands and the unprocessed fruits of *Pistacia terebinthus* L. and their fatty and essential oil analyses. Food Chem. 2012, 130, 882– 888.
- Özcan, M. and Sağdıç, O. (2003). Antibacterial activity of Turkish spice hydrosols. Food Control.
- Rabe, T. and Van Staden, J. (1997). Antibacterial activity of South African plants used for medicinal purposes. Journal of Ethnopharmacology.
- Wetherilt, H., Pala, M., 1994. Herbs and spices indigenous to Turkey. In: Charalambous, G. (Ed.), Spices, Herbs and Edible Fungi. Developments in Food Science, vol. 34. Elsevier, Amsterdam, pp. 285 – 307.



# Extracts of *Hibiscus rosa-sinensis L*. and *Paeonia peregrina Mill*. in Turkey: Antimicrobial Activity and GC-MS Analysis

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**Abstract:** In this study, the antimicrobial activities of the ethanolic extracts of *Hibiscus rosa-sinensis L*. (Chinese hibiscus) and *Paeonia peregrina Mill*. (Balkan peony), used in the folk-medicine, were tested against gram-negative and gram positive bacteria as well as yeast-like fungi by the agar diffusion method. 19 microorganisms namely, *Enterobacter aerogenes* ATCC 13048, *Salmonella infantis, Listeria monocytogenes* ATCC 7644, *Klebsiella pneumoniae, Pseudomonas aeruginosa* DSMZ 50071, *Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis* ATCC 29212, *Listeria innocua, Salmonella enteritidis* ATCC 13075, *Enterococcus durans, Salmonella typhimurium* SL 1344, *Candida ablicans* ATCC 10231, *Enterococcus faecium* ATCC 25923, *Staphylococcus aureus, Staphylococcus epidermidis* DSMZ 20044, *Bacillus subtilis* DSMZ 1971, *Escherichia coli* ATCC 25922 and *Serratia marcescens* were used. Gas-Chromatography Mass Specrometry (GC-MS) analysis of their ethanol extracts were carried out in Shimadzu GCMS-QP-2010 system. The extracts of Chinese hibiscus and Balkan peony showed antibacterial activity against tested microorganisms at different levels.

Keywords: Hibiscus rosa-sinensis L., Paeonia peregrina Mill. Disc Diffusion, GC-MS.

#### **1. INTRODUCTION**

Plants have been used in the treatment of many diseases from prehistoric to today. In addition, people from all cultures use this tradition (Karou at all., 2005). However, it has been studied on the pharmaceutical composition of these products such as antioxidant, antimicrobial and anticancer effect. In addition, the extract compositions vary depending on the type of solvent and their different effects.

The malvaceae family contains about 120 genera and 1700 to 2000 species (Nunes at all., 2006). The species of this family, especially *Hibiscus rosa-sinensis* has been used as herbal plants in folk medicine for treatment of different diseases. Moreover, it has been stated that Hibiscus rosa possesses anti-complementary, anti-diarrhetic and anti-phlogistic activity. Otherwise, *Hibiscus rosa-sinensis* flowers have anti-spermatogenic, androgenic, anti-tumor and anti-convulsant activities (Lans, 2007).

The genus Paeonia L. (Paeoniaceae) include several species. Among these, in Turkey 12 taxa of Paeonia were recorded (Kit Tan, 2000). Paeonia species and especially their roots have an important place in traditional Chinese medicine (Zhu, 1998). They have been said to own sedative, analgesic, and anti-inflammatory properties. They have been utilised as a cure for cardiovascular diseases, female genital diseases, and eczema (Prajapati at all., 2003). In traditional medicine, P. peregrina have been utilised against ulcers, coughs, and epilepsy in Anatolia (Baytop, 1999).

This paper is concerned with antimicrobial activity of ethanol (65%) extracts of *Hibiscus rosa-sinensis* L. and *Paeonia peregrina* Mill. against 19 microorganisms by using the disk diffusion method, and volatile components of the *Hibiscus rosa-sinensis* L. and *Paeonia peregrina* Mill. were determined by GC-MS analysis (Gas Chromatography-Mass Spectroscopy).

#### 2. MATERIALS AND METHODS

#### **Plant Samples**

*Hibiscus rosa-sinensis* L. was obtained from sea level Gözce village/Bozyazı/Mersin/Turkey, in July at 2018. *Paeonia peregrina* Mill. was obtained from 1000 m Pinarbaşı/Kastamonu/Turkey, in August at 2018. Plant materials were dried at room temperature and on draft for three weeks. All the plants was identified by Assist. Prof. Dr. Kerim GÜNEY, Department of Forest Engineering, Faculty of Forestry, Kastamonu University.

#### **Extraction method**

The plants were washed thoroughly 2-3 times with water, and then they were air dried under shade. Afterwards, the dried plant materials were ground in a mixer, the powder was kept in the amber glass bottle. About 10-30 g ground plant samples were extracted with 250 mL of ethanol (%65) in a Soxhlet apparatus by continuous heat extraction for 24 hours. All extract solutions were filtered through Whatman No.1 paper. Then, filtrates were evaporated with a rotary evaporator. The filtrates were freeze dried and stored in refrigerator at about 4°C after sealed with paraffin for further studies.

#### **Determination of Antimicrobial Activities**

#### **Preparation of Extract Stock**

Extract stocks to test the antimicrobial activity were prepared by dissolving 1 mg of extract in each 3 mL of ethanol for disk diffusion test.

#### Strains

In order to analyse the antimicrobial activity of plants extracts, 19 microorganisms namely, *Enterobacter aerogenes* ATCC 13048, *Salmonella infantis, Listeria monocytogenes* ATCC 7644, *Klebsiella pneumoniae, Pseudomonas aeruginosa* DSMZ 50071, Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis ATCC 29212, *Listeria innocua, Salmonella enteritidis* ATCC 13075, *Enterococcus durans, Salmonella typhimurium* SL 1344, *Candida ablicans* ATCC 10231, Enterococcus faecium ATCC 25923, *Staphylococcus aureus, Staphylococcus epidermidis* DSMZ 20044, *Bacillus subtilis* DSMZ 1971, *Escherichia coli* ATCC 25922 and *Serratia marcescens* were used.

#### **Preparation of Innocula**

All strains were incubated according to their requirements as it was previously mentioned by Altuner and Canlı (2012) and Canlı et al (2015).

For the inocula, morphologically similar colonies of each organism were transferred into physiological saline (Canlı et al, 2016a and b, Onbasli, 2013) and to adjust equal the number of the colonies in the solution, 0.5 McFarland standard was used (Hammer et al., 1999; Altuner et al., 2012a and b). Thus standard inocula is adjusted to contain approximately 108 cfu/mL for bacteria and 107 cfu/mL for C. albicans (Canlı et al, 2016c and d).

#### **Disk Diffusion Test**

The disk diffusion test was applied as it was mentioned in the previous studies (Andrews, 2003; Canlı et al, 2014). Petri dishes containing Mueller Hinton Agar were used for disk diffusion test (Ilhan et al., 2006; Canlı et al, 2016e). 20, 40 and 80  $\mu$ L of extracts were loaded on empty sterile antibiotic disks (SAD). Disks were kept at 40°C for 24 h in aseptic conditions (Altuner et al., 2010). Microorganism suspensions were inoculated on the surfaces of the Mueller Hinton plates and left in aseptic conditions for 2-3 minutes before applying disks as described in the previous studies (Altuner and Akata, 2010). Inhibition zones were defined in mm by the method mentioned by Altuner et al. (2014).

#### Controls

Empty SAD was used as negative controls for disk diffusion test, where broth medium inoculated with each microorganism was used to control microorganisms.

#### GC-MS (Gas chromatography-mass spectroscopy) Analysis

GC-MS analysis was performed using Shimadzu GCMS QP 2010 ULTRA series device. Samples were passed through RTX-5MS capillary column (30 m x 0.25 mm x 0.25 µm). Helium gas was used as carrier gas. The injection temperature was maintained at 250° C. GC-MS analysis was performed according to the procedure in the literature. The oven is heated to 40° C and is allowed to stand at this temperature for 3 minutes. Then the temperature is increased by 4 degrees per minute to 240° C and waited for 10 minutes. Finally, the temperature is increased by 4 degrees per minute to 260° C and

kept for 65 minutes and is completed to a total of 78 minutes. The injection temperature was maintained at 250° C and the injection volume was determined as 1  $\mu$ l. The intermittent temperature is 250° C and the ion boiling temperature is 200°C. It was treated with hexane.

#### 3. RESULTS AND DISCUSSION

The GC-MS results of the extracts are presented in Table 1, and the antimicrobial activity test results are presented in Table 2.

GC- MS analysis was made on ethanol (%65) extracts of *Hibiscus rosa-sinensis* L. and *Paeonia peregrina* Mill. 21 phytoconstituents were detected in *Paeonia peregrina* Mill. Besides 11 phytoconstituents were detected in *Hibiscus rosa-sinensis* L. The active principles with their retention time (RT), molecular formula, molecular weight and peak area (%) are presented in Table -1. The peaks in the chromatogram were associated and were hold a candle to the database of spectrum of known components stored in the Gas Chromatography-Mass Spectrometry library. GC-MS analysis of the ethanol extract of *Hibiscus rosa-sinensis* and *Paeonia peregrina* revealed the presence of different fatty acids, fatty acids methyl esters and some volatile organic compounds.

 Table 1. Hibiscus rosa-sinensis L. and Paeonia peregrina Mill. GC-MS aroma analysis results

|      |            | Hibiscus  | s rosa-sinensis L.   | Paeonia peregrina Mill. |           |  |  |
|------|------------|-----------|--|-------------------------|-----------|--|--|
| Peak | R.<br>Time | Area<br>% | Compenent Name   | R.<br>Time              | Area<br>% | Compenent Name   |  |
| 1    | 3.702      | 1.97      | 2,2-dimethoxybutane  | 3.704                   | 3.03      | 2,2-dimethoxybutane  |  |
| 2    | 9.984      | 1.43      | 4-oxo-5-methoxy-2-<br>penten-5-olide   | 13.850                  | 1.73      | Decane, 3,7-dimethyl-  |  |
| 3    | 22.511     | 32.2<br>0 | Anethole   | 22.243                  | 1.72      | Nonane, 5-butyl-   |  |
| 4    | 29.033     | 5.16      | cis-(-)-2,4a,5,6,9a-<br>Hexahydro-3,5,5,9-<br>tetramethyl(1H)benzocy<br>cloheptene | 23.865                  | 1.19      | Hexadecane   |  |
| 5    | 41.707     | 9.38      | Methyl Palmitate   | 29.475                  | 1.69      | Eicosane   |  |
| 6    | 45.846     | 13.0<br>3 | Methyl Linoleate   | 32.680                  | 2.14      | Diethyl Phthalate  |  |
| 7    | 45.999     | 10.1<br>7 | 9,12,15-Octadecatrienoic acid, methyl ester (CAS)                                  | 35.858                  | 1.54      | Eicosane   |  |
| 8    | 46.589     | 1.81      | Methyl stearate  | 39.109                  | 1.70      | Octadecamethylcyclonon<br>asiloxane                              |  |
| 9    | 47.522     | 2.32      | Ethyl Oleate   | 41.710                  | 7.26      | Methyl Palmitate   |  |
| 10   | 60.689     | 1.04      | Eicosane   | 42.659                  | 1.13      | Tetracosane  |  |
| 11   | 68.127     | 5.41      | Pentacosane  | 45.839                  | 6.61      | Methyl Linoleateester  |  |
| 12   |            |           |  | 45.974                  | 8.07      | Methyl elaidate  |  |
| 13   |            |           |  | 46.586                  | 1.34      | Methyl stearate  |  |
| 14   |            |           |  | 50.416                  | 5.45      | Glyceryl 1,3-Distearate  |  |
| 15   |            |           |  | 54.154                  | 2.85      | Linoleoyl chloride   |  |
| 16   |            |           |  | 54.246                  | 11.1<br>7 | Di-(9-Octadecenoyl)-<br>Glycerol                                 |  |
| 17   |            |           |  | 54.405                  | 2.56      |  |  |
| 18   |            |           |  | 54.483                  | 2.50      | Oleic acid, (2,2-<br>dimethyl-1,3-dioxolan-4-<br>yl)methyl ester |  |
| 19   |            |           |  | 56.288                  | 1.34      | 1H-Purin-6-amine, [(2-<br>fluorophenyl)methyl]-<br>(CAS)         |  |
| 20   |            |           |  | 65.852                  | 5.33      | Squalene   |  |
| 21   |            |           |  | 77.181                  | 1.32      | Vitamin E  |  |

Disk diffusion test results showed that *Hibiscus rosa-sinensis L*. is active against *S. infantis, K. pneumoniae, P. aeruginosa, P. fluorescens, S. kentucky and E. faecium* with inhibition zones between 7-10 mm, where *Paeonia peregrina Mill.* is active against *L. monocytogenes, K. pneumoniae, S. kentucky, E. durans, C. ablicans, E. faecium and S. aureus* with inhibition zones between 7-9 mm (Table 2).

|                  | Hib<br>si | Hibiscus rosa- |          |       | Paeonia peregrina<br>Mill. |          |  |  |
|------------------|-----------|----------------|----------|-------|----------------------------|----------|--|--|
|                  | 20<br>µL  | 40<br>μL       | 80<br>μL | 20 µL | 40<br>μL                   | 80<br>μL |  |  |
| E. aerogenes     | -         | -              | -        | -     | -                          | -        |  |  |
| S. infantis      | 7         | 8              | 10       | -     | -                          | -        |  |  |
| L. monocytogenes | -         | -              | -        | -     | -                          | 9        |  |  |
| K. pneumoniae    | 8         | 8              | 8        | 8     | 8                          | 8        |  |  |
| P. aeruginosa    | -         | -              | 8        | -     | -                          | -        |  |  |
| P. fluorescens   | -         | 7              | 7        | -     | -                          | -        |  |  |
| S. kentucky      | -         | 7              | 7        | -     | 8                          | 8        |  |  |
| E. faecalis      | -         | -              | -        | -     | -                          | -        |  |  |
| L. innocua       | -         | -              | -        | -     | -                          | -        |  |  |
| S. enteritidis   | -         | -              | -        | -     | -                          | -        |  |  |
| E. durans        | -         | -              | -        | -     | -                          | 7        |  |  |
| S. typhimurium   | -         | -              | -        | -     | -                          | -        |  |  |
| C. ablicans      | -         | -              | -        | -     | -                          | 9        |  |  |
| E. faecium       | -         | 7              | 7        | 7     | 7                          | 8        |  |  |
| S. aureus        | -         | -              | -        | -     | 7                          | 7        |  |  |
| S. epidermidis   | -         | -              | -        | -     | -                          | -        |  |  |
| B. subtilis      | -         | -              | -        | -     | -                          | -        |  |  |
| E. coli          | -         | -              | -        | -     | -                          | -        |  |  |
| S. marcescens    | -         | -              | -        | -     | -                          | -        |  |  |

# Table 2. Disk Diffusion Test Results of extracts at the Hibiscus rosa-sinensis L. and Paeonia peregrina Mill. concentration

#### 4. CONCLUSION

Plants are natural reservoirs of various phytonutrients and compounds which are urgent and essential to life in general. The phytochemical component of ethanol extract of *Hibiscus rosa-sinensis* L. and *Paeonia peregrine* Mill. revealed by GC-MS analysis shows its significance in phytopharmaceuticals, cosmetic and food industries.

On the basis of antimicrobial activity, it was monitored that the ethanol extracts were effective only for few of the tested species.

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#### REFERENCES

- Altuner, E.M. 2011. Investigation of antimicrobial activity of Punica granatum L. fruit peel ash used for protective against skin infections as folk remedies especially after male circumcision. African Journal of Microbiology Research. 5(20):3339-3342.
- Altuner, E.M. and I. Akata. 2010. Antimicrobial activity of some macrofungi extracts. Sakarya Üniversitesi Fen Bilimleri Enstitüsü Dergisi. 14(1):45-49.
- Altuner, E.M. and K. Canlı. 2012. In vitro antimicrobial screening of Hypnum andoi A.J.E. Sm. Kastamonu U. J. For. Fac. 12:97-101.
- Altuner, E.M., I. Akata and K. Canlı. 2012a. In vitro antimicrobial screening of Bovista nigrescens (Pers.). Kastamonu U. J. For. Fac. 12:90-96.
- Altuner, E.M., I. Akata and K. Canlı. 2012b. In vitro antimicrobial screening of Cerena unicolor (Bull.) Murrill (Polyporaceae Fr. Ex Corda). Fresen. Environ. Bullet. 21:3704-3710.
- Altuner, E.M., K. Canlı and I. Akata. 2014. Antimicrobial screening of Calliergonella cuspidata, Dicranum polysetum and Hypnum cupressiforme. Journal of Pure and Applied Microbiology. 8(1):539-545.

- Andrews, J.M. 2003. BSAC standardized disc susceptibility testing method (version 6). Journal of Antimicrobial Chemotherapy. 60:20-41.
- Baytop, T. Therapy with Plants in Turkey (Past and Present), Nobel Medical Bookhouse, Istanbul, 1999.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016d. In vitro antimicrobial screening of Aquilaria agallocha roots. African Journal of Traditional, Complementary and Alternative Medicines. 13(5):178-181.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016e. In vitro antimicrobial activity screening of Rheum rhabarbarum roots. International Journal of Pharmaceutical Sciences Invention, 5(2):1-4.
- Canlı, K., B. Çetin, E.M. Altuner, Y. Türkmen, U. Üzek and H. Dursun. 2014. In vitro antimicrobial screening of Hedwigia ciliata var. leucophaea and determination of the ethanol extract composition by gas chromatography/mass spectrometry (GC/MS). Journal of Pure and Applied Microbiology. 8(4):2987-2998.
- Canlı, K., E.M. Altuner and I. Akata. 2015. Antimicrobial screening of Mnium stellare. Bangladesh Journal of Pharmacology. 10:321-325.
- Canlı, K., E.M. Altuner, I. Akata, Y. Türkmen and U. Üzek. 2016a. In vitro antimicrobial screening of Lycoperdon lividium and determination of the ethanol extract composition by gas chromatography/mass spectrometry. Bangladesh Journal of Pharmacology. 11(2):389-394.
- Canlı, K., I. Akata and E.M. Altuner. 2016c. In vitro antimicrobial activity screening of Xylaria hypoxylon. African Journal of Traditional, Complementary and Alternative Medicines. 13(4):42-46.
- Hammer, K.A., C.F. Carson and T.V. Riley. 1999. Antimicrobial activity of essential oils and other plant extracts. Journal of Applied Microbiology. 86:985-990.
- Ilhan, S., F. Savaroğlu, F. Çolak, C.F. Iscen and F.Z. Erdemgil. 2006. Antimicrobial activity of Palustriella commutata (Hedw.) Ochyra extracts (Bryophyta). Turk. J. Biol, 30:149-152.
- Kartal, M. (2004). Avrupa birliği ülkelerinde tıbbi bitkisel ürünlerin ruhsatlandırılması, Ankara Üniversitesi Eczacılık Fakültesi Farmakognozi Anabilim Dalı.
- Kit Tan, R.M. Flora of Turkey and the East Islands, Vol. 11 (Suppl.); Eds.; Edinburgh University Press, Edinburgh, 2000.
- Nychas, G. J. E. (1995). Natural antimicrobials from plants. In: Gould, G. W. (Ed.), New Methods of Food Preservation. (58pp) London, Blackie: Academic Professional.
- Onbasli, D., G. Yuvali Celik, E.M. Altuner, B. Altinsoy and B. Aslim. 2013. In vitro antimicrobial, antioxidant, and antibiofilm activities of Bryum capillare, a bryophyte sample. Current Opinion in Biotechnology, 24 (Supplement 1): 113.

Özcan, M. and Sağdıç, O. (2003). Antibacterial activity of Turkish spice hydrosols. Food Control.

Prajapati, N. D.; Purohit, S. S.; Sharma, A. K.; Kumar, T. A Handbook of Medical Plants, Agrobios, India, 2003.

Rabe, T. and Van Staden, J. (1997). Antibacterial activity of South African plants used for medicinal purposes. Journal of Ethnopharmacology.

Zhu, Y. P. Chinese Materia Medica, OPA, Amsterdam, 1998.



# Extracts of Common and Black Hawthorn Grown in Turkey: Antimicrobial Activity and GC-MS Analysis

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Abstract: In this study, the antimicrobial activities of the ethanolic extracts of *Crataegus pentagyna* Waldst. & Kit. ex Willd. (Small-flowered Black Hawthorn/Kömüş dikeni) and *Crataegus monogyna* Jacq. (Common Hawthorn /Yemişen), used in the folk-medicine, were tested against gram-negative and gram positive bacteria as well as yeast-like fungi by the agar diffusion method. 19 microorganisms namely, *Enterobacter aerogenes* ATCC 13048, *Salmonella infantis, Listeria monocytogenes* ATCC 7644, *Klebsiella pneumoniae, Pseudomonas aeruginosa* DSMZ 50071, *Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis* ATCC 29212, *Listeria innocua, Salmonella enteritidis* ATCC 13075, *Enterococcus durans, Salmonella typhimurium* SL 1344, *Candida ablicans* ATCC 10231, *Enterococcus faecium* ATCC 25923, *Staphylococcus aureus, Staphylococcus epidermidis* DSMZ 20044, Bacillus subtilis DSMZ 1971, *Escherichia coli* ATCC 25922 and *Serratia marcescens* were used. The extracts of Small-flowered Black Hawthorn and Common Hawthorn showed antibacterial activity against tested microorganisms at different levels.

Keywords: Crataegus pentagyna, Small-flowered Black Hawthorn, Kömüş dikeni, Crataegus monogyna, Common Hawthorn, Yemişen, disc diffusion, GC-MS.

#### **1. INTRODUCTION**

The use of medicinal aromatic plants for different purposes is based on very old times. Medicinal aromatic plants and plant-derived medicines prepared from these plants are nowadays in many countries around the world; herbal medicines, herbs, phytotherapeutics, phytopharmaceuticals and traditional drugs. In today's European Union countries, the name bulun Herbal Medicinal Products inal has been found to be suitable for the use of a common term by the European Medicines Agency (EMEA). Antioxidant flavon and derivatives found in the extracts obtained from the plants are generally found to be antioxidant and the essential oils obtained from plant taxons have been proven to have antimicrobial activities. To list the conditions affecting the antimicrobial activity; taxa of the plant, structure, concentration and expected to show the genus of the microorganism, preservation conditions, etc. They are conditions. Protein, temperature, oils and pH determine the antimicrobial effects of phenolic compounds found in plants. This paper is concerned with antimicrobial activity of ethanol (65%) extracts of Small-flowered Black Hawthorn (*Crataegus pentagyna*) and Common Hawthorn (*Crataegus monogyna*) against 19 microorganisms by using the disk diffusion method, and volatile components of the Small-flowered Black Hawthorn were determined by GC-MS analysis (Gas Chromatography-Mass Spectrometry).

#### 2. MATERIALS AND METHODS

#### **Plant Samples**

In this study, Small-flowered Black Hawthorn (*Crataegus pentagyna*) and Common Hawthorn (*Crataegus monogyna*) were collected from "Sarıkum Nature Conservation Area" (Sinop/Turkey) at sea level. C. pentagyna and C. monogyna fruits were used. Plant materials were dried at room temperature and on draft for three weeks. All the plants were identified by Assist. Prof. Dr. Kerim GÜNEY, Department of Forest Engineering, Faculty of Forestry, Kastamonu University.

#### **Extraction method**

The plants were washed thoroughly 2-3 times with water, and then they were air dried under shade. Afterwards, the dried plant materials were ground in a mixer, the powder was kept in the amber glass bottle. About 10-30 g ground plant

samples were extracted with 250 mL of ethanol (%65) in a Soxhlet apparatus by continuous heat extraction for 24 hours. All extract solutions were filtered through Whatman No.1 paper. Then, filtrates were evaporated with a rotary evaporator. The filtrates were freeze dried and stored in refrigerator at about 4°C after sealed with paraffin for further studies.

#### **Determination of Antimicrobial Activities**

#### **Preparation of Extract Stock**

Extract stocks to test the antimicrobial activity were prepared by dissolving 1 mg of extract in each 3 mL of ethanol for disk diffusion test.

#### Strains

In order to analyse the antimicrobial activity of plants extracts, 19 microorganisms namely, *Enterobacter aerogenes* ATCC 13048, *Salmonella infantis, Listeria monocytogenes* ATCC 7644, *Klebsiella pneumoniae, Pseudomonas aeruginosa* DSMZ 50071, *Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis* ATCC 29212, *Listeria innocua, Salmonella enteritidis* ATCC 13075, *Enterococcus durans, Salmonella typhimurium* SL 1344, *Candida albicans* ATCC 10231, *Enterococcus faecium* ATCC 25923, *Staphylococcus aureus, Staphylococcus epidermidis* DSMZ 20044, *Bacillus subtilis* DSMZ 1971, *Escherichia coli* ATCC 25922 and *Serratia marcescens* were used.

#### **Preparation of Innocula**

All strains were incubated according to their requirements as it was previously mentioned by Altuner and Canlı (2012) and Canlı et al (2015). For the inocula, morphologically similar colonies of each organism were transferred into physiological saline (Canlı et al, 2016a and b, Onbasli, 2013) and to adjust equal the number of the colonies in the solution, 0.5 McFarland standard was used (Hammer et al., 1999; Altuner et al., 2012a and b). Thus standard inocula is adjusted to contain approximately 108 cfu/mL for bacteria and 107 cfu/mL for C. albicans (Canlı et al, 2016c and d).

#### **Disk Diffusion Test**

The disk diffusion test was applied as it was mentioned in the previous studies (Andrews, 2003; Canlı et al, 2014). Petri dishes containing Mueller Hinton Agar were used for disk diffusion test (Ilhan et al., 2006; Canlı et al, 2016e). 20, 40 and 80  $\mu$ L of extracts were loaded on empty sterile antibiotic disks (SAD). Disks were kept at 40°C for 24 h in aseptic conditions (Altuner et al., 2010). Microorganism suspensions were inoculated on the surfaces of the Mueller Hinton plates and left in aseptic conditions for 2-3 minutes before applying disks as described in the previous studies (Altuner and Akata, 2010). Inhibition zones were defined in mm by the method mentioned by Altuner et al. (2014).

#### Controls

Empty SAD was used as negative controls for disk diffusion test, where broth medium inoculated with each microorganism was used to control microorganisms.

#### **Statistical Analysis**

All tests were done in triplicates. All the results given were mentioned as the mean values for these three parallel studies. The statistical analysis was performed using a non-parametric method Kruskal-Wallis one-way analysis of variance. Significance level for p was accepted as 0.05. A value of p < 0.05 was considered statistically significant (Altuner, 2011).

#### GC-MS (Gas chromatography-mass spectroscopy) Analysis

GC-MS analysis was performed using Shimadzu GCMS QP 2010 ULTRA series device. Samples were passed through RTX-5MS capillary column (30 m x 0.25 mm x 0.25 µm). Helium gas was used as carrier gas. The injection temperature was maintained at 250° C. GC-MS analysis was performed according to the procedure in the literature. The oven is heated to 40° C and is allowed to stand at this temperature for 3 minutes. Then the temperature is increased by 4 degrees per minute to 240° C and waited for 10 minutes. Finally, the temperature is increased by 4 degrees per minute to 260° C and kept for 65 minutes and is completed to a total of 78 minutes. The injection temperature was maintained at 250° C and the injection volume was determined as 1 µl. The intermittent temperature is 250° C and the ion boiling temperature is 200°C. It was treated with hexane.

#### 3. RESULTS AND DISCUSSION

The GC-MS results of the extracts are presented in Table 1, and the antimicrobial activity test results are presented in Table 2.

Disk diffusion test results showed that Small-flowered Black Hawthorn is active against K. pneumoniae, E. durans and E. faecium with inhibition zones between 7-8 mm, where Common Hawthorn is active against L. monocytogenes, K. pneumoniae, P. fluorescens, S. kentucky, L. innocua, S. enteritidis, S. typhimurium, E. faecium, E. coli and S. marcescens with inhibition zones between 7-9 mm.

#### 4. CONCLUSION

Plants are natural reservoirs of various phytonutrients and compounds which are urgent and essential to life in general. The phytochemical component of ethanol extract of Crataegus pentagyna Waldst. & Kit. ex Willd. (Small-flowered Black Hawthorn/Kömüş dikeni) and Crataegus monogyna Jacq. (Common Hawthorn/Yemişen) Mill. revealed by GC-MS analysis shows its significance in phytopharmaceuticals, cosmetic and food industries. On the basis of antimicrobial activity, it was monitored that the ethanol extracts were effective only for few of the tested species.

#### Acknowledgements

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|      |         | Small-fl | owered Black Hawthorn   | Common Hawthorn |        |  |
|------|---------|----------|---|-----------------|--------|--|
| Peak | R. Time | Area %   | Component Name  | R. Time         | Area % | Component Name                                 |
| 1    | 3.702   | 3.56     | 2,2-dimethoxybutane   | 3.705           | 4.62   | 2,2-dimethoxybutane                            |
| 2    | 13.849  | 1.08     | Decane, 3,7-dimethyl-   | 13.849          | 1.37   | Decane, 3,7-dimethyl-                          |
| 3    | 29.629  | 2.61     | -   | 22.241          | 1.37   | Hexadecane                                     |
| 4    | 32.676  | 1.34     | Diethyl Phthalate   | 23.863          | 1.05   | Hexadecane                                     |
| 5    | 34.703  | 1.69     | -   | 29.474          | 1.50   | Eicosane                                       |
| 6    | 41.708  | 3.35     | Hexadecanoic acid, methyl ester                               | 29.627          | 3.77   |  |
| 7    | 45.838  | 4.40     | 9,12-Octadecadienoic acid (Z,Z)-,<br>methyl ester             | 32.689          | 1.20   | Diethyl Phthalate                              |
| 8    | 45.977  | 5.24     | 9-Octadecenoic acid, methyl ester,<br>(E)-                    | 34.702          | 2.18   | -  |
| 9    | 46.585  | 1.29     | Methyl stearate   | 35.859          | 1.30   | Docosane (CAS)                                 |
| 10   | 50.414  | 2.59     | Hexadecanoic acid, 1-<br>(hydroxymethyl)-1,2-ethanediyl ester | 41.581          | 1.01   | Docosane (CAS)                                 |
| 11   | 54.145  | 3.75     | 9,12-Octadecadienoyl chloride, (Z,Z)-                         | 41.720          | 2.59   | Hexadecanoic acid, methyl ester                |
| 12   | 54.244  | 7.21     | DI-(9-Octadecenoyl)-Glycerol                                  | 45.852          | 3.62   | 9,12-Octadecadienoic acid (Z,Z)-, methyl ester |
| 13   | 54.395  | 2.11     | -   | 45.982          | 3.92   | 9-Octadecenoic acid, methyl ester, (E)-        |
| 14   | 54.486  | 1.24     | -   | 46.590          | 1.44   | Octadecanoic acid, methyl ester (CAS)          |
| 15   | 60.669  | 1.85     | Pentacosane   | 50.407          | 2.13   | Octacosane                                     |
| 16   | 62.020  | 1.71     | Tetracosanoic acid, methyl ester                              | 54.160          | 2.88   | 9,12-Octadecadienoyl chloride, (Z,Z)-          |
| 17   | 65.384  | 8.41     | .gammaSitosterol  | 54.244          | 4.57   | DI-(9-Octadecenoyl)-Glycerol                   |
| 18   | 65.846  | 6.88     | Squalene  | 54.376          | 2.36   | Bicyclo[10.1.0]tridec-1-ene                    |
| 19   | 68.111  | 12.8     | Nonacosane  | 54.500          | 1.78   | -  |
| 20   | 69.257  | 1.34     | Hexacosanoic acid, methyl ester                               | 54.686          | 1.93   | Pentacosane                                    |
| 21   | 77.181  | 7.3      | dlalphaTocopherol   | 60.682          | 3.22   | Hexatriacontane                                |
| 22   | 77.370  | 3.5      | Octacosanoic acid, methyl ester                               | 62.023          | 1.25   | Tetracosanoic acid, methyl ester               |
| 23   |         |          |   | 64.722          | 2.33   | Ethyl tetracosanoate                           |
| 24   |         |          |   | 65.390          | 2.31   | -  |
| 25   |         |          |   | 65.856          | 7.22   | Squalene                                       |
| 26   |         |          |   | 68.119          | 12.77  | Pentacosane                                    |

Table 1. GC-MS Analysis of Small-flowered Black Hawthorn and Common Hawthorn

| 27 |  | 71.232 | 4.75 | Octadecanoic acid, butyl ester (CAS) |
|----|--|--------|------|--------------------------------------|
| 28 |  | 73.729 | 3.04 | Lauric acid, 2-methylbutyl ester     |
| 29 |  | 77.193 | 2.00 | -                                    |
| 30 |  | 77.240 | 1.52 | -                                    |

 Table 2. Disk Diffusion Test Results of Extracts at the Small-flowered Black Hawthorn and Common Hawthorn

 Concentration

|                  | Small-flov | Small-flowered Black Hawthorn |       |       | Common Hawthorn |       |  |
|------------------|------------|-------------------------------|-------|-------|-----------------|-------|--|
| Microorganisms   | 20 µL      | 40 µL                         | 80 µL | 20 µL | 40 µL           | 80 µL |  |
| E. aerogenes     | -          | -                             | -     | -     | -               | -     |  |
| S. infantis      | -          | -                             | -     | -     | -               | -     |  |
| L. monocytogenes | -          | -                             | -     | -     | 8               | 8     |  |
| K. pneumoniae    | 8          | 8                             | 8     | 8     | 8               | 8     |  |
| P. aeruginosa    | -          | -                             | -     | -     | -               | -     |  |
| P. fluorescens   | -          | -                             | -     | 8     | 8               | 8     |  |
| S. kentucky      | -          | -                             | -     | 8     | 8               | 8     |  |
| E. faecalis      | -          | -                             | -     | -     | -               | -     |  |
| L. innocua       | -          | -                             | -     | 7     | 8               | 9     |  |
| S. enteritidis   | -          | -                             | -     | -     | -               | 8     |  |
| E. durans        | -          | 7                             | 8     | -     | -               | -     |  |
| S. typhimurium   | -          | -                             | -     | -     | 8               | 8     |  |
| C. ablicans      | -          | -                             | -     | -     | -               | -     |  |
| E. faecium       | -          | 7                             | 7     | 7     | 7               | 7     |  |
| S. ayreus        | -          | -                             | -     | -     | -               | -     |  |
| S. epidermidis   | -          | -                             | -     | -     | -               | -     |  |
| B. subtilis      | -          | -                             | -     | -     | -               | -     |  |
| E. coli          | -          | -                             | -     | -     | -               | 7     |  |
| S. marcescens    | -          | -                             | -     | 7     | 8               | 9     |  |

#### REFERENCES

- Altuner, E.M., I. Akata and K. Canlı. 2012a. In vitro antimicrobial screening of Bovista nigrescens (Pers.). Kastamonu U. J. For. Fac. 12:90-96.
- Altuner, E.M., I. Akata and K. Canlı. 2012b. In vitro antimicrobial screening of Cerena unicolor (Bull.) Murrill (Polyporaceae Fr. Ex Corda). Fresen. Environ. Bullet. 21:3704-3710.
- Altuner, E.M. and I. Akata. 2010. Antimicrobial activity of some macrofungi extracts. Sakarya Üniversitesi Fen Bilimleri Enstitüsü Dergisi. 14(1):45-49.
- Altuner, E.M., K. Canlı and I. Akata. 2014. Antimicrobial screening of Calliergonella cuspidata, Dicranum polysetum and Hypnum cupressiforme. Journal of Pure and Applied Microbiology. 8(1):539-545.
- Altuner, E.M. and K. Canlı. 2012. In vitro antimicrobial screening of Hypnum andoi A.J.E. Sm. Kastamonu U. J. For. Fac. 12:97-101.
- Altuner, E.M. 2011. Investigation of antimicrobial activity of Punica granatum L. fruit peel ash used for protective against skin infections as folk remedies especially after male circumcision. African Journal of Microbiology Research. 5(20):3339-3342.
- Andrews, J.M. 2003. BSAC standardized disc susceptibility testing method (version 6). Journal of Antimicrobial Chemotherapy. 60:20-41.
- Canlı, K., I. Akata and E.M. Altuner. 2016c. In vitro antimicrobial activity screening of Xylaria hypoxylon. African Journal of Traditional, Complementary and Alternative Medicines. 13(4):42-46.
- Canlı, K., E.M. Altuner, I. Akata, Y. Türkmen and U. Üzek. 2016a. In vitro antimicrobial screening of Lycoperdon lividium and determination of the ethanol extract composition by gas chromatography/mass spectrometry. Bangladesh Journal of Pharmacology. 11(2):389-394.

- Canlı, K., E.M. Altuner and I. Akata. 2015. Antimicrobial screening of Mnium stellare. Bangladesh Journal of Pharmacology. 10:321-325.
- Canlı, K., B. Çetin, E.M. Altuner, Y. Türkmen, U. Üzek and H. Dursun. 2014. In vitro antimicrobial screening of Hedwigia ciliata var. leucophaea and determination of the ethanol extract composition by gas chromatography/mass spectrometry (GC/MS). Journal of Pure and Applied Microbiology. 8(4):2987-2998.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016d. In vitro antimicrobial screening of Aquilaria agallocha roots. African Journal of Traditional, Complementary and Alternative Medicines. 13(5):178-181.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016e. In vitro antimicrobial activity screening of Rheum rhabarbarum roots. International Journal of Pharmaceutical Sciences Invention, 5(2):1-4.
- Hammer, K.A., C.F. Carson and T.V. Riley. 1999. Antimicrobial activity of essential oils and other plant extracts. Journal of Applied Microbiology. 86:985-990.
- Ilhan, S., F. Savaroğlu, F. Çolak, C.F. Iscen and F.Z. Erdemgil. 2006. Antimicrobial activity of Palustriella commutata (Hedw.) Ochyra extracts (Bryophyta). Turk. J. Biol, 30:149-152.
- Kartal, M. (2004). Avrupa birliği ülkelerinde tıbbi bitkisel ürünlerin ruhsatlandırılması, Ankara Üniversitesi Eczacılık Fakültesi Farmakognozi Anabilim Dalı.
- Nychas, G. J. E. (1995). Natural antimicrobials from plants. In: Gould, G. W. (Ed.), New Methods of Food Preservation. (58pp) London, Blackie: Academic Profesional.
- Onbasli, D., G. Yuvali Celik, E.M. Altuner, B. Altinsoy and B. Aslim. 2013. In vitro antimicrobial, antioxidant, and antibiofilm activities of Bryum capillare, a bryophyte sample. Current Opinion in Biotechnology, 24 (Supplement 1): 113.
- Özcan, M. and Sağdıç, O. (2003). Antibacterial activity of Turkish spice hydrosols. Food Control.
- Rabe, T. and Van Staden, J. (1997). Antibacterial activity of South African plants used for medicinal purposes. Journal of Ethnopharmacology.



# The Effects of Maternal Obesity from The Fetus Period to The Adolescent Period

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Abstract: This study aimed to investigate the short- and long-term effects of maternal obesity on fetus, newborn, childhood and adolescence.

Worldwide, the prevalence of obesity is increasing day by day. Obesity is the most common medical condition in women during the reproductive age. Maternal obesity and excessive gestational weight gain result in fetal overfeeding, leading to obesity and metabolic disturbances in children. Maternal obesity causes pregnancy complications and has short- and long-term negative consequences for both mother and child. Prematurity, stillbirth, multiple maternal and fetal complications such as congenital anomalies, macrosomia, birth traumas, failure to breastfeed are associated with maternal obesity and excessive gestational weight gain. Maternal obesity is associated with impairment in cognitive function as well as triggering obesity, cardiovascular diseases, diabetes, atopic diseases and asthma in children. Maternal obesity constitutes cyclical obesity between generations.

All these health problems can be eliminated by the prevention of obesity in women of reproductive age who should be a global public health priority. Pre-pregnancy counseling is important to optimize maternal health and improve prepregnancy BMI. Women of reproductive age should be aware of the effects of obesity on the short and long-term health of children, and weight gain should be ensured within the limits determined by the Medical Institute in pregnancy. The importance and protective properties of breast milk should be explained, and mothers should be encouraged for breastfeeding. Therefore, in view of the increase in global obesity, effective interventions for ethnicity and cultures are needed to improve the health of women and children.

Keywords: Maternal obesity, gestational weight gain, body mass index, fetal complications, childhood obesity.



# High Performance Computing on Big data in Geographic Information Systems

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Abstract: It is increasingly necessary to process data every day, therefore, it is necessary to use high-performance computing to process these big data. In Geographic Information systems (GIS), there are billions of spatial points to be handled in a reasonable amount of time. One of the basic operations is to prepare triangulation data. There are many triangulation methods. In this study which we utilized Delaunay triangulation (DT). The Delaunay triangulation is a method to find triangles of a set of points, on a plane. The Delaunay triangles are defined as the triangles of a set P of n points where three points of P form a triangle and the circumscribed circle of each triangle contains no points of P internally. Each point of P is on the circumcircles. These triangles create a connected graph of all the points, which makes it possible to maximize the smallest angle of the set, which gives a mesh of triangles containing few lean triangles. Since the mesh has few thin triangles, this can be an advantage for applications such as humans or machines. Due to the complexity of the triangulation process, the calculation time increases as the amount of points increases. To overcome this problem in this paper, we propose a three-stage divide and conquer method which we call "sewing method". At the first stage, we run the triangulation method for each sub partition. At the next stage, we recognize the border of each sub partition by using convex hull. The convex hull is usually the first step in determining the Delaunay triangulation as it can be used a boundary for algorithms to avoid searching for triangles outside P. convex hull is a subgraph of the Delaunay triangulation of P and is where most of the thinner triangles are located. This is since the circumcircles for these edges often do expand outward from P as possibly very large circles. For the last step, we implemented a new sewing method which joins these sub partitions.

In our implementation, when we applied our sewing technique we achieved 26% effeicency on two million points. Also, triangulation of twenty million points required 170.2 second in the new implementation. On the other hand, it took 527.0 second for the usual method. Thus we accomplished 67.7% efficiency. Therefore, we proved that "sewing method" is effective on big data compared to the conventional procedures.

Keywords: Delaunay triangulation, GPS, Big Data, High performance, convex hull, sewing.



# A Literature Review of Ambient Intelligence (AmI)

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**Abstract:** Today, more than half of the world's population uses powerful processors, sensors, cameras, high-speed communications and personal devices with intelligent applications. All these technologies are expected to be an environmental intelligence revolution that we can use under the conditions of our environment in the coming years. Embedded devices with Ambient Intelligence (AmI) enable users to interact with the users in the environment by means of interfaces and respond accordingly. Machine learning, embedded systems, the internet of things (IoT), artificial intelligence (AI), robotics, nanotechnology and other emerging technologies is the most popular architecture of the fourth industrial revolution. The complexity of technology and their markets is defined quickly and appropriately by ambient intelligence. A holistic approach has been adopted, and it has been determined which areas should work on in order to realize media intelligence on issues such as technology, society and business. In this article, information about the ambient intelligence is given and the predictions have been presented with the studies.

**Keywords:** Ambient Intelligence (AmI), Artificial Intelligence (AI), Decision Making, Context Awareness, Internet-of-Things, Sensors.

#### **1. INTRODUCTION**

Ambient Intelligence (AmI), which is a sub-branch of artificial intelligence, is known as the technology of the future with its sensitive and adaptive features wherever it is informatics. AmI is a hidden smart network that is sensitive to the challenges in our daily life. Sensitive to the needs, habits, gestures and emotions of assets, the main factor of this digital media tool is intelligence.

AmI researchers first created a requirement as a result of problems arising from information engineering, electrical engineering and medical developments. AmI in informatics is formed by the presence of people and electronic devices. As can be seen in Figure 1, an ambient intelligence methodology supports devices to inform people about their daily living activities and tasks through a network. Intelligence and intelligence features of the devices used for ambient intelligence are everywhere. Thanks to these characteristics, there are many interconnected embedded systems running in the background. These systems recognize the people in the environment they live in and learn their behavior and program themselves to meet their needs. These programs and systems can be used and configured seamlessly in computing environments without the need for users to program or connect each device.





Media intelligence is based on three new basic technologies. These;

• Ubiquitous Computing: It is the integration of everyday used objects with microprocessors. They are computers that operate in the background in a way that people are not aware of the existence of people using information technologies.

• Ubiquitous Communication: Objects that enable technologies such as ad-hoc and wireless networking to communicate with each other and the user.

• An Intelligent User Interface: The AmI environment can be controlled and interacted in a natural and personalized environment.

As shown in Figure 2, ambient intelligence can detect the environment through software algorithms and use this information in a different task to change the environment and the environment. This sensing is carried out with the help of sensors while the sensors are designed for position measurement in situations such as the detection of chemicals and moisture. The sensors are integrated into small and embedded systems. A problem with wireless sensors and wireless sensor networks is an important resource to support long-term data collection.



Figure 2. General architecture of ambient intelligence [9].

The data from the sensors can be read using conventional data analysis techniques, although the complexity of the multiplicity and multiplicity of data is complicated. These data analysis techniques consist of algorithms that need to be carried out in a number of ways, such as user modeling, estimation and recognition, decision-making and reasoning. Security and confidentiality are important issues in all areas of informatics as well as security problems in these media intelligence technologies. Sensor reliability, operating errors and installation errors can create security risks. To ensure the safety of sensor networks, designers should pay attention to the safety and reliability of the sensor communication channel together with the data security issues. However, other adversities in the ambient intelligence technology are limited battery life for wireless sensors and software - user errors.



Figure 3. Structures forming the ambient intelligence.

As shown in Figure 3, technologies that create media intelligence are; Smart agent, lifelog, personal health record, context awareness, IoT device, environment and ambient sensor.

Today, applications that can exemplify the ambient intelligence; smart homes, health related applications, public transport sector, education system, emergency services, production-oriented places.

#### 2. LITERATURE

The concept of rapidly developing media intelligence has been mentioned in many studies in the literature from past to present.

In 2004, Hagras et al. [1] developed an adaptive control, iDorm, which envisages online, lifelong, personalized learning to develop learning and adaptation techniques for embedded systems. IDorm has three types of embedded computing artifacts belonging to the network infrastructure. The first is a physically static calculation that is closely related to the building. With the help of sensors over the network, it takes the information containing the user's behavior and calculates the appropriate control actions with the fuzzy ISL system. Then it sends them to the iDorm effectors. The second type is a physical mobile service robot. This robot serves users by performing ambient intelligence and working online. All these operations can be realized thanks to the rich sensors it has. The third type is a physically portable computing device. iDorm is a wearable technology that can be monitored and controlled wirelessly. This type, which has a standard Java encoding, can be used as remote control. At the same time, Bluetooth and wireless network connections make it possible to set up and use all in and out of the environment.

In 2005, Kaiser et al. [2] reported network-based mechanical systems (NIMS) using media intelligence. NIMS is used to analyze complexities in three-dimensional environments with its infrastructure and mobility. At the same time, it plays an active role in the restructuring of uncertainty in the physical perception of infrastructure and sensors. This basket offers a unique feature for obtaining and transporting all physical numbers. The study also discussed NIMS, sensor diversity capability, sensor uncertainty reduction, adaptive sensor fusion, and the benefits of speed-distortion, bandwidth, and extension of energy limits in scattered sensor networks.

In 2006, Nehmer et al. [3] made predictions about the use of ambient intelligence in life support systems. Automated life support systems represent a promising approach to enable people with disabilities and older people to live independently and to improve their quality of life and to minimize social and medical care needs. For this reason, while mentioning that life support systems should be able to adapt and adapt at an algorithmic, architectural and human interface level to an unknown extent in existing systems, they also talk about human-computer interfaces, software architectures, resource efficiency and security challenges.

In 2009, a study by Eisenhauer et al. [4] developed a project called HYDRA to create ambient intelligence applications based on wireless devices and sensors. This project has been developed for the development of intermediate layers to network visual systems. Thanks to the combination of service-based architecture (SoA) and Semantic-based model architectures, this software has enabled the development of public services based on open standards.

In a study by Kim [5] in 2018, he designed an intelligent classroom system using an ambient intelligence algorithm to assess students' level of participation in the classroom. The algorithm evaluates the students' psychological status by measuring the thermal infrared images of the students in the classroom. An innovative algorithm was used because the algorithm used in the study gave feedback to the students in real time against their students. At the same time, this study encourages the education and training of objects in the Internet. In particular, a model was created to measure student participation levels using thermal infrared imaging. The mobile applications of the teachers in the class communicate with the system and warn the user in real time. Biomedical signals such as electrocardiography, or blood pressure monitoring methods were collected. It is likened to the use of traffic lights as a method of expressing the student's level of participation. Light levels were used to minimize complexity. The green light represents the good participation of the student in the class, while the yellow light participation is moderate and the red light represents little.

In another study published by Kim et al. [6] in 2018, they developed an adaptive neural network-based estimation model for ambient intelligence. In the study, similarities in a life platform were calculated. In a conventional estimation

procedure, an error rate is used to update a weight in the neural network. In the proposed model, the rates of similarity of users to the environment were evaluated. In order to calculate this similarity network, user clustering and average deviations were taken into account. Collaborative filtering technology was used to estimate the values of the user and to find out the similarity rates repeatedly using the neural network. According to the performance results, the proposed neural network-based similarity weight method gave the highest estimation accuracy when the learning rate was 0.001. As a result, they have proved that the media addiction is a new technology that can be improved in order to contribute to the development of the health sector and improve the quality of life.

In another study conducted in 2018, Sanders et al. [7] proposed a decision-making system for energy savings in compressed air systems using ambient intelligence and artificial intelligence. In this research, by combining real-time environment perception with artificial intelligence and information management systems, the efficiency of energy in intensive production has been automatically increased. It also aimed to minimize energy use for air compressors based on real-time production conditions. He gave detailed information about the performance of the study. The ambient data were automatically interpreted by artificial intelligence and then exhibited behavior for the future. Information management was used to archive information about actions to reduce energy use and maintain productivity. New techniques have been used to save energy in compressed air systems.

In a study published by Chen et al. [8] in 2018, they talked about an application for the sustainable development of industry on industrial engineering concepts and techniques using ambient intelligence. For this reason, they have applied two important industry concepts and techniques such as planning cycle and cost-benefit analysis to AmI system. In the first stage of this, the user side, the server side and the AmI system were considered as a whole and then a five-stage planning cycle in which a detailed cost-benefit analysis of the collective targets was calculated was proposed. They explained this proposed methodology on a restaurant recommendation system. The experimental results proved that the proposed system and methodology can perform a reliable cost-benefit analysis and improve system performance.

#### 3. RESULTS AND DISCUSSION

In this study, firstly the methodology of ambient intelligence is mentioned and the studies that are close to the present and past in the literature are mentioned. Approximately 20 years ago, the concept of ambient intelligence emerged, but has become technically feasible under today's conditions. Many areas have turned their business models upside down, but ambient intelligence has gained the trust of users. In order to maintain this situation, it will require much more improvement in the future, such as accuracy, security, reliability and privacy. However, users have begun to take advantage of the environmental intelligence offered by smart home-like applications. In this way, it will affect many aspects of our lives with all usage models of ambient intelligence.

#### REFERENCES

- [1] Hagras, Hani, et al. "Creating an ambient-intelligence environment using embedded agents." IEEE Intelligent Systems 19.6 (2004): 12-20.
- Kaiser, William J., et al. "Networked infomechanical systems (nims) for ambient intelligence." Ambient Intelligence. Springer, Berlin, Heidelberg, 2005. 83-113.
- [3] Nehmer, Jürgen, et al. "Living assistance systems: an ambient intelligence approach." Proceedings of the 28th international conference on Software engineering. ACM, 2006.
- [4] Eisenhauer, Markus, Peter Rosengren, and Pablo Antolin. "A development platform for integrating wireless devices and sensors into ambient intelligence systems." Sensor, Mesh and Ad Hoc Communications and Networks Workshops, 2009. SECON Workshops' 09. 6th Annual IEEE Communications Society Conference on. IEEE, 2009.
- [5] Kim, Pyoung Won. "Ambient intelligence in a smart classroom for assessing students' engagement levels." Journal of Ambient Intelligence and Humanized Computing (2018): 1-6.
- [6] Kim, Joo-Chang, and Kyungyong Chung. "Neural-network based adaptive context prediction model for ambient intelligence." Journal of Ambient Intelligence and Humanized Computing (2018): 1-8.
- [7] Sanders, D., et al. "Making Decisions about Saving Energy in Compressed Air Systems using Ambient Intelligence and AI." IEEE Proceedings of the SAI Conference on Intelligent Systems. 2018.
- [8] Chen, Toly, and Horng-Ren Tsai. "Application of industrial engineering concepts and techniques to ambient intelligence: a case study." Journal of Ambient Intelligence and Humanized Computing 9.2 (2018): 215-223.
- [9] Augusto, Juan Carlos, and Paul J. McCullagh. "Ambient intelligence: Concepts and applications." Comput. Sci. Inf. Syst.4.1 (2007): 1-27.



# **ORAL PRESENTATION**

# Quantum Fourier Transform: Using Five Qubits On QISKIT

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Abstract: Quantum computing, the use of quantum computers that use quantum physics to process data, arrange, store and so on. Classical computer deals with data called bits. On the other hand, quantum computers operate on quantum bits (qubits). Qubits are represented by basis vectors expressed as  $|0\rangle$  and  $|1\rangle$ . In quantum computing, a quantum logic gate (quantum gate) is a basic quantum circuit operating on qubits. They are the building blocks of quantum circuits. While some gates operate on a single qubit. Also, some quantum gates such as Toffoli and Controlled NOT gate manipulate multiple qubits. Pauli gates, Hadamard are examples of single-qubit gates. In Quantum computing there are basic operations defined such as Quantum Fourier Transform (QFT). QFT is the main operation in Shor's Prime factorization algorithm announced in 1995. on qubits, we use QFT for a linear transformation.

Because the lack of resources, we cannot handle our huge applications on physical quantum computers. So we are able to simulate on classic computers. There are some simulator tools such as QISKIT that developed by IBM and QX by QU Touch company.

In this research, we used QISKIT simulator which is an open-source platform. There are Education and Business options for QISKIT. We can install It on a local machine or we access online and it includes a code editor (QSM) and a graphical user interface.

We implemented a 5-qubit QFT algorithm with QISKIT. We used 5 Hadamard gates and 10 phase flip gates. The longest path has 5 gates. We tested our design on IBM Quantum Computer in Yorktown.

Keywords: Quantum computing, QFT, Quantum computer, QUBIT, quantum operator, QISKIT.



# Nuclei Cells Detection and Segmentation with Deep Neural Network

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Abstract: The breast cancer is the most popular in mammography because in the world many people die from this type of cancer. From 8 women 1 one them has these problem. Mammography is the x-ray picture of the breast. Mammography can be used to check for breast cancer in women who have no signs or symptoms of the disease. It can also be used if you have a lump or other sign of breast cancer. Recently, the breast cancer is one of the most massive disease in the world. Also, the most people in world die from this disease. As United State medical statistic center about 81% of the people that they died include the women, other percentages are men. Hematoxylin and Eosin stain (H&E stain or HE stain) is one of the principal stains in histology. It is the most widely used stain in medical diagnosis and is often the gold standard; for example, when a pathologist looks at a biopsy of a suspected cancer, the histological section is likely to be stained with H&E. A combination of hematoxylin and eosin, it produces blues, violets, and reds.

Deep NN is just a deep neural network, with a lot of layers. It can be CNN, or just a plain multilayer perceptron. CNN, or convolutional neural network, is a neural network using convolution layer and pooling layer. The convolution layer convolves an area, or a stuck of elements in input data, into smaller area to extract feature. It is done by filtering an area, which is as same as to multiplying weights to an input data. The pooling layer picks a data with the highest value within an area. These layers act to extract an important feature from the input for classification.

In this study we worked on the nuclei image segmentation and detection on public breast cancer images which we used from Tumor Proliferation Assessment Challenge (TUPAC) and the Triple Negative Breast Cancer (TNBC) data sets.

For accuracy evaluation we used the confusion matrix that calculate the true negative (TN), true positive (TP), False positive (FP) and false negative (FN). We compared the output result with the ground truth images. For each pixel we calculated these criteria.

Keywords: Image segmentation, Nuclei Cells Detection, Deep Neural Network.



# High Performance Quantum Chemical Calculations for Polymers

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**Abstract:** Polymers are classes of natural or synthetic substances composed of very large molecules, called macromolecules, which are multiples of simpler chemical units called monomers. Polymers establish a considerable lot of the materials in living creatures, including, proteins, cellulose and nucleic acids.

Quantum chemistry is the application of quantum mechanical principles and equations to understand molecules. Quantum chemistry distinguishes itself from other computational methods. It is established on laws of nature: "Quantum mechanics". The Quantum chemical calculations allow us to accurately predict the structures of molecules and the spectroscopic behavior of atoms and molecules for their previous study.

Computational chemistry can be described as the way we use chemistry using computers instead of real chemical components. Over the years, powerful molecular modeling tools have been developed that can predict the structures, energy, reactivities and other properties of molecules.

These developments have occurred largely due to the dramatic increase in the speed of the computer and the design of efficient quantum chemical algorithms.

All quantum chemical calculations use a special system of units which are very natural and greatly simplify expressions for various quantities.

The purpose of this research is to perform simulations and calculations of chemical polymers using the open source package PSI4. PSI4 allows us to perform efficient and highly accurate simulations of a variety of molecular properties.

Keywords: Quantum, Polymers, Chemistry, High performance, Quantum chemistry, PSI4.



# The Comparison of Graph Database Query Languages

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Abstract: The popularity of graph databases among NoSQL databases is increasing. Graph databases allow you to store data as entities (nodes) and relationships (columns), and to query the data as a graph. In this study, SPARQL (Query Language for RDF) and GraphQL languages, which are two of the graph database query languages, were comparatively examined. The advantages and disadvantages of the query languages used on the DBPedia data set are given. GraphQL is the client-level query language developed by Facebook GraphQL is one of the most modern ways of creating and querying APIs. GraphQL is a syntax that explains how to request data. It is usually used to load data from a server to the client. With GraphQL, the user can make a single call to get the required information instead of creating a few REST requests to get the same information. SPARQL is both a protocol and query language that is able to manipulate data stored in RDF (Resource Description Framework) format. It is considered as a standard and one of the key technologies of the semantic web. As a result; The biggest disadvantage of GraphQL compared to SPARQL is that it does not have the concept of semantics, so it requires an interface specific schema. Therefore, it makes it difficult to combine GraphQL data from different sources. This is further complicated by the fact that GraphQL does not have the concept of global identifiers possible in the RDF through the use of URIs. Also, GraphQL is not as meaningful as SPARQL. GraphQL queries represent trees, and not full graphs are not as in SPARQL.

Keywords: GraphQL, SPARQL, Query Languages, REST, DBPedia

#### **1. INTRODUCTION**

The DBpedia data set is a central resource in the connected open data cloud that can be counted among the largest datasets in the database, in terms of data and topics. Nowadays, DBpedia data set is usually taken from Wikipedia based on the contents of the page and collected using 16 extractors (Gandon et al., 2016). The DBpedia data set also aims to extract content made from information created on Wikipedia.

GraphQL, developed by Facebook, is a type of NoSQL database query language developed to implement web-based APIs. SPARQL is both a protocol and a query language that can manage data stored in the RDF (Resource Description Framework) format.

Graph database is a kind of NoSQL database system. The difference from other NoSQL databases is that it can communicate with nodes. Relationships in SQL databases are usually kept on separate tables, indexes and logical keying of the data are made with the primary key. In NoSQL databases, relations are kept on a single structure. Some of the graph database query languages are GraphQL and SPARQL.

In different studies, GraphQL and SPARQL have been applied successfully on different data sets. In this study, the advantages and disadvantages of GraphQL and SPARQL are given by using ready tools on DBpedia data set.

#### 2. MATERIALS AND METHODS

#### **Overview on DBPedia**

DBpedia is created in LOD Cloud Diagram and is data from Wikipedia. The class establishes the category hierarchy with 359 classes and the classes are defined with 1775 different features. The data set contains multiple data files, and these data files contain different data from Wikipedia. Wikipedia is not semantic. Because it is not a natural language (Zhang et al., 2014). However, the dataset establishes a semantic relationship according to the attribute property.

Wikipedia contains a lot of information about the world, and most of this information includes information about a number of features or subjects of people who are part of DBpedia. The DBpedia data set contains structural information from Wikipedia (Lehmann et al., 2015). It is a study that can be accessed with SPARQL query language, which is one of Graph database types.

### GraphQL: A New Perspective on Data Access

GraphQL is an open-source data query for APIs and a runtime for manipulation language and execution of queries with existing data. GraphQL query on the left side of the Figure 1 is given. On the right side, there is an answer to our query. The classic Rest/MVC structure requires at least 2-3 different controllers and can be executed with a few HTTP Requests.



Figure 1. GraphQL query

GraphQL consists of server (Application Level Query Layer) and client. Information about the data to be sent to the client by the server (GraphQL Schema, Mutation, etc.). GraphQL provides an interface between the server and the client, allowing clients to create their own queries. The GraphQL client does not have a very complex structure and is not technically necessary. In the server part, it can be performed for all kinds of languages without substructure ready by following the GraphQL standards. The GraphQL server has the following structure.

- schema: It needs data layer with data type obligation, i.e. data schemes.
- resolve functions: A function that defines how and how the server retrieves data.

The GraphQL is designed with some broad objectives in mind (Bryant, 2017):

- Reduce data transfer by considering other web service models, such as REST.
- Reduce the potential for error due to invalid queries in client part.
- Supporting a developing data model without API version.

Figure 2 shows how to query an RDF repository through a GraphQL server. In response, it contains linked data as a JSON-LD object. JSON-LD is the use of a JavaScript object in the HTML to define the data.



Figure 2. Querying DBPedia with GraphQL

#### SPARQL: RDF Query Language

RDF sets standards for the metadata definition of computers so that computers can understand and process data on the web and integrate them into different web content (Zhou et al., 2017). The structure underlying any expression in the RDF is a collection of triples, each of which consists of a subject, a predicate, and an object. A group of such triples is called an RDF graph. RDF requires a standard data query language to facilitate the design process. SPARQL (SPARQL Protocol and RDF Query Language) is a query language used for RDF data (Soussi et al., 2017). DBpedia is using Virtuoso as the underlying triple store. Figure 3 shows an example of a query to know about living in Turkey and having a date of birth less than 01.01.2000.

| Default   | Data Set Name (Graph IRI)   |
|---|---|
| nup.//db  | bedia.org   |
| Query T   | ext   |
| PREFIX<br>PREFIX<br>PREFIX<br>SELECT<br>WHERE<br>{                            | <pre>db: <http: dbpedia.org="" resource=""></http:> foaf: <http: 0.1="" foaf="" xmlns.com=""></http:> dbo: <http: ?birth="" ?death="" ?name="" ?person<="" dbpedia.org="" ontology="" pre=""></http:></pre> |
| <pre>?pers<br/>?perso<br/>?perso<br/>?pers<br/>FILTEF<br/>}<br/>ORDER E</pre> | on dbo:birthPlace db:Turkey .<br>n dbo:birthDate ?birth .<br>n foaf:name ?name .<br>on dbo:deathDate ?death .<br>(?birth < "2000-01-01"^^xsd:date)<br>Y ?name   |
| LIMIT 1   | 00  |

Figure 3. Example of SPARQL query on DBPedia data set

#### 3. RESULTS AND DISCUSSION

SPARQL query language;

- RDF is a metadata model defined by W3C that is available for use.
- The SPARQL query language is a standardized language on the W3C platform for RDF queries.
- The RDF and SPARQL query language eliminates the need to create or learn a new API.
- Creating an endpoint is highly possible in the SPARQL query language.

GraphQL query language;

• To use the GraphQL query language, it is not necessary to create a semantic ontology and even if it is possible to use the GraphQL query language.

• As with the SPARQL query language, GraphQL also tries to avoid the use of multiple requests in the query language.

• GraphQL allows the creation of different types of data sources. REST APIs, which are a distributed system that mostly use web protocols and technologies.

The similar and different aspects of GraphQL and SPARQL query languages in terms of low developer effort, wide availability of professional development tools, high expression and reusability of queries between datasets are given in Table 1.

|  | GraphQL | SPARQ |
|--|---------|-------|
| Low developer effort                                     | +       | -     |
| Extensive availability of professional development tools | +       | -     |
| High expression  | -       | +     |
| Reusability of queries between datasets                  | -       | +     |

Table 1. Differences Between GraphQL and SPARQL

#### REFERENCES

- Bryant, M., 2017. GraphQL for Archival Metadata: An Overview of the EHRI GraphQL API, IEEE International Conference on Big Data (BIGDATA), 2226.
- Gandon, F., Boyer, R., Corby, O., and Monnin, A., 2016. Wikipedia editing history in DBpedia extracting and publishing the encyclopedia editing activity as linked data, 2016 IEEE/WIC/ACM International Conference on Web Intelligence, pp. 479-482.
- Lehmann, R., Isele, M., Jakob, A., Jentzsch, D., Kontokostas, P., Mendes, N., Hellmann, S., Morsey, M., Kleef, P., Auer, S., and Bizer, C., 2015. DBpedia A large-scale, multilingual knowledge base extracted from Wikipedia. Semantic Web, 6(2): 167-195.
- Soussi, N., Boumlik, A., and Bahaj, M., 2017. Mongo2SPARQL: Automatic and semantic query conversion of MongoDB query language to SPARQL, 2017 Intelligent Systems and Computer Vision (ISCV), pp. 1-6.
- Zhou, X., Luo, J., and He, T., 2017. Multi-Query Optimization via Common Sub Query Elimination for SPARQL, 10th International Symposium on Computational Intelligence and Design, Hangzhou, pp. 213-218.
- Zhang, X., Li, X., and Zhao, Y., 2014. Knowledge Extraction and Application for Metal Materials Based on DBpedia, 10th International Conference on Semantics, Knowledge and Grids.



# Smart Houses with Internet of Things: Investigation of Security Issues and Solutions

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**Abstract:** The Internet of Things (IoT) is a network in which physical objects are linked to each other or to larger systems. IoT technology is used today in many areas from smart homes to city management. Smart homes are used as a technology that facilitates the daily lives of people, which are the result of adapting the technologies applied in many areas of the industry to the home environment. IoT technology can control all items and objects in your home with your smartphone. IoT can also aim to establish a connection between devices, systems and services beyond machine-machine communication. IoT can accommodate a wide variety of protocols, sites and applications, and the ability of these devices to communicate with each other enables automation in every field. In this study, information is given about 11 protocols (Bluetooth, Wi-Fi, ZigBee, Z-Wave, Thread, Celluar, NFC, Sigfox, Neul, LoRaWAN and 6LoWPAN) used for safe communication in IoT technology. Although IoT is a technology that facilitates our lives, it has brought many problems. In this context, the problems experienced in intelligent home systems based on IoT technologies have been examined and security objectives and security requirements have been discussed as a solution to these problems.

Keywords: Internet of Things, smart home, security, communication protocols, remote access

#### **1. INTRODUCTION**

The IoT term was first introduced by Kevin Aston in 1999 at MIT's AutoID laboratory. The Internet of Things was designated as a global network infrastructure. IoT consists of connecting physical and virtual objects via the Internet (Kim, 2017). IoT is a permeable and ubiquitous network that allows monitoring and control of the physical environment by collecting, observing, processing and analyzing data generated by sensor devices and intelligent objects (Ali et al., 2017).

The areas of use for IoT are quite broad and can be made as varied as possible in daily use. The fact that all devices capable of communicating over the internet can be included in this category means that many different samples have been used in daily life for some time. IoT samples can be seen in almost every area. Both the principle of operation is simple and where it can be used in all sectors can be encountered with a variety of examples. Examples of sectors; Environment, energy, transportation, health, agriculture and daily use can be given. In everyday use, the internet of objects can spread to many different areas. As mentioned earlier, effective uses can be made to facilitate daily life and improve the quality of life. It is possible to improve the quality of life with IoT technology in many different areas, from smart home appliances to security systems.

IoT creates a dynamic global network infrastructure by linking various physical and virtual objects. Thus, IoT aims to improve user comfort, control strategies and to strengthen cooperation between smart devices. IoT technology increases productivity in many sectors and affects user'daily lives from different angles by offering many valuable benefits to users. For the end user, IoT's biggest return is to improve the advanced user experience. Real-time data collection provides a great advantage in profile-based and historical data analysis and processing solutions (Pradeep vd., 2016).

One of the areas where IoT will add the most value to our daily lives is our homes. Smart home automation systems play an important role in bringing together the interconnected benefits in our homes. In the smart home systems, all devices, sensors and services within the house communicate with certain communication protocols. In some systems, these smart devices are interconnected by cables, while others benefit from the advantages of wireless technology. The following parts of the study are as follows: In Chapter 2, IoT communication protocols are described in detail. Chapter 3 provides detailed information on smart home systems. In Chapter 4, security objectives of smart home systems are presented in detail. In chapter 5, security problems and solutions in smart home systems are explained. The last part is the results of the study.

# 2. COMMUNICATION PROTOCOLS USED BY THE INTERNET OF THINGS

One of the indispensable elements for the internet of things is communication protocols. Because the devices can communicate with each other is one of the key points of the internet of the objects and the communication between the devices will be provided by communication protocols. IoT has different communication protocols. Choosing the appropriate communication type according to the requirements of the product and the system allows the process to function efficiently. Depending on the application, the most appropriate protocol is determined by taking into account factors such as data requirements, security, power demand and battery life. The communication protocols used by the Internet of Things are listed below:

• Bluetooth: It continues to be used as an important short-range communication technology in computers and in many electronic products. While it offers many disadvantages due to data size when used in file transfer, it has a significant advantage with its advantages in mobile device connections. It is preferred as a safe, inexpensive way of connecting and transferring data (Samuel, 2016).

• Wi-Fi: It is preferred that the existing infrastructure is very large, it is widely used in homes and offices and it is possible to transfer large files. However, it is used in more complex applications because it causes excessive power consumption for small size devices.

• ZigBee: It is used as a low-power wireless network. It is preferred in consumer and industrial equipment requiring infrequent data exchange and provides a short-range communication.

• Z-Wave: It is a low-power wireless communication protocol. Especially in remote control applications, home automation is preferred in residential and light commercial environments. The biggest advantage is that it is independent and compatible with team work (Samuel, 2016).

• Thread: It is a preferred network protocol in home automation. It is a wireless meshed network structure. Because of low energy, hundreds of devices can be connected to each other or to the cloud.

• Celluar: GSM/3G/4G cellular communication capabilities can be used for internet applications of long distance objects. Especially low cost development kits such as Raspberry Pi and Arduino are frequently preferred in this type of applications.

• NFC (Near Field Communication): NFC is becoming more and more widespread in our lives. By keeping the two devices close to each other, NFC technology enables the devices to exchange data in the magnetic field which is formed. Due to the nature of the technology, even though the communication is one-way, the data transmitted in both directions is limited. Therefore, in general, the data communication between these devices contains the keys of the communication protocols to be connected to each other.

• Sigfox: It is a kind of cellular communication that uses to minimize power consumption. It is used especially in smart home and smart cities, especially in home counters, alarm systems and security systems.

• Neul: Due to its high coverage, low power consumption and low cost, it is often preferred among wireless networks.

• LoRaWAN: It is providing communication for long-range wide area network. It is a protocol with optimum features for smart city and industrial applications. Low cost, mobile and bidirectional communication is the biggest advantage.
• 6LoWPAN: It is used as IPv6 network technology. It is especially used in stock monitoring and health monitoring devices for enterprises engaged in production or sales. It is preferred because it is safe and long lasting.

The communication protocols used in IoT and the properties of these protocols are shown in Table 1. The standards of the communication protocols, frequency bands, nominal range and data rate are shown in this table. In this way, the communication protocols are compared. With this table, the appropriate connection type can be selected according to the requirements of the product and the system and the process can be run efficiently.

| Protocols |  | Characteristics                         |                        |   |
|-----------|--|---|------------------------|---|
|           | Standard                                   | Frequency Band                          | Nominal<br>Range       | Data Rate                                   |
| Bluetooth | Bluetooth 4.2 core specification           | 2.4 GHz (ISM)                           | 50-150m<br>(Smart/Ble) | 1Mbps                                       |
| Wi-Fi     | 802.11n                                    | 2.4 GHz ve 5 GHz                        | 50m                    | Max<br>600Mbps                              |
| ZigBee    | Zigbee 3.0 IEEE802.15.4                    | 2.4 GHz (ISM)                           | 10-100m<br>(Smart/Ble) | 250Kbps                                     |
| Z-Wave    | Z-Wave Alliance ZAD12837 /<br>ITU-T G.9959 | 900 MHz (ISM)                           | 30m<br>(Smart/Ble)     | 9.6/40/100<br>Kbps                          |
| Thread    | Thread (IEEE 802.15.4 ve<br>6LowPAN)       | 2.4 GHz (ISM)                           | N/A                    | N/A   |
| Cellular  | GSM/GPRS/2G/3G/4G                          | 900/1800/1900/2100 MHz                  | GSM max<br>35km        | 4G 3-10<br>Mbps, 3G 600<br>kbps -10<br>Mbps |
| NFC       | ISO/IEC 18000-3                            | 13.56 MHz (ISM)                         | 10cm                   | 100-420 kbps                                |
| Sigfox    | Sigfox                                     | 900 MHz                                 | 3-50km                 | 10-1000 bps                                 |
| Neul      | Neul                                       | 900 MHz (ISM)                           | 10km                   | 100 kbps                                    |
| LoRaWAN   | LoRaWAN                                    | Variable                                | 2-5km, 15km            | 0,3-50 kbps                                 |
| 6LoWPAN   | RFC 6282                                   | Bluetooth Smart 2.4 GHz /<br>ZigBee/ RF | N/A                    | N/A   |

Table 1. Properties of communication protocols

### **3. SMART HOME SYSTEMS**

The availability of IoT can only be achieved with user-oriented applications that users can easily use in their daily lives. A smart home consists of many technologies through home networks to improve the quality of life. Smart home is a place with highly developed automated systems to control and monitor lighting and temperature, household appliances, multimedia devices and security systems and many other functions. IoT plays an important role in smart home construction. Thanks to IoT, almost every object of our daily life in a home can be connected to the internet. IoT allows all these connected objects to be monitored and controlled regardless of time and place (Malche vd., 2017).



### Figure 1. IoT used smart home systems

The smart home systems using IoT is shown in Figure 1. This system is a framework that uses computers, mobile phones and/or smartphones to control basic home appliances wisely and automatically over the internet. The main purpose of this system is to design a smart home automation system using IoT, that is, to convert a traditional house, to a smart home to remotely access and control devices (Govindraj vd., 2017).

The smart home systems consist largely of three main components: Home server, smart home devices and home gateway. First, the home server provides the function of storing, integrating and distributing information collected from home environments. The main gateway then performs a subscriber function or enables the interconnection between the access network and a wired or wireless home network. Finally, smart home devices exchange information between devices and the external internet access function (Han vd., 2015).

Resources used in smart home systems are very important for users. Users should learn and use smart home systems well. If the opposite situation is shown, problems may arise within the system.

Since the user will control the home system via the internet, the internet network must work properly and should not have disconnected connections. The disconnection of the mobile device with the internet means that the home system cannot be controlled. Therefore, the importance of the internet in these systems is very large.

As the number of devices such as lamps, refrigerators and televisions increases, this system becomes more difficult to control. In order to prevent this, the design of the instruments inside the house should be done well. In this way, problems that may occur are prevented.

Building designers, managers and IT professionals must work together to identify and mitigate potential security risks. In this way, the needs are more accurately determined and a more appropriate system is revealed.

### 4. SECURITY GOALS IN SMART HOME SYSTEMS

Most of the devices connected to the IoT are interconnected and always open to attack. A security problem brings with it many challenges. At home or workplaces where many devices are connected, these challenges are greater. For this reason, securing devices connected to the internet is very important for IoT success. In this section, the five most important security objectives for the smart home are addressed. These objectives include:

• Authentication: Confirming the accuracy of a piece of data that an entity has verified. In other words, the communication of the parties or users is a message of the request sent by the author. An appropriate and robust authentication procedure is required for the smart home system. Such an approach is the use of a mutual authentication protocol of the public key with pre-shared keys between a gateway and a new device (Santoso vd., 2015). For example,

when logging into the system, the user may be asked for a password and opening the application when the user enters the correct password.

• Authorization: It can be said that certain rights and obligations specific to electronic communication services are given to users who want to provide electronic communication services or to provide electronic communication network. In these systems, the access rights of each user and the resources of the system must be defined. As an example, the user can view invoices but prevent them from making changes to the invoice.

• Confidentiality: The confidentiality of information, documents and data belonging to the parties performing the transaction from other persons or institutions. It allows only authorized users to access private data in the system.

• Integration: It means integrity and harmony. In these systems, it is defined as a guarantee that the data is maintained in a consistent and accurate manner.

• Availability: Man-made things are easy to use and learn. These systems must also ensure that all services are accessible to a user at all times, and these resources must be protected against all threats.

## 5. SMART HOME SYSTEMS SECURITY ISSUES AND SOLUTIONS

The security of many internet-enabled devices connected to smart homes and organizations is very low. These devices are vulnerable to attacks and can create security issues. Security and privacy should be given more attention when designing IoT devices. New technologies are often misused. In order to prevent this, it makes more sense to solve it before the problem occurs (Jindal vd., 2018). Smart homes connected to the internet also allow malicious attackers to intrude into systems connected to the same network. This weakness is a very important problem that needs to be solved. In this section, problems in smart homes will be explained.

• The smart meter in a smart home is used to collect the user's consumption information. As a result of this information, measurement data is processed and billing information is generated. In this case, security issues such as message changes, customer data leaks may occur.

• Another problem with smart meters is that users try to change the software of the smart meter and reverse the counter to avoid paying the electricity bill. This situation causes many problems.

• The attacker can access the consumer's consumption data via electronic media and use this data to reflect the lifestyle of the user. This issue also causes message change threats. Changes the messages sent to the user. Therefore, undesirable results may occur.

• IoT-connected objects, such as household electrical appliances, door lock, in smart homes, can be used for illegal monitoring. Objects connected to this internet can allow attackers to gain much more information. For example, attackers can connect to a door lock that is connected to the internet and can easily monitor when a person enters his/her home and leaves his/her home.

• IoT enables the creation, storage and sharing of large amounts of data related to a person's habits, preferences and behaviors. In this situation, when attackers reach this data, they can use this data in many places and threaten users.

In order to control and minimize risks in IoT-based smart homes, the following safety requirements must be observed. In this way, problems that may occur are detected in advance and bigger problems are prevented.

• Authentication: Since smart home devices are connected to the internet, software updates between these devices and data exchange between devices are required. A strong authentication mechanism should be established for unauthorized devices and users, and unauthorized users should be prevented from entering the system.

• Monitor the Network: Smart homes connected to a network should also be equipped with a monitoring and intrusion detection tool without monitoring means to protect against enemy attacks. In this way, it is possible to save the network from many attacks (Ali vd., 2017).

• Checking for Updates: The manufacturer's website should be checked at regular intervals to see if there are updates to the software of a device.

• Security Settings: Security settings of devices in smart homes should be reviewed. All passwords must always be changed to be difficult to predict. Remote access to devices must be disabled unless necessary.

• Integrity: Ensures accurate and consistent transmission of information. The data should not change and should not be lost. The message verification code for smartphones plays an important role in ensuring integrity.

• Confidentiality: Enables the privacy of users. The user's private information is stored and only authorized users can access this information. Encryption and key management strategy should be used to ensure confidentiality (Ali vd., 2017).

#### 6. CONCLUSION

Smart homes are defined as homes that can meet the needs of users and offer users a more comfortable and more economical life. Smart homes also aim to increase the standard of living and safety, to save energy and resources. For this reason, IoT technology is often preferred in smart homes. IoT technology has made a quick introduction to our lives with smart homes. With IoT, almost any object in a home can be connected to the Internet and controlled. However, IoT has brought with it many problems. In this study, many problems with IoT have been identified. In order to avoid these problems, the security literature has been examined and security objectives for the smart home have been determined by using these techniques. In this way, it was emphasized that pre-measures should be taken and that such problems should be avoided from the very beginning. In addition, there are many connection protocols in IoT technology and selecting the appropriate connection type ensures that the communication takes place efficiently. In this study, these connection protocols are mentioned.

In addition, the standards, frequency bands, nominal range and data rate of the connection protocols are shown and compared. With this table, the appropriate connection type can be selected according to the requirements of the product and the system and the process can be run efficiently.

#### REFERENCES

- Kim, J. (2017). Analyses of Secure Authentication Scheme for Smart Home System based on Internet on Things, International Conference on Applied System Innovation, 335-336.
- Ali, W., Dustgeer, G., Awais, M. & Shah, M. (2017). IoT based Smart Home: Security Challenges, Security Requirements and Solutions, 23rd International Conference on Automation and Computing.
- Malche, T. & Maheshwary, P. (2017). Internet of Things (IoT) for Building Smart Home System, International Conference on I-SMAC (IoT in Social, Mobile, Analytics and Cloud), 65-70.
- Govindraj, V., Sathiyanarayanan, M. & Abubakar, B., (2017). Customary Homes to Smart Homes using Internet of Things (IoT) and Mobile Application, International Conference On Smart Technology for Smart Nation, 1059-1063.
- Samuel, S. (2016). A Review of Connectivity Challenges in IoT-Smart Home, 3rd MEC International Conference on Big Data and Smart City.
- Pradeep, S., Kousalya, T., Suresh, K. & Edwin, J. (2016), IoT and Its Connectivity Challenges in Smart Home, International Research Journal of Engineering and Technology, 1040-1043.
- Han, J., Jeon, Y., Kim, J. (2015). Security Considerations for Secure and Trustworthy Smart Home System in the IoT Environment, 2015 International Conference on Information and Communication Technology Convergence, 116-1118.
- Jindal, F., Jamar, R. & Churi, P. (2018). Future and Challenges of Internet of Things, International Journal of Computer Science & Information Technology, 13-25.

Santoso, F. & Vun, N. (2015). Securing IoT for Smart Home System, 2015 IEEE International Symposium on Consumer Electronics.



# Territorial Aspects of Ecologization of Recreational Land Use in Urban Agglomerations

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Abstract: This research is devoted to the theoretical principles of recreational territories` zoning on the way to ecologization of urban agglomerations. Ecological factor of territories' modernization in the current conditions of world's development and interests of economic entities, should take into account the priority areas of the economy. In this sense, it is considered appropriate to ecologize recreational lands. According to the author, ecologization is the process of purposeful and consistent implementation of measures for preservation and improvement of the environment in the system of organizational and economic activity of the country in the field of economic activity. It is important to introduce the appropriate process in the development of urban agglomerations. According to research of UN experts, the number of urban agglomerations in the world is calculated by many hundreds and 1.3 billion people or about 50% of the urban population of the world live there. In 30 largest agglomerations with a population of more than 10 million people in the future will be concentrated 478.8 million people, or about 10% of the urban population of the world. It is expected that by 2050, increase the number of urban people will be 3.1 billion people. In the research we have analyzed the scientific approaches to the definition of the concepts of "ecologization" and "urban agglomerations", deficiencies of the urban agglomeration systems. The use of land in the world is characterized by significant disproportions; contradictory structure of the economic complex, high level of pollution of the environment; small part of territories of nature protection, recreation, health, historical and cultural destination. Today most urban agglomerations use fundamentally unstable methods as a result of the existence of of such barriers on the way to ecologization of territories. For proper leveling of anthropogenic impact on the environment and eliminating the above threats increase of recreational territories and their zoning is necessary requirement. We have proposed ecological&economic zoning of recreational territories. It involves a departure from economic activity and focuses on the areas of environmental factor, but also involves greening of economies, taking into account existing economic efficiency, needs of stakeholders in the system of recreational lands. During research of features and scientific principles of zoning of territories, we have formed the principles and predicted effect of the proposed ecological&economic zoning of territories. Given the specificity and complexity of recreational use of land, we have developed proposals for the distribution of recreational areas for ecological&economic zones: zone of seasonal residence, zone of security restriction, green zone, healing-medical zone, cultural-entertaining zone, commercial-economic zone, aesthetic-cognitive zone, service zone. It is envisaged that the green, aesthetic-cognitive and healing-medical zones will serve as an ecological base of territories' development. And the zones of seasonal residence, cultural-entertaining and commercial-economic zones will be the basis for the economic development of the territories. The appropriate distribution will be the basis for determining the ecological status and calculating the ecological stability of the territories. This will serve as a guarantee of ecologization of recreational land use in urban agglomerations.

**Keywords:** Ecologization, recreational land-use, urban agglomerations, ecological&economic zoning, sustinable development



# Reduction of Pollution by Activated Carbon Columns and Activated Carbon Backwash Applications

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**Abstract:** In this study, the wastewater of a factory producing organic peroxide was treated with two serial connected activated carbon columns. Wastewater treatment effluents (equalization, flotation and chemical treatment) have been passed through a series of activated carbon columns. Activated carbon columns were 2 m long and 5 cm in diameter.

As a result of the variety of products and as a result of the different products of the wastewater, the efficiency of activated carbon COD removal was changing. There were dozens of product profiles available, such as methyl ethyl ketone peroxide, butyl peroxybenzoate, benzoyl peroxide, acetyl acetone peroxide. Plant effluent COD ranged from 10,000 mg/l to 25,000 mg/l. Activated carbon influent COD values vary depending on physical and chemical treatment efficiency. In this study, if the inluent COD value was 15980 mg/l, the 2nd column activated carbon effluents were 669, 903,1152,5301 and 8217 mg/l at the end of one, two, three, five and six hours. Activated carbon was regenerated (backwash) with hot water and tap water. Regeneration due to chemical adsorption of wastewater containing high COD was limited. The adsorption method and the backwash were not suitable for this type of wastewater.

Keywords: Activated carbon, Organic Peroxide, COD, Regeneration.

## **1. INTRODUCTION**

In this study, the characterization and treatment of wastewaters of a chemical industry producing organic peroxide, initiator, accelerator and dye dryers were studied. The wastewater from the treatment plant balancing tank was used in the study. Samples used in the study were wastewaters from the equalization tank of a chemical industry producing organic peroxide, initiator, accelerator and dye dryers and from 6 different process sections. The H2O2 ratio in the samples was very high. H2O2 was reduced using an anti-peroxide solution (Catalase) in case of high H2O2 before each COD test.

It was observed that the pollutant concentrations in the wastewater formed at this plant have a very high and very variable structure. It was thought that the main reason for this was the variability of the rate of chemical used in the process and the amount of production. Activated carbon was generally activated in the environment of CO2, CO, O2, H2O vapor, air or other selected gases (N2) at temperatures between 300-1100oC. Due to the impure structure of the raw materials used in the production of commercial carbons and the concentration and temperature gradients developed in the carbon deposits during activation, very heterogeneous surfaces may ocur (Küçükgül, 2004).

The large surface area, microporous structure and increased surface reactivity associated with granular activated carbon (GAC) [Horning et al.,2008; Mumford et al., 2014) was one of the strongest adsorbent materials used in purification processes. The renewal of the GAC aims to restore the original adsorptive and increase the capacity of the GAC by removing the pollutants accumulated on the carbon surface and causing as little damage to the carbon as possible [Van Vliet,1991]. Various regenerative techniques were handled extensively and wase now used in industrial applications; thermal, chemical and microbial methods were included. However, these methods were met by limitations such as high energy consumption and carbon wear, pore clogging, and often the need for secondary processes and slow regeneration rates [Rebecca, 2018].

In this study, it has been tried to reduce the pollution of the effluents of the treatment plant by adsorption. In the second step, backwashing of saturated active carbon was studied.

### 2. MATERIALS AND METHODS

Activated carbon as adsorbent has been preferred. Experiments were carried out in downstream configuration. With a control valve connected to the bottom of the filter base in each column, a constant water level can be maintained in the GAC filter. The pilot scale reactor shown in Figure 1 was 2 m long and it was made of plexiglass with an inner diameter of 5 cm. The column was filled between two supporting layers with an activated carbon of 5 mm particle size (Figure 1). The experiments consist of two activated carbon columns connected in series. Treatment plant effluent wastewater was stored in 30-liter tank and fed to activated carbon columns. The samples were taken from the first and second column exits in certain time periods and COD measurements were made. For activated carbon regeneration, tap water and hot water plus air methods were applied. In certain periods of time, the regeneration of the system has been checked by measuring the COD. COD, pH and peroxide measurements were made in the system. Measurements were made according to Standard Methods.



Figure 1. Adsorption (Activated Carbon) Columns

## 3. RESULTS AND DISCUSSION

In this study, wastewater influent samples were taken from the equalization tank at different times in order to determine the wastewater characterization. As a result of the wastewater characterization from the balancing tank where the effluent waters were stored for 24 hours, the COD value was found to be 23725 mg/l. The amount of oil and gress was 899 mg/l, total nitrogen 6.2 mg/l and chloride concentration 9042 mg/l.

| it i. Equalization Fank wastewater Characteristic |      |         |  |  |  |  |  |  |
|---|------|---------|--|--|--|--|--|--|
| Parameter   | Unit | Results |  |  |  |  |  |  |
| COD   | mg/L | 23725   |  |  |  |  |  |  |
| Oil and gress                                     | mg/L | 899     |  |  |  |  |  |  |
| Total Kjeldahl Nitrogen(TKN)                      | mg/L | 6.2     |  |  |  |  |  |  |
| Free Chlorine                                     | mg/L | < 0.05  |  |  |  |  |  |  |
| Chloride  | mg/L | 9042    |  |  |  |  |  |  |

Table 1. Equalization Tank Wastewater Characteristic

The wastewater stored in the equalization tank was passed through the flotation unit and then through the chemical treatment unit. Chemical treatment effluent COD:19825 mg/l, oil and gress 580.6 mg/l, total kjeldahl nitrogen 5.2 mg/l, chloride concentration 9032 mg/l was found. Chemical treatment efficiency was low due to dissolved substances.

Table 2. Chemical Treatment Effluent Wastewater Characterization

| Parameter                    | Unit | Results |
|------------------------------|------|---------|
| COD                          | mg/L | 19825   |
| Oil and gress                | mg/L | 580.6   |
| Total Kjeldahl Nitrogen(TKN) | mg/L | 5.2     |
| Free Chlorine                | mg/L | < 0.05  |
| Chloride                     | mg/L | 9032    |

pH and COD were measured in the samples taken from the plants. The measurement results were given in Table1. The current treatment (flotation + coagulation + flocculation + precipitation) system does not provide discharge standards. Therefore, adsorption method was applied after chemical treatment. Granular activated carbon was preferred as adsorbent. As seen from Table 1, samples were taken from active carbon columns 1 and 2 in one-hour interval in three different experiments. At the end of one hour at the first column exit, the COD value decreases from 15980 to 4866 mg/l. At the end of the second and sixth hours, COD values increased to 7260 and 13850, respectively. When the same wastewater was passed through the second filter, the COD value decreased to 669 mg / l after 1 hour. After 4, 5 and 6 hours, the effluent COD increased to 2424,5301 and 8217 mg, respectively. Depending on the product profile, the wastewater characteristics change in the existing plant. Therefore, samples were taken from the equalization tank on different dates. In the second series of experiments, at the end of four hours, the first and second filter effluent COD decreased from 10000 mg / l to 1295 and 875 mg / l respectively. After 14 hours, 9135 and 2541 mg/l COD were measured at the first and second filter effluents, respectively. The different performance of activated carbon in the first and second series experiments was related to the wastewater characteristics. More than 30 different chemical products were produced in the facility. In the third series, the first and second filter effluent COD values were 1316 and 1032 mg /l, respectively. After 12 hours, the first filter effluent was 12392 mg/l and the second filter effluent was 8760 mg/l.

| COD    |   | 1.hou | 2.hou | 3.hou | 4.hour | 5.hour | 6.hou | 7.hou | 10.hou | 12.hou | 14.hou |
|--------|---|-------|-------|-------|--------|--------|-------|-------|--------|--------|--------|
| (mg/l) |   | r     | r     | r     |        |        | r     | r     | r      | r      | r      |
| İnflue |   |       |       |       |        |        |       |       |        |        |        |
| nt     |   |       |       |       |        |        |       |       |        |        |        |
|        | 1 | 4866  | 7260  | 7780  | 9850   | 12680  | 1385  |       |        |        |        |
| 15980  | _ |       |       |       |        |        | 0     |       |        |        |        |
|        | 2 | 669   | 903   | 1152  | 2424   | 5301   | 8217  | 1254  |        |        |        |
|        |   |       |       |       |        |        |       | 5     |        |        |        |
|        | 1 |       |       |       | 1295   |        | 1435  |       | 2583   | 4312   | 9135   |
| 10000  | 2 | 798   |       |       | 875    |        | 889   |       | 1568   | 1596   | 2541   |
|        | 1 |       | 1316  | 1520  | 2976   | 5296   | 6828  | 9156  | 10968  |        | 12392  |
| 13000  | 2 |       | 1032  |       | 1020   | 1156   | 1212  |       | 4040   | 6745   | 8760   |

**Table 3.** Activated carbon effluent COD results

It was examined whether active carbon was regenerated after adsorption by passing 5.0 liters of wastewater from 800 g activated carbon. Two different influent wastewater were studied. After each passage the filter was washed backwards. The filters were washed with tap water and 75oC of hot water. There was no difference in the efficiency of the filters after backwashing. As seen from Table 4, it was seen that the filter was not cleaned by simple washing. The influent COD was 12120 mg/l. After each passage the filter was washed backwards. The filter effluent COD after the 1st pass was 1995 mg/l. When the filter was washed back and an additional one liter of wastewater was passed, filter effluent COD value 7980 mg/l was found. The filter effluentt COD values were measured as 7585, 9420,11030 mg/l respectively as a result of washing the filter backwards after each pass and using 5.0 liter wastewater at each pass. When the influent COD was 14920 mg/l, it was seen that the effluent COD values increased more.

 Table 4. Change of filter effluent COD depending on the amount of wastewater.

| COD=12120 | 1.0   | 1.0   | 1.0   | 1.0   | 1.0   | Total=5lt  |
|-----------|-------|-------|-------|-------|-------|------------|
| mg/l      | Liter | Liter | Liter | Liter | Liter | wastewater |
| 1.pass    | 1995  | 3150  | 5625  | 7265  | 7585  |            |
| 2.pass    | 4445  | 5770  | 7640  | 8650  | 9420  |            |
| 3.pass    | 7980  | 10115 | 8320  | 10760 | 11030 |            |

| COD=14920 |      |       |      |       |       |  |
|-----------|------|-------|------|-------|-------|--|
| mg/l      |      |       |      |       |       |  |
| 1.pass    | 8060 | 9590  | 9230 | 10460 | 10790 |  |
| 2.pass    | 8900 | 11000 |      | 10650 | 10290 |  |
| 3.pass    | 9080 | 11220 | 9770 | 12360 | 12380 |  |

Influent COD=12120 and 14920 mg/l \* The filter was washed back after each filtration pass 150 g of activated carbon was placed in 500 ml beakers and stirred for 30 minutes at 190 rpm. It was mixed with tap water in mechanical mixer. As a result of this process, the effluent COD was 2240 mg/l in the backwashed filtration water. As a result of washing with 10% sodium hydroxide solution, the backwash water COD was 3000 mg/l. Backwash water COD value was low, activated carbon was not cleaned mechanically, chemical adsorption has been found in the system.

Table 5. Backwashing of the saturated filter using mechanical stirrer

| COD=11280     | Backwash  |  |
|---------------|-----------|--|
| mg/l          | COD       |  |
| Backwash with | 2240 mg/l |  |
| tap water     |           |  |
| Backwashing   | 3000 mg/l |  |
| with 10%      |           |  |
| NaOH          |           |  |

#### REFERENCES

- Küçükgül, E.Y., (2004). Ticari aktif karbon üretimi ve özelliklerin belirlenmesi. DEÜ Mühendislik Fakültesi Fen ve Mühendislik Dergisi 6(3): 41-56.
- [2] Horning, G., Northcott, K., Snape, I., Stevens, G.W., (2008). Assessment of sorbent materials for treatment of hydrocarbon contaminated ground water in cold regions, Cold Reg. Sci. Technol 53:83–91.
- [3] Mumford, K.A., Rayner, J.L., Snape, I., Stevens, G.W., (2014). Hydraulic performance of a permeable reactive barrier at casey station, Antarctica, Chemosphere 117: 223–231.
- [4] Van Vliet, B.M., (1991). The regeneration of activated carbon, J. South. Afr. Inst. Min. Metall. 91: 159–167.
- [5] Rebecca, V. McQuillan., Geoffrey, W., Stevens, Kathryn, A. Mumford., (2018). The electrochemical regeneration of granular activated carbons: A review. Journal of Hazardous Materials 355: 34–49.



## Examination of Traffic Education Parks in the Scope of Landscape Architecture: Bartın Case

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**Abstract:** Nowadays, the number of traffic accidents increases due to rapid urbanization and population density. According to the Statistics of Road Traffic Accidents received from Turkey Statistical Institute (TUIK) in 2008, 1.182.491 traffic accident (of which 104.212 were mortal and injury) occurred. This number increased to 1.202.716 (of which 182.619 were mortal and injury) in 2017. Accordingly, the importance of transportation planning in our daily life is increasing. The main reason for the traffic accidents is the human element consisting of drivers, pedestrians and passengers. In ensuring traffic safety, the awareness of traffic safety in individuals is effective. This reveals the necessity of planned and scheduled training to prevent accidents.

It is known that education is effective in increasing the quality of life of individuals of all ages who make up the society in which the concept of lifelong learning is now discussed and accepted. However, the education, which will start at a young age, may contribute to a stronger foundation of society. In this direction, traffic education parks are emerging as a tool for creating a traffic culture to be sustained through fun learning. Traffic education parks offer a realistic learning environment and combine elements of the traffic system together. These elements are; road, sidewalk, street, intersection, road lines, pedestrian crossing, lighted traffic devices, traffic signs, vehicles, pedestrians and green areas. Traffic education parks planned for use in different age groups are part of the open-green area system.

In this study; it is aimed to determine landscape planning and design criteria by examining the traffic education parks which should be arranged within the scope of the Regulation on Children Traffic Education Parks dated 24.10.1998 numbered 23503 in terms of landscape architecture. For this purpose, the study was carried out in four stages as data collection, analysis, evaluation, development of results and recommendations. During the data collection stage, domestic and foreign literature on the subject was reviewed; data on regulations, plans, projects and reports, and examples of traffic education parks in Turkey and abroad were obtained. All these data were analysed and evaluated, and the ideas of traffic education park projects, which is planned to be built in Bartin province, was discussed in the last stage of the study. Recommendations on land usage, plant design and urban equipments in traffic parks are presented based on the headings in the Child Traffic Education Parks Regulation.

The regulation does not contain any statements regarding the landscape design of the traffic education parks that should be included in the open and green area system. In order to prevent the negative consequences of incomplete/wrong structural and planting design in these areas where users are children, city and region planners, traffic engineers and landscape architects should work together. Therefore, landscape architects are to be in the planning and design process as well as their duties and responsibilities with changes to be made in legal and managerial dimensions.

Keywords: Traffic education parks, traffic safety, planting design, landscape design, Bartın.

### **1. INTRODUCTION**

Today, 55% of the world's population lives in cities. This rate is expected to rise to 68% by 2050 (UN, 2018). The population living in the cities in Turkey is indicated to be 92.3% as of 2018 (TSI, 2019). It is possible to say that urbanization is accelerated by considering these data. There is an increase in the number of traffic accidents with the effect of rapid urbanization and increasing population density. Transportation has become one of indispensable aspects of our era and our daily life as a result of rapid urbanization. Although air, sea and railway transportation facilities are developed in Turkey, transportation throughout the country is mostly provided by highways (Bolat et al., 2017). Since the road traffic accident statistics are established in decennial periods by TSI and they are the most current data available,

TSI 2008-2017 data were used in the study. Numbers of registered vehicles, accidents, the dead and injured for these years are given in Table 1.

|      |                                      | 0                         | · 1   |  | 5     |                         | · ,                       | ,                               |
|------|--------------------------------------|---------------------------|---|--|-------|-------------------------|---------------------------|---------------------------------|
|      | Number of                            | Total                     | Number of                                     | Number of  | Numb  | er of persons           | s killed                  | _                               |
| Year | vehicles<br>registered<br>to traffic | number<br>of<br>accidents | accidensts<br>involving<br>death or<br>injury | accidensts<br>involving<br>material<br>loss only | Total | At<br>accident<br>scene | Accident<br>follow-<br>up | Number<br>of persons<br>injured |
| 2008 | 13.765.395                           | 950.120                   | 104.212                                       | 845.908  | 4.236 | 4.236                   | -                         | 184.468                         |
| 2009 | 14.316.700                           | 1.053.346                 | 111.121                                       | 942.225  | 4.324 | 4.324                   | -                         | 201.380                         |
| 2010 | 15.095.603                           | 1.106.201                 | 116.804                                       | 989.397  | 4.045 | 4.045                   | -                         | 211.496                         |
| 2011 | 16.089.528                           | 1.228.928                 | 131.845                                       | 1.097.083  | 3.835 | 3.835                   | -                         | 238.074                         |
| 2012 | 17.033.413                           | 1.296.634                 | 153.552                                       | 1.143.082  | 3.750 | 3.750                   | -                         | 268.079                         |
| 2013 | 17.939.447                           | 1.207.354                 | 161.306                                       | 1.046.048  | 3.685 | 3.685                   | -                         | 274.829                         |
| 2014 | 18.828.721                           | 1.199.010                 | 168.512                                       | 1.030.498  | 3.524 | 3.524                   | -                         | 285.059                         |
| 2015 | 19.994.472                           | 1.313.359                 | 183.011                                       | 1.130.348  | 7.530 | 3.831                   | 3.699                     | 304.421                         |
| 2016 | 21.090.424                           | 1.182.491                 | 185.128                                       | 997.363  | 7.300 | 3.493                   | 3.807                     | 303.812                         |
| 2017 | 22.218.945                           | 1.202.716                 | 182.669                                       | 1.020.047  | 7.427 | 3.534                   | 3.893                     | 300.383                         |

| Table 1. Number of vehicles registered | d, accident, persons | s killed and injured | , 2008-2017 ( | TSI, 2018) |
|--|----------------------|----------------------|---------------|------------|
|--|----------------------|----------------------|---------------|------------|

According to Arslan (2002); preventing traffic accidents is possible by ensuring traffic safety. While traffic safety can be ensured by adhering to traffic rules, adapting to these rules depends on the presence of traffic culture. Planned and scheduled training activities are needed for the creation of traffic culture (Güner and Genç, 2012; Bolat et al., 2017). According to Böcher (1995) and E.U. Report (2005), traffic safety training is based on three main goals. These goals include knowledge of traffic rules and situations, development of skills through training and experience, risk awareness, strengthening and/or changing attitudes on personal security and security of other users (Assailly, 2017). The human factor is the most dominant among the various causes of accidents due to human, vehicle and environmental factors (Acar, 1992; Gökdağ and Atalay, 2015; Assailly, 2017). While human behaviour is effective in 90% of accidents, the environmental factor is effective in 30%, and vehicle factor is effective in 10% of all accidents (Assailly, 2017). According to Cengiz (2013), raising conscious and responsible individuals is the most effective solution to traffic problems (Bolat et al., 2017). According to Sönmez (1991), traffic training refers to raising awareness among human element involved in traffic in training institutions or outside traffic and in terms of traffic accidents and to provide necessary knowledge and skills to prevent problem of traffic. According to another definition, traffic training includes all works required to be done to make traffic rules set by the legal conditions people's natural behavior and to ensure the safety of people (Hatipoğlu et al., 2012; Erturk, 2016).

In our country, education starts with preschool training. Training should not be limited to buildings only. Open areas of educational institutions are as important as buildings. The presence of spaces that are integrated with the garden, with the ability to meet and develop physical, spiritual and mental needs of children, affects the success of education in a positive way (Karakaya and Kiper, 2013). Children's Traffic Education Parks (CTEP) are also one of the socio-cultural areas supportive of physical development. However, it is regarded as a tool to ensure that training is permanent through fun learning and continuity in outdoors. CTEP is created as a simulated road environment where children can learn to use routes safely and consciously without exposing themselves to life-threatening traffic hazards (Fourie, 2005). This environment offers the opportunity to easily implement information that is difficult and dangerous to consolidate in the real traffic environment.

The traffic education parks, which are part of the urban open-green area system, were established in 1998 with by regulating "CTEP Regulation" and published in the Official Gazette no.23503. In the regulation, these parks are defined as stationary or mobile children's traffic education park, which is built and to be built with the aim of teaching preschool and school age children traffic rules and making a habit out of these trainings". The purpose of arrangement of these parks is indicated as "giving the opportunity to preschool children and primary and secondary education students to practice theoretical traffic information learned in the lessons, and helping to create traffic culture and minimize traffic accidents by teaching behaviors of performing traffic rules by means of creating real life traffic environment in these parks".

In Turkey, there are numerous CTEPs affiliated to Directorates of Police, Municipal and Provincial Education Directorates. In Figure 1, examples of children's traffic education parks applied in different provinces of Turkey are given.



**Figure 1.** Examples of TEP in Turkey: a) Bağcılar TEP/İstanbul b) Diyarbakır TEP c) Sakarya TEP d) Tarsus TEP/Mersin e) Kocaeli TEP f) Malatya TEP g) Küçükçekmece TEP/İstanbul (URL-1, URL-2, URL-3, URL-4, URL-5, URL-6, URL-7, 2019).

There are many CTEPs in different countries abroad. One of them is Pyongyang Traffic Safety Education Park in North Korea (Figure 2). This park has an application area that helps consolidate information about traffic arrangements through practice. The miniatures of streets, buildings and facilities in the city with roads, tunnels, pedestrian bridges, railway observation poles for vehicles and bicycles provide a variety of traffic conditions in this area. Children can use electric vehicles and bicycles to explore the theoretical information they learn in lessons in a real way by using roads. In the facility, there are classes for teachers to provide theoretical information related to traffic courses, and accidents based on violation of highway safety rules are shown in the simulation room. It is possible to reach information about traffic regulations, laws, tools and other relevant information in the e-library in the facility.



Figure 2. Pyongyang Traffic Safety Education Park for Children in Korea (URL-8, 2019).



Figure 3. Fenyves TEP in Budapest (URL-9, 2019).

## 2. MATERIALS AND METHODS

Bartin CTEP with 2 different landscape design idea projects that have not yet been applied constitutes main material of the study. Previously conducted studies regulations, plans and reports related to the field and the subject are materials that help to complete the study. The area selected for Bartin CTEP, which is planned to be applied, is located on the D10 Highway which provides transportation to Karabük-Bartin-Istanbul and covers an area of 3382 m<sup>2</sup>. The position of the working area on the country and province is given in Figure 4. The aerial photograph of the area is given in Figure 5.



Figure 4. Location of the selected area for Bartin CTEP on country and province (Google Earth, 2019).



Figure 5. Aerial photo of the working area (URL-10, 2019).

This study was conducted in 4 phases including data collection, analysis, evaluation, results and preparing proposals. In the data gathering phase, the literature on the subject was scanned with the keywords such as CTEP, traffic security and urban open-green field. Regulations, plans, projects and reports, and data related to local and foreign CTEP samples were obtained. Traffic education park idea projects planned to be done in Bartin province based on criteria belonging to "Land selection, Park Project and Additional Facilities" discussed in "Properties and Equipment of Children Traffic Education Parks" in the regulation were analyzed. In the final stage of the study, recommendations were given by making evaluations of the field usage, planting design and reinforcement elements in idea projects in accordance with the principles of landscape architecture.

### 3. RESULTS AND DISCUSSION

In the third part of the CTEP regulation, the following statement is included: "The following provisions apply for selection of the location where children's traffic training parks shall be established, the establishment of the traffic signs, devices and signs in place, and the construction of additional facilities". In this section, Properties and Equipment of Children's Traffic Education Parks, properties of these parks are examined under 3 main headings. These include plot selection, park project and additional facilities. The first of these main titles, the plot selection is based on 4 criteria. Accordingly, the selected plot for the CTEP should be suitable for use by as many schools as possible, it should be convenient for transportation, where children can be used at any time from public transport in their departure and arrival. Plot should be available for services such as water, electricity, sewage, and should be at least 4000 m<sup>2</sup>.

Accessibility is defined as the distance taken within 10-15 minutes, which is an ideal walking time, by O'Neill et al. (1992), Hsiao et al. (1997) and Phillips and Edwards (2002) in their studies. The ideal walking distance is regarded as 400 m (Yenice, 2013). Bartin CTEP was evaluated in terms of plot selection criteria by creating a circular area with a diameter of 400 m from the area boundary (Figure 6). Accordingly, while there is a kindergarten at a distance of 50 m, there is a secondary school and a private rehabilitation center 200 m away from the north of the area border, and a student residence in the southeast, 200 m away from the area border. It was determined that only 1 school were found within walking distance. Therefore, it was observed that the field did not meet the statement "it shall be suitable for use by as many schools as possible" in the regulation.



Figure 6. Accessibility of the working area (Google Earth, 2019).

It is located on Karabük-Bartın-Istanbul highway D10 surrounded by residences. The area is located on the route with important links to the city transportation, bus stops, pedestrian crossings and pedestrian overpasses (Figure 7). In this context, it is possible to say that the area is accessible and fulfills the requirements of the article "Shall be in places that can be used at all times in public transport, suitable for transportation, for children's departures and their arrival" of the regulation.



Figure 7. Working area route (Google Earth, 2019).

The area is located in a region located in Kemerköprü Neighborhood 151. Cad., where Bartın city center is spreading and urban development is felt intensely in the last 10 years. A regular infrastructure system has also been established with the emerging urban texture. In this context, the field fulfills the requirements of the item "must be in the condition that services such as water, electricity, sewage, etc." of the regulation. The land size of Bartın Children's Traffic Education Park is 3382 m<sup>2</sup> (Figure 8). This feature of the field was determined to partially meet the phrase "must be at least 4000 m<sup>2</sup>" in the regulation.

The second main title of the regulation is the Park Project. Under this title, it was indicated that the parking plot should be rectangular. Furthermore, it was mentioned that traffic signs, illuminated devices, road lines, etc. must be maintained in accordance with the projects in children's traffic education parks. Accordingly, when the idea projects were examined, it was observed that the terrain shape was in accordance with the expression specified in the Regulation (Figure 8) and that the projects contain mandatory equipment to be kept in the regulation.



Figure 8. Limit and size of working area (Google Earth, 2019).

The third parent header is the Additional Facilities. Accordingly, outside of the training circuit in children's traffic education parks, training vehicles and equipment in the park must be protected from external factors and there is a sufficiently large warehouse for the vehicles in question. In addition, among the mandatory uses as specified in the regulation, there is a gradual constructed tribune where the students visiting the park will follow hands-on training, and there is an inspection tower where a supervisor can look out for participants of hands-on training, guide them and warn them if required.

There should be sufficient number of WCs in the park, and it was indicated in the regulation. When examining the ideas projects prepared for Bartin, all these uses and equipments were found (Figure 9) but no supervision towers were found.



Figure 9. Usage areas and equipments in Bartin CTEP idea projects.

2 different design project ideas for Children's Traffic Education Park, which will be built by Bartin Municipality, were produced but implementation of projects has not yet begun. These projects have been announced to the public through one three-dimensional (3D) visual media. Project proposals were evaluated under the headings of track/circulation, parking lots, indoor/outdoor buildings, seating areas and equipments. The plant design, which is an important element in traffic education parks, was been evaluated because there were no data in the project samples. The evaluations on the first project proposal were shown on the images given in Figure 10.



Figure 10. The first Project idea for Bartin CTEP.

**Track/Circulation:** Circulating system is constructed in 2 ways. The first line surrounds the entire area. The car passes are not interrupted except for the parking entrances in question. The track is separated from other circulatory lines using light-colored concrete material on the floor. Along this line, there is a possibility of monitoring the entire track from its surroundings and accessing the desired point in a short time. Pedestrian road expands into the entrance, the administrative building, the warehouse, the parking lot and the living areas and unites with these spaces. The second line is the cycle path that continues adjacent to the parallel of the pedestrian path. The bicycle path is also part of the course, which is designed with a blue background color in accordance with the bicycle paths regulation.

**Car Parks:** Parking lots designed within the area seem inadequate, but it threatens pedestrian safety because it circulates around the course.

**Indoor Spaces/Buildings:** In the proposal project, there is an administrative building located in the south of the area and a warehouse in the north where the appliances are maintained. The administrative building where the entrance control and consultation section are located near the area entrance will provide convenience for users. The fact that the warehouse is positioned in the background of the area will reduce the reflection of view resulted from work and material density. The buildings are suitable aesthetically since they are designed in small scale and color harmony is watched at this stage.

**Seating Areas:** In landscape design projects, seating areas are considered at multiple points of the area in accordance with different user requests and climatic conditions. The project features a seating area designed as a wooden half-circle pergola. The seating units are also listed as semi-circular in harmony with the space. Although it is favorable for the use of staff to be positioned very close to the administrative building of the pergolas, it is thought that other users may negatively affect their preferences. In addition, the two covered spaces are independent of each other but very close to the design with a negative effect. Pergolas that are not supported by the design of the planning are not available in four seasons, given the climatic conditions of Bartın province. The selected equipment and materials should have long-lasting choices, taking into consideration the climatic conditions. In addition, there is a 4-grade seating unit in the amplifier/tripod for monitoring for 48 people. The tribune, which is an important element in traffic education parks, is designed using materials that are suitable for climatic conditions.

**Equipments:** The principle of harmony between materials in the selection of equipments has been ignored. Different materials and living units designed in different colors weaken the aesthetic perception.

The evaluations on the second project proposal were shown on the visual given in Figure 11.



Figure 11. The second Project idea for Bartin CTEP.

**Track/Circulation:** Outside the park, there is pedestrian circulation only in the section reaching the entrance. A section of the pedestrian road that provides access to the park entrance, administrative buildings and trails is expanded to be used as a seating area. There are no roaming networks surrounding the course or a separate bicycle path. In this way, it can be said that the course is isolated from distracting elements from the perspective of the children who receive training. However, traffic education park remains inadequate since there were no routes for bicycles which have begun to spread in traffic. From the perspective of upholstery; the granite cube stone used in the pedestrian entrance is resistant to climatic conditions and is environmentally compatible. Its surface is noticeably indented-protruding, it can reduce the risk of slipping, but may create difficulty in walking.

**Car Parks:** Although parking lots designed outside the area (7 pcs) seem inadequate, they are positioned at the right spot for pedestrian safety.

**Indoor Spaces/Buildings:** In the proposal project, there is an administrative building in the entrance section south of the area. The warehouse where the appliances are maintained is also resolved within the same building. It is thought to be the right choice for the reduction of structures.

**Seating Areas:** There is one outdoor seating area in the entrance of the park. Designing seating areas at multiple points of the field in accordance with different user requests and climatic conditions is important for user satisfaction. In order to track the course in the project, there are no tribunes/amphitheaters which are mandatory in regulation and is one of the important elements of traffic education parks.

**Equipments:** The principle of harmony between materials in the selection of equipments has been ignored. Different materials and living units designed in different colors weaken the aesthetic perception.

### 4. CONCLUSION

CTEP provides a safe environment where information about traffic education can be taught to children. By teaching different skills in a specific order, it allows these skills to be tested and persisted through the application repeatedly. There are no statements in the regulation regarding the landscape design of "Children's Traffic Education Parks" that enable the learning through application of information that is difficult and dangerous to consolidate in the real traffic environment and that should be considered as part of the open-green field system.

The usage areas, sizes, relations between each other, the width of the track and the vehicle-pedestrian circulation are required to be established in the traffic education parks. In this context, it is important to produce holistic designs that are compatible with the environment. Given these considerations, the comprehensive proposals for the removal of the deficiencies of children's traffic parks are presented below in the examples of the CTEP idea project, which is considered to be built in Bartun.

### Suggestions towards planning:

• In areas where child users are expected to be dense, pedestrian circulation must be constructed independently of the vehicle entries and circulations. For this reason, pedestrian circulation in children's traffic education parks should not be divided by vehicle transitions.

- The circulation specified in the course must be created in accordance with the traffic system of our country. Transportation planners/traffic engineers and landscape architects should work together in this regard.
- There are no standard measurements of the width of the vehicle and pedestrian roads in the regulation. These parks are artificial environments where the real traffic environment is represented. Therefore, the dimensions of roads that provide circulation in the park should be determined proportionally with the current highway connections.
- Bicycle routes indicated in place of course and route must be planned to circulate the entire area without interruption.

### **Suggestions Towards Design:**

- In order to ensure the safety of children, parking spaces must be designed to be completely outside the area, complying with parking standards and supported by the design of plants.
- Although the number and direction of the vehicle paths differ according to the design, it is necessary to design these paths in a way to allow passage of at least two battery operated vehicles used in the child traffic education parks.
- Pedestrian roads to be designed on the course should not be divided by vehicle traffic. It is recommended that the pedestrian roads be designed at least 1.2 m, which allows two people to walk side by side.
- Bicycle paths must be arranged according to the principles outlined in the bicycle paths regulation.
- The exterior design of the buildings that are constructed in the area should contain the exterior façade and roofing materials that are compatible with the climatic conditions and reflect the traditional urban texture. In areas with rainy climatic conditions, a cradle or crushing roof should be preferred instead of a flat roof to prevent the accumulation of rainfall load.
- In designed process for children's use in the design process of Children's Traffic Education Parks, security of the target user audience should be prioritized. Plants to be selected in the design of planting should be chosen among stringing and non-toxic species that will not cause a destructive effect with their leaves, branches and bodies (in terms of physiological properties). Species with dense leaves or fruit shed and frequent pruning should be avoided by considering maintenance difficulties. The provision of ecological demands of selected species is vital for plants, in this context, it should be placed on plant species that are resistant to local climatic conditions. The general maintenance and pruning of the plants included in the design should be performed periodically.
- The choice of equipments to be ergonomically, disabled-friendly, compatible with climate conditions and traditional texture, and recyclable, will improve the impact of projects as it will also fulfill the principle of sustainability in the design. It is important to regularly maintain the equipment (seating and lighting units, illuminated traffic devices, traffic signs, vehicles, road lines) and renew it if necessary.

### Suggestions towards management:

- In order to create a healthy and safe traffic environment, as stated in the CTEP regulation, traffic instructors in the park must be present at times when the parks are open. For the prevention of possible accidents, at least one of the medical personnel needs to be on the task.
- In the parks, the personnel assigned by the administrative unit (police directorates, mayors and provincial national education directorates) where the traffic education park is affiliated must be available in terms of controlled use of battery-operated cars. It is recommended that these individuals be competent in terms of operations in the park and trained on this subject.
- In order to transfer the necessary theoretical information within the scope of applied traffic educations, a class environment of at least 30 people must be created. Thus, the environment will be created for the applications to be held under the Traffic Safety Courses in the education curriculum of Turkey. This should be considered a high-scale management decision.

Consequently, this study has allowed the evaluation of these projects, which are still in the idea stage, in terms of the profession discipline of landscape architecture. The fact that the Children's Traffic Education Parks are part of the urban open and green area system requires that these areas be analyzed with the urban planning and design principles in mind. The profession that should have an active role in planning and design of these areas is landscape architecture. Location selection during the planning of traffic education parks, preparation of landscape design project, spatial organization of space, material selection, urban reinforcement design, plant design, type selection, irrigation and maintenance are all related to expertise area of landscape architects.

#### REFERENCES

- Acar İ H (1992). Kent içi Trafik Sorunlarının Hafifletilmesinde Güncel Yöntemler ve İstanbul'un Durumu [Current Methods in Alleviating Urban Traffic Problems and the Situation of Istanbul] Kentiçi 2. Ulaşım Kongresi, 16 Aralık, İstanbul, s.468-480.
- Assailly J P (2017). Road safety education: What works? Patient education and counseling, 100: 24-29.
- Bolat Y, Özbek E & Kaygusuz S (2017). Çocuk Trafik Eğitim Parkının 4. Sınıf Trafik Güvenliği Dersinde Öğrenci Başarısı Üzerindeki Etkisi [The Effect of Park of Child Traffic Training On The Success of Students at Fourth-Grade Traffic Safety Course]. Elektronik Sosyal Bilimler Dergisi, 16(63): 1155-1165.
- Ertürk R (2016). İlkokul Öğrencilerinin Uygulamalı Trafik Eğitimlerine Yönelik Algıları [Applied For Primary Education Students Perception Of Traffic]. Akademik Sosyal Araştırmalar Dergisi, 34: 546-553.
- Fourie E, Coetzee J L & Raven A (2005). Road Safety Education for Children Tthe Use of Junior Training Centres. Proceedings of the 24th Southern African Transport Conference, 11–13 July, Pretoria, South Africa, pp.567-575.
- Google Earth (2019). https://earth.google.com/web/ (29.03.2019).
- Gökdağ M & Atalay A (2015). Trafik Eğitiminin Trafik Kazaları Üzerindeki Etkisi [Traffic Education And Effective On The Traffic Accidents]. EÜFBED- Fen Bilimleri Enstitüsü Dergisi, 8(2): 272-283.
- Güner F & Genç Z S (2012). İlköğretim Öğrencilerinin Trafik Güvenliği Bağlamında Kitle İletişim Araçlarına İlişkin Görüşlerinin İncelenmesi (Çanakkale İli Örneği) [The Examination Of Primary School Students' Opinions Related To Mass Media In The Terms Of Traffic Security (Sample: Çanakkale Province)]. Selçuk Üniversitesi İletişim Fakültesi Akademik Dergisi, 7(2): 44-57.
- Hatipoğlu S, Özdemir S & Öztürk E A (2012). Türkiye'de İlköğretim Okullarında Verilen Trafik Eğitiminin Farklı Ülkelerde Verilen Eğitimle Karşılaştırılarak İncelenmesi ve Geliştirilmesi İçin Öneriler [Examination of Traffic Education in Primary Schools in Turkey Through Comparison with Traffic Education Given in Different Countries and Suggestions for Improvement]. Abant İzzet Baysal Üniversitesi Eğitim Fakültesi Dergisi, 12(2): 9-22.
- Karakaya B & Kiper T (2013). Edirne Kent Merkezindeki Bazı İlköğretim Okul Bahçelerinin Peyzaj Tasarım İlkeleri Açısından Mevcut Durumunun Belirlenmesi [According to Landscape Design Principles Determination of Current Situations of Orchards of Some Elementary School in Edirne City Center]. Tekirdağ Ziraat Fakültesi Dergisi, 10(1): 59-71.
- Resmi Gazete (1998). Çocuk Trafik Eğitim Parkları Yönetmeliği. Resmî Gazete Sayı: 23503, Tarih: 24.10.1998.
- TSI (2018). Haber Bülteni. Karayolu Trafik Kaza İstatistikleri, 2017, Sayı: 27668, Tarih: 27 Haziran 2018. http://tuik.gov.tr/PreHaberBultenleri.do?id=27668.
- TSI (2019). Haber Bülteni. Adrese Dayalı Nüfus Kayıt Sistemi Sonuçları, 2018, Sayı: 30709, Tarih: 01 Şubat 2019. http://tuik.gov.tr/PreHaberBultenleri.do?id=30709.
- UN (2018). World Urbanization Prospects: The 2018 Revision. United Nations Department of Economic and Social Affairs, https://population.un.org/wup/Publications/Files/WUP2018-KeyFacts.pdf
- URL-1 (2019). Bağcılar TEP. https://uym.ibb.gov.tr/kurumsal/haberler-ve-duyurular/ibb-ba%C4%9Fc%C4%B1lar-trafike%C4%9Fitim-park%C4%B1-a%C3%A7%C4%B1ld%C4%B1 (22.04.2019).
- URL-2 (2019). Diyarbakır TEP. http://www.hurriyet.com.tr/yerel-haberler/diyarbakir/diyarbakirda-cocuk-trafik-egitim-parki-acildi-41046971 (22.04.2019).
- URL-3 (2019). Sakarya TEP. https://marmarabasin.com/yasam/serdivan-trafik-park-yaz-doneminde-de-cok-renkli-63443.htm (22.04.2019).
- URL-4 (2019). Tarsus TEP. http://www.tarsusbeyazhaber.com/cocuklara-trafik-egitim-parki.html (22.04.2019).
- URL-5 (2019). Kocaeli TEP. http://www.basiskele.bel.tr/haber/1/2889/park-acilmadan-cocuklarin-ilgi-odagi-oldu (22.04.2019).
- URL-6 (2019). Malatya TEP. http://www.percininsaat.com.tr/resim/userfiles/images/trafk parki 15 1280.jpg (22.04.2019).
- URL-7 (2019). Küçükçekmece TEP. http://kucukcekmece.istanbul/icerikler/tamamlanan-projeler/trafik-cocuk-egitim-parki/4315 (22.04.2019).
- URL-8 (2019). Pyongyang Traffic Safety Education Park for Children. https://exploredprk.com/articles/traffic-safety-education-parkfor-children/ (29.04.2019).
- URL-9 (2019). Fenyes Traffic Education Park. http://www.ujirany.com/project/traffic-education-park (29.04.2019).
- URL-10 (2019). Bartin Peyzaj. https://www.bartinpeyzaj.com/bartin\_belediyesi\_trafik\_egitim\_parki.htm (04.04.2019).
- Yenice M S (2013). İlköğretim Okulları İçin Mekânsal Yeterlilik Analizi; Burdur Örneği [A Spatial Sufficiency Analysis for Primary Schools; A Case Study of Burdur]. Hacettepe Üniversitesi Eğitim Fakültesi Dergisi, 28(3): 430-439.



## Effects of Different PCL Concentrations on the Nanofiber Morphology

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Abstract: From past to present there have been many research about nanofibers. In conclusion, it came to clear that; nanofibers have wide surface to volume ratios and unique magnetic, electrical and chemical properties. These properties have provided extensive usage in different areas, including tissue scaffold, drug release, textile, protective clothing, optical electronics, sensors, and filtrations. The method is based on applying an electrical field to the tip of the syringe loaded with polymer solution and collecting the polymer jet on the collector plate as nanofibers. Nanofiber formation and its morphology can be changed and/or effected depending to the properties of the polymer solutions (density, viscosity, conductivity and so on), process parameters (applied power, flow rate, distance between tip and collector plate and so on,) and medium condition (temperature and humidity). Changing of these parameters cause the differentiation on the fiber morphology such as nano-beads, beaded-fibers, beaded-free fibers, and smooth fibers. Therefore, the aim of the study was to determine the effects of different Polycaprolactone (PCL) concentrations (18, 20, 22 and 24%) on the fiber morphology. PCL is a linear, hydrophobic and synthetic polymer with high mechanical strength and was used in nanofiber optimization in this study. In the method section, PCL was weighed to obtain different concentrations and solved in 5 mL MeOH and 5 mL CHL. Then, all polymer solutions were electro-spinned in Inovenso NE200 equipment. Process properties were kept constant (flow rate: 2.65 mL/h, distance between tip and the collector plate: 15.7 cm, and applied electrical power: 17 kV). As a result, when the SEM micrographs were examined, beaded fibers were obtained and the diameters of fibers according to different polymer concentrations (18, 20, 22, and 24%) were measured 1.682, 1.859, 1.904, and ,017 µm, respectively. ImageJ software were used to determine the diameter of the fibers.

Keywords: Polycaprolactone (PCL), electrospining, nanofiber, polymer, morphology

## **1. INTRODUCTION**

Fundamentals of nanotechnology first was first mentioned in annual meeting of American Physicists Association 1959 by Richard P. Feynman who was the faculty member of California Technology Institute (Bushan, 2004). Thus, research on the development of materials with more functional and unique properties for the purpose has been started with atomatom and/or molecule-molecule sequence. Then, nanotechnology has been used in in polymer studies and became widespread in these areas. With the idea of enhancing efficiency and microminiaturization of polymers, researches in recent years have been concentrated on nanofiber technology. Nanofiber technology, has influenced science and related engineering disciplines on a large scale, because nanofibers have high mass – volume ratio, mechanical characteristics and unique magnetic, electrical and chemical qualifications. They play a crucial role in human life. According to that; tissue skeletons, textile, protective clothing, optical electronics, sensors, drug distributions and filtration are just some of the areas where nanofiber technology extensively used. For example, Akhgari and coworkers (2017) have proved that, nanofibers could be used as an approach to preparation of column drug distribution. Also Katti and coworkers (2004) have proved that, poly(lactit-co-glikolit) (PLAGA)nanofibers could be used for healing wounds by delivering antibiotics. In a similar way Ki and coworkers (2007) have reported that; membranes with nanofibers, produced by electro spinning process, are applicable for removal of heavy metals from water.

For producing nanofibers; template synthesis, melt spinning, phase inversion, electro spinning and other similar technics could be used. Among these; electro spinning is one of the easiest way to produce continuous and uniform scale nanofibers.

Electro spinning technique which commonly used for producing variable scaled nanofibers by using polymer solution and formation of electrostatic fiber; was patented first time in USA, 1902 but for a long time it could not find application area. (Ramakrishna 2005; Bhardwaj 2010) After Sir Taylor explained stability of viscose jets in the presence of high electric field; there had been many researches about physics of electro spinning and producing of nanofibers by electro spining. (Li 2004; Ramaskrishna 2005; Subbiah 2005; Naraghi 2011) These membranes show some properties like; high porosity; wide surface per volume/mass unit, high gas permeability and small internal pore size. Strings made of nanofibers could be produced at the rate of 70 meter/minute. Every polymer which have high enough molecular weight could be spinned by electro spinning.

This method is based on basis of polymer jets, produced by charging polymer solution or dilution, from high voltage exposed point; forming fibers on concentrator layer. (Li vd 2006; Croiser and Jerome 2013) Fiber films made by electro spinning method, have high porosity and surface. This makes them have widespread area to be used.



Figure 1. Method of Electrospining

Three parameters have effect on this method: i) Solution, ii) Process, iii) setting parameters. Solution parameters are density, molecular mass, viscosity, surface potential and conductivity. Process parameters are electrical power that used in process, flow rate, distance between needle and layer, inner diameter of needle, concentrator layer. Setting parameters are warmth and humidity. (Li Wang 2013) Also by making slight changes in these parameters; unwanted bead formations could be prevented (Liu.vd 2008). Thompson and friends (2007) have reported that; density, viscosity and distance between layer and needle have great effect on diameter of fibers, in their study. Similarly, Zhou and coworkers (2006) have done scientific research about setting parameters and proved that changes in warmth also effects diameter of fiber. Vrieze and coworkers (2009) have analyzed effect of humidity on nanofibers by using cellulose acetate (CA) and polyvinylpyrrolidone (PVP) As a result, with rise in humidity, average diameters of CA nanofibers go up but for PVP, it goes down. On the other hand, Liu and friends (2008) proved that, low surface potential causes bead formations and higher surface potential causes fewer and smaller beads. Ultimately they reported that, all of parameters have significant role in fiber morphology.

Polycaprolactone (PCL) is a polymer that is biodegradable, biocompatible, chemically and heat-resistant and approved by the Food and Drug Administration (FDA), used in some implantable devices known for its good mechanical properties. It is also a polymer with a semi-crystalline structure and does not like water (Karuppuswamy et al., 2015). It was used in this study due to the mentioned properties. In the light of the above information, the effects of different concentrations of polycaprolactone on nanofiber morphologies were investigated. Thus, the purpose of this study was to determine the effect of solution concentration on PCL fiber morphology.

### 2. MATERIALS AND METHODS

Materials: Poly (vinyl pyrrolidone) (Mw=360000 g/mol) from Sigma Aldrich and methanol (MeOH) and chloroform were received from Carlo Erba

**Methods**: In the study, PCL solutions were prepared at rates of 18, 20, 22, and 24%. For this0,9, 1, 1,1 ve 1,2 g PCL were weighed and added to the solvent containing 5 mL of MeOH-CHL (1:1 v/v) and stirred at room temperature until spinning.

The process parameters for all prepared solutions are kept constant and the solutions were spun at the conditions of 2.65 mL/h pump flow rate, 17.0 kV electrical power, 15.7 cm distance between the needle and the collector plate and with 0.6-inch (0.6 mm) tip inside diameter. To electrospinning of the solutions, Inovenso NE200 brand and model electrospinning device in the Central Research Laboratory of Kastamonu University, Polymer Research Laboratory was used.

**Characterization of Fiber Morphology:** FEI Quanta FEG250 brand and model scanning electron microscope (SEM) in Kastamonu University Central Research Laboratory were used for morphology. Fiber diameters were measured with ImageJ software.

## 3. RESULTS AND DISCUSSION

In this study, the effect of different PVP concentrations (18, 20, 22, and 24%) on nanofiber morphology was investigated. The SEM images of the nanofibers obtained with PCL solutions were presented below (Figure 2).



**Figure 2.** The SEM images of the nanofibers obtained with PCL solutions A, B, C and D belongs to 18, 20, 22, and 24% respectively.

According to SEM images not only smooth fiber formation but also the beadles fiber not obtained for all concentration. It was also observed that homogeneity of the fibers deteriorated with the increasing of the concentrations. When fiber diameters were examined, it was determined that the average fiber diameters for PCL at rates of 18, 20, 22, and 24% were 0.995, 1.682, 1.859, 1.904, 2.017  $\mu$ m, respectively, and nano to micron sized fibers were obtained.

As mentioned before fiber morphology has been affected many parameters like solution concentration and viscosity. There are four critical concentration values for solution concentrations:

- 1. If concentration is too low, polymeric nanoparticles (electrospray) during spinning are obtained,
- 2. Beaded-fibers are obtained when the concentration is increased a little,
- 3. Smooth, homogeneous and nano-sized fibers are obtained when a suitable concentration is used and
- 4. When the concentration is too high, either micron-sized fibers or micron-sized beaded-fibers are obtained (Li and Wang, 2013).

Therefore, the viscosity increases with increasing the concentration of the solution, the surface tension decreases. Similarly, as the concentration decreases, the surface tension will increase. Consequently, when the concentration of the solution increases, the viscosity becomes predominant in the electrospinning. when the concentration is reduced, the opposite occurs. Thus, when the surface tension increases, the polymeric nanoparticles are obtained, and when the viscosity increases, more uniform fibers are obtained (Liu et. al., 2008; Li and Wang, 2013). Thicker fibers are obtained by increasing the concentration of the solution and increasing in viscosity (Abutalep et al., 2017). In the same way, fiber diameters increased with the increase of solution density in this study.

In this study, it is possible that the density of the probable agents is low due to the fact that smooth and non-homogeneous fibers cannot be obtained.

#### 4. CONCLUSION

Fiber formation was not clearly visible for PCL spinning. The fibers were not smooth and beadless for all concentrations. Average fiber diameter were recorded as 0.995, 1.682, 1.859, 1.904, 2.017 µm respectively for different PCL concentration of 18, 20, 22, and 24%. The homogeneity of the fibers was deteriorated with increasing concentrations. As future study higher PCL concentration will be investigated to be able to get smooth and proper fiber formation

#### REFERENCES

- Abutaleb, A., Lolla, D., Aljuhani, A., Shin, H., Rajala, J., & Chase, G. (2017). Effects of surfactants on the morphology and properties of electrospun polyetherimide fibers. Fibers, 5(3), 33.
- Akhgari, A., Heshmati, Z., Garekani, H. A., Sadeghi, F., Sabbagh, A., Makhmalzadeh, B. S., & Nokhodchi, A. (2017). Indomethacin electrospun nanofibers for colonic drug delivery: In vitro dissolution studies. *Colloids and Surfaces B: Biointerfaces*, 152, 29-35.
- Bhardwaj, N., & Kundu, S. C. (2010). Electrospinning: a fascinating fiber fabrication technique. *Biotechnology advances*, 28(3), 325-347.
- Croisier, F., & Jérôme, C. (2013). Chitosan-based biomaterials for tissue engineering. European Polymer Journal, 49(4), 780-792.
- De Vrieze, S., Van Camp, T., Nelvig, A., Hagström, B., Westbroek, P., & De Clerck, K. (2009). The effect of temperature and humidity on electrospinning. *Journal of materials science*, 44(5), 1357-1362.
- Karuppuswamy, P., Venugopal, J. R., Navaneethan, B., Laiva, A. L., & Ramakrishna, S. (2015). Polycaprolactone nanofibers for the controlled release of tetracycline hydrochloride. Materials Letters, 141, 180-186.
- Katti, D. S., Robinson, K. W., Ko, F. K., & Laurencin, C. T. (2004). Bioresorbable nanofiber-based systems for wound healing and drug delivery: Optimization of fabrication parameters. Journal of Biomedical Materials Research Part B: Applied Biomaterials: An Official Journal of The Society for Biomaterials, The Japanese Society for Biomaterials, and The Australian Society for Biomaterials and the Korean Society for Biomaterials, 70(2), 286-296.
- Ki, C. S., Park, S. Y., Kim, H. J., Jung, H. M., Woo, K. M., Lee, J. W., & Park, Y. H. (2008). Development of 3-D nanofibrous fibroin scaffold with high porosity by electrospinning: implications for bone regeneration. *Biotechnology letters*, 30(3), 405-410.
- Li, C., Vepari, C., Jin, H. J., Kim, H. J., & Kaplan, D. L. (2006). Electrospun silk-BMP-2 scaffolds for bone tissue engineering. *Biomaterials*, 27(16), 3115-3124.
- Li, D., & Xia, Y. (2004). Electrospinning of nanofibers: reinventing the wheel?. Advanced materials, 16(14), 1151-1170.
- Li, Z., & Wang, C. (2013). Effects of working parameters on electrospinning. In *One-dimensional nanostructures* (pp. 15-28). Springer, Berlin, Heidelberg.
- Liu, Y., He, J. H., & Yu, J. Y. (2008). Bubble-electrospinning: a novel method for making nanofibers. In *Journal of Physics: Conference Series* (Vol. 96, No. 1, p. 012001). IOP Publishing.
- Naraghi, M., Arshad, S. N., & Chasiotis, I. (2011). Molecular orientation and mechanical property size effects in electrospun polyacrylonitrile nanofibers. *Polymer*, 52(7), 1612-1618.

Ramakrishna, S. (2005). An introduction to electrospinning and nanofibers. World Scientific.

- Subbiah, T., Bhat, G. S., Tock, R. W., Parameswaran, S., & Ramkumar, S. S. (2005). Electrospinning of nanofibers. *Journal of applied polymer science*, *96*(2), 557-569.
- Thompson, C. J., Chase, G. G., Yarin, A. L., & Reneker, D. H. (2007). Effects of parameters on nanofiber diameter determined from electrospinning model. Polymer, 48(23), 6913-6922.
- Zhou, H., Green, T. B., & Joo, Y. L. (2006). The thermal effects on electrospinning of polylactic acid melts. *Polymer*, 47(21), 7497-7505.



# Effects of Different PVP Concentrations on Nanofiber Morphology

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Abstract: From past to present there have been many research about nanofibers. In conclusion, it came to clear that; nanofibers have wide surface to volume ratios and unique magnetic, electrical and chemical properties. These properties have provided extensive usage in different areas, including tissue scaffold, drug release, textile, protective clothing, optical electronics, sensors, and filtrations. The method is based on applying an electrical field to the tip of the syringe loaded with polymer solution and collecting the polymer jet on the collector plate as nanofibers. Nanofiber formation and its morphology can be changed and/or effected depending to the properties of the polymer solutions (density, viscosity, conductivity and so on), process parameters (applied power, flow rate, distance between tip and collector plate and so on,) and medium condition (temperature and humidity). Changing of these parameters cause the differentiation on the fiber morphology such as nano-beads, beaded-fibers, beaded-free fibers, and smooth fibers. Therefore, the aim of the study was to determine the effects of different PVP concentrations (6, 8, 10, 12, 14, and 16%) on the fiber morphology. Poly (vinyl pyrrolidone) is an amorphous polymer having a high glass transition temperature (Tg). In addition, since it has good electrical properties, easy processability, moderately electrical conductivity and load carrying capacities. Therefore, it was selected for the nanofiber optimization in this study. In the method section, PVP was weighed to obtain different concentrations and solved in 5 mL MeOH. Then, all polymer solutions were electro-spinned in Inovenso NE200 equipment. Process properties were kept constant (flow rate: 0.75 mL/h, distance between tip and the collector plate: 10 cm, and applied electrical power: 15 kV). As a result, when the SEM micrographs were examined, both bead-free (smooth) nano and micro fibers were obtained and the diameters of fibers according to different polymer concentrations (6, 8, 10, 12, 14, and 16%) were measured 0,995, 1,308, 1,537, 1,953, 4,535, and 3,563 µm, respectively. ImageJ software were used to determine the diameter of the fibers.

Keywords: Poly (vinyl pyrrolidone) (PVP), electrospinning, nanofiber, polymer, morphology

### **1. INTRODUCTION**

The idea of nanotechnology was first put forward in the speech of Richard P. Feynman from the California Institute of Technology professors at the annual general meeting of the American Physical Society in 1959 (Bushan, 2004). Thus, research on the development of materials with more functional and unique properties for the purpose has been started with atom-atom and/or molecule-molecule sequence. In the field of polymer, studies have mostly shifted to nanofiber technology. Nanofiber technology has greatly influenced science and engineering disciplines. Because nanofibers have high mass volume ratio and mechanical properties and unique magnetic, electrical and chemical properties (Kiyak, 2014; Haider et. al., 2018).

These features play an important role in human life today (Chen and Kotek, 2008). So recently nanofiber technology are researched and used in the field of tissue scaffolds, textiles, personal protective equipments, optical electronics, drug delivery and filtration (Subbiah, 2005).

For instance, Katti et. al., (2004) showed that poly(lactite-co-glycolite) (PLAGA) nanofibers could be used as antibiotic delivery systems for the treatment of wounds. Similarly, Ki et. al., (2007) reported that the nanofiber membranes prepared by the electro-spinning process were very suitable for the removal of heavy metals in the water.

It is used some different technics as template synthesis, melt spinning, phase inversion and electro-spinning to produce nanofibers. Among them, electrospinning is one of the simplest methods to produce continuous and uniform diameter nanofibers (Li et. al., 2004).

Membranes produced by electro-spinning method show many superior features as high porosity, large surface area per unit volume/mass, high gas permeability and small inner pore size (Saeed et. al., 2008). Nano fibers can be produced at a speed of 70 m/min. Almost any polymer with sufficiently high molecular weight can be spun with electro-spinning (Ramakrishna et. al., 2005).

The electrospinning method is based on applying an electrical field to the tip of the syringe loaded with polymer solution and collecting the polymer jet on the collector plate as nanofibers (Li et. al., 2006; Croisier and Jerome, 2013). The method is influenced by three main parameters as solution parameters, process parameters and environmental conditions. Solution parameters are concentration of polymers, the density of the solution, molecular weight, viscosity, surface tension and conductivity. Process parameters are applied electrical power used, flow rate, distance between plate and tip, tip inside diameter and collector plate. Environmental parameters are relative humidity and temperature (Li and Wang, 2013).

When sensitive changes are made in these parameters, unwanted bead formations can be prevented with fiber structure (Liu et. al., 2008). Many studies on fiber morphology have been carried out. For example; Liu et al. (2008) showed in their study that bead formation is mainly caused by low surface tension and the size of the beads in the fibers was small and the number of the beads were few at higher surface tension. Finally, all of these parameters are of great importance to determine fiber morphology.

Thompson et al. (2007) were reported in their study that the parameters (concentration, the distance between the tip and the collector plate and viscosity) had the greatest impact on the nanofiber morphology.

The aim of this study was investigated the effects of different concentrations (6, 8, 10, 12, 14, and 16%) of PVP on nanofiber morphologies.

## 2. MATERIALS AND METHODS

Poly (vinyl pyrrolidone) (Mn=360000 g/mol) from Sigma Aldrich and methanol (MeOH) from Carlo Erba were bought.

In the study, PVP solutions were prepared at rates of 6, 8, 10, 12, 14, and 16%. For this, 0.6, 0.8, 1.0, 1.2, 1.4, and 1.6 g PVP were weighed and added to the solvent containing 10 mL of MeOH and stirred by means of magnetic stirrer at room temperature and solutions were prepared for spinning.

The process parameters for all prepared solutions are kept constant and the solutions were spun at the conditions of 0.75 mL/h pump flow rate, 15.0 kV electrical power, 10.0 cm distance between the needle and the collector plate and with 0.6-inch (0.6 mm) tip inside diameter.

To electrospinning of the solutions, Inovenso NE200 brand and model electrospinning device in the Central Research Laboratory of Kastamonu University, Polymer Research Laboratory was used. FEI Quanta FEG250 brand and model scanning electron microscope (SEM) in Kastamonu University Central Research Laboratory were used for morphology. Fiber diameters were measured with ImageJ software.

## 3. RESULTS AND DISCUSSION

### Results

In this study, the effect of different PVP concentrations (6, 8, 10, 12, 14, and 16%) on nanofiber morphology was investigated.

The SEM images of the nanofibers obtained with PVP solutions can be seen in Figures 2.



Figure 2. The SEM images of the nanofibers obtained with different PVP solutions concentrations

When the SEM images were examined, it was observed that both the fiber formation and beadless fibers obtained for all concentrations. Meanwhile, it was observed that the homogenity of the fibers deteriorated with the increasing of the concentrations. When fiber diameters were examined, it was determined that the average fiber diameters for PVP at rates of 6, 8, 10, 12, 14, and 16% were 0,995, 1,308, 1,537, 1,953, 4,535 ve 3,563 µm, respectively, and nano to micron sized fibers were obtained.

## Discussion

There are four critical concentration values for solution concentrations:

- 1. If concentration is too low, polymeric nanoparticles (electrospray) during spinning are obtained,
- 2. Beaded-fibers are obtained when the concentration is increased a little,
- 3. Smooth, homogeneous and nano-sized fibers are obtained when a suitable concentration is used and

4. When the concentration is too high, either micron-sized fibers or micron-sized beaded-fibers are obtained (Li and Wang, 2013).

While the viscosity increases with increasing the concentration of the solution, the surface tension decreases. Similarly, as the concentration decreases, the surface tension will increase. Therefore, when the concentration of the solution increases, the viscosity becomes predominant in the electrospinning. when the concentration is reduced, the opposite occurs. Thus, when the surface tension increases, the polymeric nanoparticles are obtained, and when the viscosity increases, more uniform fibers are obtained (Liu et. al., 2008; Li and Wang, 2013). Thicker fibers are obtained by increasing the concentration of the solution and increasing in viscosity (Abutalep et al., 2017). In the same way, fiber diameters increased with the increase of solution density in this study. At the same time, our results were also consistent with the results of Thompson et al. (2007).

### 4. CONCLUSION

Fiber formation was clearly visible for PVP spining. The fibers were smooth and beadless for all concentrations. The homogeneity of the fibers was deteriorated with increasing concentrations.

#### REFERENCES

- Abutaleb, A., Lolla, D., Aljuhani, A., Shin, H., Rajala, J., & Chase, G. (2017). Effects of surfactants on the morphology and properties of electrospun polyetherimide fibers. Fibers, 5(3), 33.
- Croisier, F., & Jérôme, C. (2013). Chitosan-based biomaterials for tissue engineering. European Polymer Journal, 49(4), 780-792.
- Haider, A., Haider, S., & Kang, I. K. (2018). A comprehensive review summarizing the effect of electrospinning parameters and potential applications of nanofibers in biomedical and biotechnology. *Arabian Journal of Chemistry*, 11(8), 1165-1188.
- Katti, D. S., Robinson, K. W., Ko, F. K., & Laurencin, C. T. (2004). Bioresorbable nanofiber-based systems for wound healing and drug delivery: Optimization of fabrication parameters. Journal of Biomedical Materials Research Part B: Applied Biomaterials: An Official Journal of The Society for Biomaterials, The Japanese Society for Biomaterials, and The Australian Society for Biomaterials and the Korean Society for Biomaterials, 70(2), 286-296.
- Ki, C. S., Park, S. Y., Kim, H. J., Jung, H. M., Woo, K. M., Lee, J. W., & Park, Y. H. (2008). Development of 3-D nanofibrous fibroin scaffold with high porosity by electrospinning: implications for bone regeneration. *Biotechnology letters*, 30(3), 405-410.
- Kiyak, Y. E., & Cakmak, E. (2014). Nanofiber Production Methods. *Electronic Journal of Vehicle Technologies/Tasit Teknolojileri Elektronik Dergisi*, 8(3).
- Li, C., Vepari, C., Jin, H. J., Kim, H. J., & Kaplan, D. L. (2006). Electrospun silk-BMP-2 scaffolds for bone tissue engineering. *Biomaterials*, 27(16), 3115-3124.
- Li, D., & Xia, Y. (2004). Electrospinning of nanofibers: reinventing the wheel?. Advanced materials, 16(14), 1151-1170.
- Li, Z., & Wang, C. (2013). Effects of working parameters on electrospinning. In *One-dimensional nanostructures* (pp. 15-28). Springer, Berlin, Heidelberg.
- Liu, Y., He, J. H., & Yu, J. Y. (2008). Bubble-electrospinning: a novel method for making nanofibers. In *Journal of Physics: Conference Series* (Vol. 96, No. 1, p. 012001). IOP Publishing.
- Ramakrishna, S. (2005). An introduction to electrospinning and nanofibers. World Scientific.
- Saeed, K., Haider, S., Oh, T. J., & Park, S. Y. (2008). Preparation of amidoxime-modified polyacrylonitrile (PAN-oxime) nanofibers and their applications to metal ions adsorption. *Journal of Membrane Science*, 322(2), 400-405.
- Subbiah, T., Bhat, G. S., Tock, R. W., Parameswaran, S., & Ramkumar, S. S. (2005). Electrospinning of nanofibers. *Journal of applied polymer science*, *96*(2), 557-569.
- Thompson, C. J., Chase, G. G., Yarin, A. L., & Reneker, D. H. (2007). Effects of parameters on nanofiber diameter determined from electrospinning model. Polymer, 48(23), 6913-6922.



# Investigation of The Correlation Between Diabetes Mellitus and Blood Pressure in Tripoli Central Hospital

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**Abstract:** One of the most widespread diseases related to diabetes is hypertension. Patients suffering from high blood pressure are diabetes-prone, and those suffering from diabetes are also exposed to hypertension at any time. Diabetes mellitus and arterial hypertension are common chronic disorders that frequently occur together. Also, cardiovascular events are higher in patients having both diabetes and hypertension than the patients having only one of these two diseases. 50% of patients with type 2 diabetes have hypertension at the time of the diagnosis. Cardiovascular disease is the most important cause of death in both types of diabetes.

Materials-Methods: This study was conducted at Tripoli Central Hospital. 200 of the 350 total diabetic patients having hypertension were included in this study, and the required analyses were performed. Glucose measurement was done first from the urine and then from the blood. Blood pressures were measured and recorded in each patient. Blood glucose measurement is the only criteria for the diagnosis of diabetes, and it is useful during treatment, especially in the monitoring of insulin-dependent diabetic patients, who may experience high blood sugar. The method used to measure glucose concentration in the blood depends on the events of a colored carbohydrate reaction with reagent outside the dehydrate.

Results: The rate of patients having coexisting hypertension with diabetes was 57%. According to the results of the statistics obtained from Tripoli Central Hospital, the proportion of women with high blood pressure complicated with diabetes (4-5) within two months is higher than that of men (57.9% in women and 42.1% in men). The total number of hypertension-related diabetes cases collected from diabetic patients with high blood pressure was 117 in women and 83 in men. The prevalence of blood pressure in adults with diabetes is 20-60%. This is 1.5-3 times higher than in non-diabetic individuals.

Conclusion: The optimal BP goal for diabetic patients should be personalized. However, the current literature shows that the maximum benefit of BP control in diabetic patients is achieved with systolic BP levels between 80 and 85 mmHg, except for stroke prevention.

Keywords: Hypertension, Diabetes Mellitus, Cardiovascular disease.



# Effects of Chestnut and Fat/Oil Contents On Textural and Sensorial Properties of Cocoa Chestnut Cream as A New Product

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**Abstract:** The textural and sensory properties of cocoa chestnut cream containing different fat/oil and chestnut ratios were investigated in this research. Two different chestnut (7.5 and 10%) and fat/oil ratios (25.75 and 28.25%) were used in laboratory scale model production. The main goal of this research was to develop cocoa chestnut cream as a new product.

As the content of chestnut increased, accelerated oil separation and yield stress values increased, conversely stickiness, apparent viscosity, plastic viscosity, consistency index values and body-texture score decreased. The chestnut content had no significant effect on spreadability, flavor-aroma and color-appearance. As the fat/oil rate increased, it was found that accelerated oil separation value and body-texture score increased, while spreadability, stickiness, apparent viscosity and plastic viscosity and consistency index values decreased.

In general, the panelists were convinced that the creams obtained were "good" from the sensory point of view. Considering the similarity of textural and sensory characteristics such as AOS, spreadability, stickiness, body–texture, and appearance–color to commercial samples in literature, it can be concluded that cocoa chestnut cream containing 28.25% fat/oil and 10% chestnut flour is suitable for production.

The development of cocoa chestnut cream would increase utilization of chestnut as a food, offer an alternative food product to consumer, and provide economic contribution to the chestnut grower. However, it may be useful to slightly increase the amount of liquid oil compared to solid fat to improve the spreadability of the product. To improve the flavor– aroma properties, the addition of a commercial chestnut aroma or roasting of the chestnut or chestnut flour can be applied.

Keywords: Chestnut, Cocoa chestnut cream, Texture, Sensory



# The Effect of Using Transglutaminase on Free Aminoacid Composition and Volatile Compound Profile of Pastırma

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Abstract: In the research, the effects of different ratios (0%-control, 0.25%, 0.50% and 1%) of transglutaminase enzyme preparate on free aminoacid composition and volatile compound profile in pasturma, a traditional Turkish dry-cured meat product, were investigated. The thiobarbituric acid reactive substances (TBARS) value and non-protein nitrogenous substance (NPN-S) content of pastirma samples were also detected. Pastirma production was carried out under the controlled conditions. Enzyme preparate was applied after the first drying stage of production. Enzyme application factor did not show a statistically significant effect on NPN-S content (P>0.05). NPN-S content of pastirma samples changed between 4.82% and 5.97%. As the applied enzyme level increased, the TBARS value, which is an indicator of lipid oxidation, decreased (P<0.05). The highest TBARS value was determined in the control group (30.061±2.558 µmol MDA/kg), while the lowest value was determined in the samples with 1% enzyme preparate (15.937±2.031 µmol MDA/kg). Enzyme application caused a significant (P < 0.05) effect on arginine and proline and a very significant effect (P<0.01) on cystine and phenylalanine. Total free aminoacid contents including aspartic acid, glutamic acid, serine, histidine, glycine, threonine, alanine, tyrosine, valine, methionine, isoleucine, leucine and lysine were not affected by enzyme application (P>0.05). In control group and pastirma samples treated with 0.25%, 0.50% and 1% of transglutaminase, mean FAA levels were detected 1774.76±195.69, 1660.35±11.82, 1847.51±128.86 and 1847.35±37.06 mg/100 g dry matter, respectively. On the other hand, a total of 49 volatile compounds, including alcohols, aldehydes, ketones, aliphatic and aromatic hydrocarbons, terpenes, sulfur compounds, esters, furans, acids and nitrogenous compounds were identified from pastirma samples. Aldehydes were detected as a major chemical groups and hexanal was a major compound in all pastirma groups. However, it was determined that enzyme application had no significant effect on volatile compounds (P>0.05). It was concluded that the enzyme application reduces lipid oxidation, an important quality parameter in the product, and thus has a positive effect on the quality of the product.

Keywords: Pastırma, Transglutaminase, TBARS, NPN-S, Volatile compound profile, free aminoacid composition

### **1. INTRODUCTION**

Pastrma is a traditional dry-cured meat product obtained by applying curing, drying, pressing and cemen application procedures to large pieces of meat obtained from beef or buffalo carcasses (Tekinşen and Doğruer 2000; Gökalp *et al.* 2012; Kaban 2013). In the production of pastrma, the incision procedure is applied for a better diffusion of the curing components into the meat. For this purpose, the knifepoint is held in the direction of the muscle fibers to make a 45 °angle to the surface of the meat, and deep cuts are made. The depth of these cuts is applied to half of the meat for pastrma and applied in series (Tekinşen and Doğruer 2010). However, appearance defects may be encountered in the final product due to this process. Therefore, it was considered that this problem could be eliminated by the use of transglutaminase enzyme, which provides cross-linking between lysine and glutamine in pastrma production.

In dry-cured meat products such as dry-cured ham and pastirma chemical and physicochemical changes occur during the production time (Lorenzo et al. 2008; Kaban 2009). Lipolysis and proteolysis reactions that occur during ripening lead to the formation of compounds that are the precursor of flavor (free amino acids, free fatty acids) (Toldrá and Flores 1998; Flores *et al.* 2007: Kaban 2013; Lorenzo and Carballo 2015). Furthermore, lipid oxidation occurring in these types of products also has a positive effect on the formation of typical aroma (Harkouss *et al.* 2015). On the other hand, proteolysis and lipolysis are responsible for the sensory characteristics of the final product (Lorenzo and Carballo 2015).

In this study, the effects of usage of different transglutaminase levels (0%, 0,25%, 0,50% and 1%) on TBARS, NPN-S content, volatile compound profile and free amino acid composition of pasturma were investigated.

### 2. MATERIALS AND METHODS

### **Pastirma Production**

The *M.Longissimus thoracis et lumborum* muscle obtained from two different carcasses was used in the production. The pieces obtained from ½ carcass were divided into two parts and thus 4 pieces of meat for pastirma were obtained from a carcass. One of these pieces was considered as control. The other three pieces were treated with transglutaminase enzyme by 0.25%, 0.50% and 1% upon meat weight. Enzyme application was performed before the 1st pressing stage. In the production of pastirma, 5% salt, 150 ppm nitrite and 0.3% sucrose were used in the curing stage. The production of pastirma was conducted under controlled conditions in a climate chamber (Reich, Germany).

While the analysis of thiobarbituric acid reactive substances (TBARS) was performed according to the method provided by Lemon (1975), non-protein nitrogenous substances (NPN-S) analysis was performed according to the method provided by Anonymous (1989).

The method provided by Kaban (2009) was used in determining the volatile compound profile. The extraction of free amino acids was performed according to Antoine et al. (1999). In the derivatization process, 50  $\mu$ L of the filtrate was added and mixed into 250  $\mu$ L of borate buffer (pH 10.2). Then, 50  $\mu$ L OPA reagent (phthaldialdehyde reagent, Sigma) and 50  $\mu$ L FMOC reagent (9-fluorenylmethylchloroformate in acetonitrile, Agilent Technologies) were added and mixed, and 3200 mL of distilled water was added to it. After derivatization, the samples were referred to an HPLC/FLD device (Agilent Technologies 1100 Series, USA) within 5 minutes, and the amounts of free amino acids were determined as mg/100 g dry matter.

### **Statistical Analysis**

Data were analyzed according to randomized complete block design with two replicates by using SPSS software (SPSS 20.0). Differences among means of results were tested for significance (P < 0.05) by Duncan's multiple range test.

### 3. RESULTS AND DISCUSSION

The effects of different enzyme levels on TBARS value and NPN-S content of pastirma samples are showed in Table 1. According to these results, NPN-S content was not affected by enzyme level. On the other hand, enzyme application caused differences in TBARS values (P<0,01). As the applied enzyme ratio increased, the TBARS value, an indicator of lipid oxidation, decreased (Table 1).

| <b>Enzyme Ratio</b> | NPN-S (g/100g) | TBARS(µmolMDA/kg) |
|---------------------|----------------|-------------------|
| Control             | 5,43±0,76a     | 30,061±2,558a     |
| 0,25%               | 5,35±0,60a     | 21,090±6,438b     |
| 0,50%               | 5,72±0,21a     | 18,700±1,389c     |
| 1%                  | 5.05±0.33a     | 15,937±2,031d     |

Table 1. The effects of different enzyme levels on the TBARS value and NPN-S content of pastirma samples (mean±SD)

a-d: any two means on the same colon having to the same letters are not significantly different (P>0.05)

A total of 49 volatile compounds, including alcohols, aldehydes, ketones, aliphatic and aromatic hydrocarbons, acids, sulphur compounds, esters, furans, nitrogenous compounds, were identified in pasturma samples. None of the volatile compounds were affected by the enzyme application (P>0,05) (Table 2). Akköse et al. (2017) detected 46 volatile compounds in pasturma. On the other hand 122 volatile compounds were determined in Iberian ham (Martínez-Onandi et al. 2017).

| Table 2. | The | effects | of | different | enzym | e levels | on | volatile | compound | s of | pastırma | samples | (mean±SD | )) |
|----------|-----|---------|----|-----------|-------|----------|----|----------|----------|------|----------|---------|----------|----|
|          |     |         |    |           |       |          |    |          |          |      |          |         |          |    |

| Compounds                          | Enzyme Application |              |               |              |  |  |  |
|------------------------------------|--------------------|--------------|---------------|--------------|--|--|--|
| Compounds                          | Control            | 0,25%        | 0,50%         | 1%           |  |  |  |
| Alcohols                           |                    |              |               |              |  |  |  |
| Ethanol                            | 10,46±4,43a        | 9,04±4,16a   | 14,61±5,81a   | 12,36±7,04a  |  |  |  |
| 1-propen-2-ol                      | 0,00±0,00a         | 0,35±0,70a   | 1,64±1,11a    | 0,53±1,07a   |  |  |  |
| Aldehydes                          |                    |              |               |              |  |  |  |
| Acetaldehyde                       | 6,99±1,41a         | 9,09±5,99a   | 5,27±0,73a    | 5,45±0,73a   |  |  |  |
| Heptanal                           | 0,67±0,42a         | 0,45±0,37a   | 0,56±0,43a    | 0,54±0,48a   |  |  |  |
| Pentanal                           | 5,39±3,93a         | 4,09±2,74a   | 9,13±4,00a    | 6,02±4,37a   |  |  |  |
| Hexanal                            | 64,86±41,88a       | 48,68±27,43a | 101,89±39,52a | 71,07±46,94a |  |  |  |
| 2-hexenal                          | 0,25±0,24a         | 0,25±0,15a   | 0,33±0,17a    | 0,29±0,25a   |  |  |  |
| Heptanal                           | 4,89±2,85a         | 3,75±2,15a   | 7,02±2,89a    | 5,53±3,50a   |  |  |  |
| 2-heptenal (E)                     | 0,26±0,37a         | 0,16±0,20a   | 0,09±0,18a    | 0,51±0,42a   |  |  |  |
| Benzaldehyde                       | 28,38±7,52a        | 30,08±8,13a  | 34,03±9,67a   | 23,13±7,89a  |  |  |  |
| Octanal                            | 8,09±4,13a         | 6,69±3,65a   | 11,10±4,30a   | 8,30±4,89a   |  |  |  |
| 2,4-heptadienal (E,E)-             | 0,90±0,98a         | 1,09±1,32a   | 1,28±0,77a    | 0,78±0,76a   |  |  |  |
| 2-Octenal (E)-                     | 1,60±0,86a         | 1,72±1,08a   | 1,91±0,77a    | 1,33±0,61a   |  |  |  |
| Nonanal                            | 15,19±5,61a        | 12,67±6,01a  | 19,34±6,67a   | 14,64±6,09a  |  |  |  |
| Trans-2-nonenal                    | 1,70±0,42a         | 1,70±0,37a   | 1,56±0,35a    | 1,29±0,41a   |  |  |  |
| 2,4-nonadienal                     | 0,26±0,30a         | 0,37±0,27a   | 0,41±0,30a    | 0,45±0,12a   |  |  |  |
| 2-methyl-3-phenyl-propanal         | 2,75±1,89a         | 1,97±1,25a   | 1,90±0,92a    | 1,29±0,75a   |  |  |  |
| 2-methyl-2-butenal                 | 1,57±0,99a         | 0,82±0,88a   | 1,82±0,80a    | 1,37±0,99a   |  |  |  |
| Ketones                            |                    |              |               |              |  |  |  |
| 2-cyclopenten-1-one                | 0,89±0,82a         | 0,77±0,71a   | 1,21±0,62a    | 0,06±0,12a   |  |  |  |
| 2-pentanone                        | 1,61±1,26a         | 1,25±0,91a   | 1,76±0,74a    | 1,12±0,94a   |  |  |  |
| 2-heptanone                        | 0,49±0,25a         | 0,52±0,22a   | 0,71±0,24a    | 0,52±0,28a   |  |  |  |
| 2,3-octanedione                    | 15,07±6,71a        | 11,09±10,90a | 23,32±9,54a   | 15,66±10,01a |  |  |  |
| 6-methyl-5-hepten-2-one            | 1,03±1,38a         | 0,41±0,40a   | 0,57±0,32a    | 0,32±0,21a   |  |  |  |
| 2-octanone                         | 0,22±0,17a         | 0,03±0,05a   | 0,34±0,24a    | 0,21±0,17a   |  |  |  |
| 5-methyl-3-hepten-2-one            | 1,13±1,30a         | 1,54±0,75a   | 1,97±0,41a    | 0,94±0,49a   |  |  |  |
| 3-octen-2-one                      | 1,01±0,40a         | 1,06±0,63a   | 1,27±0,41a    | 0,82±0,37a   |  |  |  |
| 3,5-octadien-2-one                 | 1,30±0,56a         | 1,08±0,43a   | 0,64±0,48a    | 0,82±0,20a   |  |  |  |
| Aromatic Hydrocarbons              |                    |              |               |              |  |  |  |
| 1-methyl-2-(1-methylethyl)-benzene | 1,66±1,31a         | 1,12±0,98a   | 1,37±0,66a    | 0,70±0,30a   |  |  |  |
| Styrene                            | 0,32±0,27a         | 0,35±0,08a   | 0,42±0,17a    | 0,30±0,10a   |  |  |  |
| Alifatic Hydrocarbons              |                    |              |               |              |  |  |  |
| 1,3-pentadiene                     | 21,74±18,38a       | 17,35±17,43a | 36,35±20,37a  | 29,41±21,27a |  |  |  |
| Hexane                             | 27,11±16,18a       | 23,15±14,96a | 25,13±15,29a  | 22,30±12,36a |  |  |  |
| Octane                             | 0,59±0,46a         | 0,42±0,62a   | 0,27±0,53a    | 0,52±0,51a   |  |  |  |
| Nonane                             | 0,30±0,23a         | 0,30±0,09a   | 0,43±0,15a    | 0,34±0,13a   |  |  |  |
| Decane                             | 0,42±0,16a         | 0,22±0,15a   | 0,39±0,12a    | 0,41±0,18a   |  |  |  |
| Undecane                           | 1,10±0,83a         | 1,05±0,45a   | 1,40±0,71a    | 0,80±0,27a   |  |  |  |
| Dodecane                           | 1,01±0,82a         | 1,21±0,99a   | 1,26±0,60a    | 1,07±0,50a   |  |  |  |
| Tridecane                          | 0,58±0,45a         | 0,47±0,60a   | 0,75±0,67a    | 0,71±0,37a   |  |  |  |
| Tetradecane                        | 0,64±0,57a         | 0,78±0,50a   | 0,45±0,54a    | 0,00±0,00a   |  |  |  |
| Sulphur Compounds                  |                    |              |               |              |  |  |  |
| Allyl mercaptan                    | 1,64±1,25a         | 0,81±0,68a   | 1,35±0,82a    | 2,33±1,68a   |  |  |  |
| 3,3'thiobis-1-propene              | 2,63±2,21a         | 2,50±2,95a   | 4,19±2,53a    | 3,36±2,51a   |  |  |  |
| Methyl 2-propenyl disulfide        | 2,11±1,20a         | 1,95±1,25a   | 2,73±1,19a    | 2,15±1,07a   |  |  |  |

| Table 2 c | ontinued |
|-----------|----------|
|-----------|----------|

| Diallyl disulphide                 | 17,90±8,14a | 18,56±8,84a | 22,32±8,11a | 22,39±7,49a |
|------------------------------------|-------------|-------------|-------------|-------------|
| 3-(methylthio)-1-propene           | 0,19±0,38a  | 0,20±0,40a  | 0,36±0,42a  | 0,33±0,38a  |
| Esteres                            |             |             |             |             |
| 2,4-hexadienoic acid, methyl ester | 3,69±0,75a  | 3,11±1,02a  | 2,95±1,66a  | 2,35±0,92a  |
| Ethyl acetate                      | 1,49±1,25a  | 1,62±1,31a  | 2,12±1,11a  | 1,50±1,40a  |
| Furans                             |             |             |             |             |
| 2-butyl furan                      | 0,15±0,12a  | 0,12±0,14a  | 0,36±0,15a  | 0,23±0,20a  |
| 2-pentyl furan                     | 0,99±0,67a  | 0,83±0,52a  | 1,51±0,73a  | 0,83±0,36a  |
| Nitrogenous Compounds              |             |             |             |             |
| 1-methyl-1H-pyrrole                | 0,50±0,46a  | 0,42±0,40a  | 0,23±0,19a  | 0,37±0,43a  |
| Acids                              |             |             |             |             |
| Asetic acid                        | 3,73±1,11a  | 3,03±1,56a  | 2,59±1,04a  | 2,16±1,51a  |

a: any to means on the same line having to the same letters are not significantly different (P>0,05)

The effects of different enzyme levels on free amino acids of pasturma samples are showed in Table 3. Proteolysis plays important role in the quality of meat products. The amino acids and peptides formed by proteolysis affect directly taste of the product. Amino acids which are precursor of some volatile compounds affect the aroma of dry-cured meat products (Toldrá and Flores 1998). In this study, glutamic acid, arginine, alanine, sistine and valine detected as dominant amino acids (Table 3). Transglutaminase enzyme application had a very significant effect (P<0,01) on cystine and phenylalanine. Arginine and proline were also affected by this treatment (P<0,05).

| Amino Aoida      | Enzyme Application |                |                 |                |  |  |
|------------------|--------------------|----------------|-----------------|----------------|--|--|
| Amino Acius      | Control            | 0,25%          | 0,50%           | 1%             |  |  |
| Aspartic Acid    | 32,28±6,63a        | 30,31±4,27a    | 31,79±3,43a     | 25,89±9,75a    |  |  |
| Glutamic Acid    | 151,95±85,17a      | 155,82±109,41a | 207,49±147,12a  | 195,08±129,54a |  |  |
| Serine           | 25,75±14,64a       | 27,39±15,57a   | 34,88±32,25a    | 30,99±28,22a   |  |  |
| Histidine        | 36,22±12,60a       | 32,27±10,07a   | 28,04±4,85a     | 33,25±8,17a    |  |  |
| Glycine          | 52,13±27,86a       | 36,48±35,59a   | 68,16±44,52a    | 66,08±42,71a   |  |  |
| Threonine        | 101,28±16,26a      | 106,05±93,33a  | 31,76±25,60a    | 69,41±23,03a   |  |  |
| Arginine         | 256,20±24,42a      | 192,17±9,37b   | 255,57±28,28a   | 274,64±59,87a  |  |  |
| Alanine          | 125,68±9,18a       | 119,68±3,39a   | 124,75±5,83a    | 125,24±6,93a   |  |  |
| Tyrosine         | 131,15±50,85a      | 110,86±11,96a  | 153,74±14,24a   | 143,39±20,44a  |  |  |
| Cystine          | 248,10±80,29c      | 279,34±66,70bc | 331,22±87,43a   | 298,20±94,49ab |  |  |
| Valine           | 230,27±30,22a      | 229,72±26,53a  | 239,61±23,51a   | 236,01±23,29a  |  |  |
| Methionine       | 121,75±56,69a      | 93,84±15,90a   | 108,69±13,22a   | 112,10±45,57a  |  |  |
| Phenylalanine    | 76,85±4,34a        | 91,64±5,71a    | 48,82±3,98b     | 57,48±22,67b   |  |  |
| Isoleucine       | 15,49±5,78a        | 14,41±2,83a    | 15,18±3,18a     | 13,76±4,90a    |  |  |
| Leucine          | 50,62±10,12a       | 44,67±3,59a    | 44,28±7,70a     | 37,83±25,95a   |  |  |
| Lysine           | 108,67±50,60a      | 89,30±29,87a   | 115,97±17,45a   | 117,59±21,20a  |  |  |
| Proline          | 10,38±4,57a        | 6,41±2,73b     | 7,58±2,10ab     | 10,42±5,68a    |  |  |
| Total Amino Acid | 1774.76±195.69a    | 1660.35±11.82a | 1847.51±128.86a | 1847.35±37.06a |  |  |

Table 3. The effect of different enzyme levels on free amino acids of pastirma samples (mean±SD)

a-c: any two means on the same line having to the same letters are not significantly different (P>0,05)

### 4. CONCLUSION

The use of transglutaminase in pastrma production reduced the lipid oxidation. However, transglutaminase caused no differences in volatile compound profile. A similar result was obtained for NPN-S content. On the other hand, this application had only a limited influence on the free amino acid composition of pastrma.

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#### REFERENCES

- Akköse, A., Ünal, N., Yalınkılıç, B., Kaban, G., and Kaya, M.,2017. Volatile compounds and some physico-chemical properties of pastırma produced with different nitrate levels. Asian-Australasian Journal of Animal Sciences 30(8): 1168-1174.
- Anonymus,1989. Untersuchung von Lebensmitteln. Bestimmung des Gehaltes an Nichprotein-Stickstoffsuntanz in Fleisherzeugnissen. 07.00-41. Amtliche Sammlung von Untersuchungsverfahren nach, 35 LMBG.
- Antoine, F.R., Wei, C.I., Littell, R.C., and Marshall, M.R.,1999. HPLC method for analysis of free aminoasits in fish using ophthaldialdehyde precolumn derivatization. Journal of Agricultural and Food Chemistry 47: 5100-5107.
- Flores, M., Gianelli MP., Pérez-Juan, M., and Toldrá F.,2007. Headspace concentration of selected dry-cured aroma compounds in model systems as affected by curing agents. Food Chemistry 102: 488–493.
- Gökalp, H.Y., Kaya, M., and Zorba, Ö.,2012. Et ürünleri işleme mühendisliği, Atatürk Üniversitesi yayın no:786, Ziraat Fakültesi yayın no: 320, Ders kitapları serisi No:70, Atatürk Üniversitesi Ziraat Fakültesi, Erzurum.
- Harkouss, R., Chevarin, C., Daudin, J.D., Sicard, J., and Mirade, P.S., 2018. Development of a multi-physical finite element-based model that predicts water and salt transfer, proteolysis and water activity during the salting and post-salting stages of the drycured ham process. Journal of Food Engineering 218: 69-79.
- IBM SPSS Statistics 20.0,2011. Statisticsal Package for the Social Sciences, United States, Chicago.
- Kaban, G., 2009. Changes in the composition of volatile compounds and in microbiological and physicochemical parameters during pasturma processing. Meat Science 82: 17-23.
- Kaban, G., 2013. Sucuk and pastirma: Microbiological changes and formation of volatile compounds. Meat Science 95: 912-918.
- Lemon, D.W., 1975. An improved TBA test for rancidity new series circular, No: 21. Halifax-Loboratory, Halifax, Nova, Scotia.
- Lorenzo, J.M., and Carballo, J., 2015. Changes in physico-chemical properties and volatile compounds throughout the manufacturing process of dry-cured foal loin. Meat Science 99: 44-51.
- Lorenzo, J.M., García Fontán, M., Franco, I., and Carballo, J., 2008. Biochemical characteristics of dry-cured laco'n (a Spanish traditional meat product) throughout the manufacture, and sensorial properties of the final product. Effect of some additives. Food Control 19: 1148-1158.
- Martínez-Onandi, N., Rivas-Cañedo, A., Ávila, M., Garde, S., Nuñez, M., and Picon, A., 2017. Influence of physicochemical characteristics and high pressure processing on the volatile fraction of iberian dry-cured ham. Meat Science 131: 40-47.
- Tekinşen, O.C., and Doğruer, Y., 2000. Her yönü ile pastırma, Selçuk Üniversitesi Basımevi, Konya.
- Toldrá, F., and Flores, M., 1998. The role of muscle proteases and lipases in flavor development during the processing of dry-cured ham. Critical Reviews in Food Science 38(4): 331-352.



# Effect of Protein Amount and the Foaming Conditions on the Stability of Carrot Juice Foams

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**Abstract:** Foams are the colloidal suspensions which are composed of gas entrapped in a liquid continuous phase. Forming of stable food foams is particularly important in foam-mat drying process because the stable foam structure reduces drying time and improves the final product quality. Food foams are generated through rigorous mixing of a solution that involves protein sources for a short period of time, but stabilizers are generally employed for improving the foam stability.

In this study, two animal protein sources namely egg albumen (A) and whey protein isolate (WPI) are employed for producing carrot juice foams. Whipping time and the foam ingredient composition (amount of A and WPI) were selected as independent variables of a full factorial central composite design to elaborate the stability of carrot juice foams. The results were discussed on basis of density and overrun of the foams in order to determine the optimum formulation and foam mixing condition.

Depending on the results, the mixing period had no significant effect individually; however, its interaction with WPI was significantly effective on density and overrun (p<0.05). The ANOVA of density and overrun responds presented that the linear, square and interaction terms of regression analysis was significant (p<0.05). Also increasing amount of albumen decreased the density and increased the overrun that supports the argument of improved the physical stability of the foams. The microscopic evaluation of the foams confirmed these results.

Keywords: Foam, protein, stability, carrot.

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# Determination of the Differences in the Composition of Erzincan Tulum Cheeses Produced from Raw and Pasteurized Sheep's Milks During Ripening

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**Abstract:** In this research; Erzincan Tulum cheese samples, from Akkaraman sheep's raw and pasteurized milk, were produced in the Dumlu (Erzurum, Turkey) plateaus with the help of the Şavak who are the original producers of cheese. The differences of cheese samples produced in duplicate by using commercial liquid rennet from raw and pasteurized milk were revealed during ripening. Produced cheeses were filled into the plastic barrels (2 kg), physical and chemical properties were examined in monthly periods and were ripened in a refrigerator at  $4 \pm 1$  °C for 90 days. It was determined that the milk variety had statistically significant effect on some components (P <0.05). In the cheese sample (P) produced from pasteurized milk, the higher dry matter, fat, fat in dry matter, total nitrogen, total protein, ash, salt, salt in dry matter amounts were determined according to the cheese sample ( $\zeta$ ) produced from raw milk. However, the pH and % acidity values in the P sample were found to be lower than the values of the  $\zeta$  sample. In cheese samples, except for pH, examined all properties (dry matter, fat, fat in dry matter, total nitrogen, total protein, ash, salt in dry matter, % acidity) increased continuously during ripening. The ripening period has been statistically significant the effect on the other components except for the fat in dry matter in cheeses (P<0.05). As a result, it was emerged that the milk variety and the ripening period affected the composition of Erzincan Tulum cheeses.

Keywords: Erzincan Tulum cheese, ripening period, commercial liquid rennet, pasteurisation.

# 1. INTRODUCTION

There are more than 100 varieties of cheese in Turkey, but three of these varieties (Beyaz Kaşar and Tulum cheeses, especially Erzincan Tulum-Şavak cheese) are the most popular (Çakmakçı, 2011; Cakir et al., 2016). Erzincan Tulum cheese, which is the best known among the Tulum cheeses, is the third most commonly produced cheese in Turkey (Cakir and Cakmakci, 2018). Erzincan Tulum cheese (Tulum cheese) has been produced in plateaus around Erzincan, Erzurum, Tunceli, Bingöl and Elazığ and in the East Anatolian regions by the Şavak tribe using Akkaraman sheep milk between May and September. The cheese was originally ripened in Tulum caves for 3 months or the period could extend up to one year (Cakir et al., 2016). In addition to; be convenient for exportation, having high nutritional value and being more expensive than butter increase the economic importance of Tulum cheese (Cakir and Cakmakci, 2018).

In this study, the effects of milk type and ripening period upon some components of Erzincan Tulum cheeses, which is produced from raw and pasteurized Akkaraman sheep's milk using the commercial liquid rennet and ripened in plastic barrels were investigated during the ripening period. For this purpose, just after the production the plastic barrels were packaged and the Tulum cheeses stored at  $4\pm1^{\circ}$ C were tested during the ripening period at the 1st 30th, 60th and 90th days.

# 2. MATERIALS AND METHODS

# Materials

Raw and pasteurized Akkaraman sheep's milk, salt and commercial liquid rennet used in the production of Erzincan Tulum cheese were obtained from Şavak tribe who is producing Tulum cheese and coming to Tortum (Dumlu) plateaus (Erzurum, Turkey) from Elazig. The plastic barrels (2kg capacity) used in the packaging of cheese were provided from Çeloğlu Inc. The packaging and production of Tulum cheese were carried out in Elazig.

# Sampling

The Tulum cheeses were stored in a refrigerator  $(4 \pm {}^{\circ}C)$  and were sampled from the same plastic barrels (2 kg capacity) at each sampling time (1, 30, 60, 90 d), but before taking samples the surface section of the cheese samples (approximately 0.5 to 1.0 cm) was discarded. After each sampling time, the surfaces of the Tulum cheeses were covered with a nylon material after sampling, and the Tulum cheeses were stored again for more sampling within the plastic barrels.

# **Cheese-Making**

Erzincan Tulum cheeses (Tulum cheeses) were made from raw and pasteurized Akkaraman sheep's milk in Tortum (Dumlu) plateau (Erzurum, Turkey). The commercial liquid rennet was added, and coagulation took place at 35°C. After, the coagulum was cut into pieces (approximately 1 cm cubes) and transferred into cotton bags for whey drainage. Drainage was carried out at approximately 20°C for 24 h, and the curd was pressed by piling the cotton bags on top of each other, with regular turning. Stacking of the curd at this temperature allows curd acidity (pH drops below 6.0) to develop and increases the removal of whey. Following this step, the curd was broken into pea-size pieces by hand and salted (3.0%, wt/wt), kneaded, and transferred into the bags. The bags were piled on top of each other and turned regularly for 10 d. Afterward, the plastic barrels were tightly filled with curd at dairy, and the packaged samples were ripened at 4°C for 90 d. Cheese making was made in duplicate, and cheese samples were analyzed for every 30 d.

# **Gross Chemical Composition**

The cheese samples were analysed during the ripening period at the 1st, 30th, 60th, and 90th days. Dry matter was investigated by the oven-drying method at 105 °C (IDF, 1982), fat content by the method of Gerber, and ash and salt contents according to the methods described by Kurt et al., (2007). Titratable acidity (lactic acid, %) was measured as suggested by AOAC, (1995). To measure pH, a grated Tulum cheese sample (10 g) was mixed uniformly with distilled water (15 mL), and the pH of the resultant slurry was measured using a digital pH meter (pH 211, Microprocessor pH Meter, Hanna Inst., Italy). Tulum cheese samples were investigated in duplicate for determination of total nitrogen (TN) by the micro-Kjeldahl method (IDF, 1993), and the total protein content was calculated (TN  $\% \times 6.38$ ).

# 3. RESULTS AND DISCUSSION

In this study, the milk variety determined to have statistically a significant effect on some components of Erzincan Tulum cheese (P < 0.05) (Table 1 and 2). According to this, the P Tulum cheese sample produced from pasteurized Akkaraman sheep's milk has higher dry matter, fat, fat in dry matter, total nitrogen, total protein, ash, salt, salt in dry matter values compared to the Ç Tulum cheese sample produced from raw Akkaraman sheep's milk. However, The P Tulum cheese sample produced from pasteurized Akkaraman sheep's milk has lower pH and % acidity values compared to the Ç Tulum cheese sample produced from raw Akkaraman sheep's milk (Table 1). In the study made by Sert et al., (2014), similarly, the Tulum cheese produced from pasteurized milk has higher dry matter, fat, fat in dry matter values compared to the Tulum cheese produced from raw milk. In the study made by Sert et al., (2014), similarly, the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has higher % acidity values compared to the Tulum cheese produced from raw milk has lower pH values compared to the Tulum cheese produced from raw milk has lower pH values compared to the Tulum cheese produced from raw milk has lower pH values compared to the Tulum cheese produced from p

The dry matter, fat, fat in dry matter, total nitrogen, total protein, ash, salt, salt in dry matter, % acidity values of Erzincan Tulum cheese samples showed continuous an increase during the ripening period (until the end of ripening) (Table 1). The ripening period has statistically a significant effect on the other components except for the fat in dry matter of Erzincan Tulum cheese samples (P<0.05) (Table 1 and 2). In the study made by Cakir and Cakmakci, (2018), similar increases were also determined during the ripening period.

# 4. CONCLUSION

In this study; due to increase the dry matter in milk of pasteurization process, the Tulum cheese sample produced from pasteurized milk thought to have higher dry matter, fat, fat in dry matter, total nitrogen, total protein, ash, salt and salt in dry matter values compared to the Tulum cheese sample produced from raw milk. In this study, it can be said that the dry matter amounts increased in Tulum cheese samples due to moisture loss during the ripening period. Depending on increase in the dry matter amounts of Tulum cheese samples; fat, fat in dry matter, total nitrogen, total protein, ash, salt and salt in dry matter values of Tulum cheese samples increased during the ripening period.

| Type of cheese    | <b>Ripening time</b> | Dry matter,                | Eat 9/                       | Fat in dry          | Total nitrogen,              | Total protein,              | Ach 0/                    | Salt, %                   | Salt in dry<br>matter, %  | <b>II</b>                     | A aidita 0/                     |
|-------------------|----------------------|----------------------------|------------------------------|---------------------|------------------------------|-----------------------------|---------------------------|---------------------------|---------------------------|-------------------------------|---------------------------------|
| (Treatment)       | (Days)               | %                          | Гац, 70                      | matter, %           | %                            | %                           | ASII, 70                  |                           |                           | рп                            | Actuity, 76                     |
|                   | 1                    | 55.51±0.06                 | 30.25±0.00                   | 54.50±0.06          | 3.025±0.016                  | 19.30±0.10                  | $4.43 \pm 0.04$           | $3.04 \pm 0.00$           | 5.48±0.01                 | $5.02 \pm 0.01$               | 0.981±0.013                     |
|                   | 30                   | 55.71±0.19                 | $30.38{\pm}0.18$             | 54.53±0.13          | $3.050 \pm 0.012$            | $19.46 \pm 0.08$            | $4.48{\pm}0.04$           | $3.07 \pm 0.04$           | $5.51 \pm 0.06$           | $4.89{\pm}0.01$               | $1.161 \pm 0.013$               |
| Ç                 | 60                   | $55.96{\pm}0.09$           | 30.63±0.18                   | 54.73±0.23          | $3.066 {\pm} 0.004$          | $19.56{\pm}0.02$            | $4.52 \pm 0.02$           | $3.10{\pm}0.00$           | $5.54{\pm}0.01$           | $4.85 \pm 0.01$               | $1.278 {\pm} 0.025$             |
|                   | 90                   | $56.18{\pm}0.03$           | $30.75 \pm 0.00$             | $54.74{\pm}0.02$    | $3.091 {\pm} 0.006$          | 19.73±0.04                  | $4.56 \pm 0.04$           | 3.13±0.04                 | $5.57 \pm 0.07$           | $5.02 \pm 0.04$               | $1.395 {\pm} 0.013$             |
|                   | Average              | 55.84±0.28 <b>a</b>        | 30.50±0.23 <b>a</b>          | 54.62±0.15 <b>a</b> | 3.058±0.027 <b>a</b>         | 19.51±0.17 <b>a</b>         | 4.49±0.06 <b>a</b>        | 3.09±0.04 <b>a</b>        | 5.52±0.05 <b>a</b>        | 4.94±0.09 <b>a</b>            | 1.204±0.164 <b>a</b>            |
|                   | 1                    | $56.96 \pm 0.05$           | 31.13±0.18                   | 54.65±0.26          | $3.138 {\pm} 0.008$          | 20.02±0.05                  | 4.63±0.04                 | 3.13±0.04                 | 5.50±0.06                 | 4.66±0.04                     | 0.963±0.013                     |
|                   | 30                   | $57.15 \pm 0.02$           | $31.25 \pm 0.00$             | 54.69±0.02          | $3.151 {\pm} 0.006$          | $20.11 \pm 0.04$            | $4.67 \pm 0.03$           | $3.16 \pm 0.00$           | $5.53 \pm 0.00$           | $4.79 \pm 0.00$               | $1.062 \pm 0.025$               |
| Р                 | 60                   | $57.31 \pm 0.06$           | $31.38{\pm}0.18$             | 54.76±0.25          | $3.169 \pm 0.012$            | $20.22 \pm 0.08$            | $4.71 \pm 0.04$           | $3.19{\pm}0.04$           | $5.57 \pm 0.06$           | 4.69±0.01                     | $1.161 \pm 0.013$               |
|                   | 90                   | $57.66 \pm 0.25$           | $31.63{\pm}0.18$             | $54.85{\pm}0.07$    | $3.186{\pm}0.010$            | 20.33±0.06                  | $4.76 \pm 0.05$           | $3.22 \pm 0.00$           | $5.59{\pm}0.02$           | 4.63±0.01                     | $1.278 \pm 0.025$               |
|                   | Average              | 57.27±0.29 <b>b</b>        | $31.34{\pm}0.23\textbf{b}$   | 54.73±0.16 <b>a</b> | $3.161 \pm 0.021 \mathbf{b}$ | $20.17{\pm}0.13\textbf{b}$  | $4.69{\pm}0.06\textbf{b}$ | $3.18{\pm}0.04\mathbf{b}$ | 5.54±0.05 <b>a</b>        | $4.69{\pm}0.07\textbf{b}$     | 1.116±0.126 <b>b</b>            |
|                   | 1                    | 56.23±0.84 <b>a</b>        | 30.69±0.52 <b>a</b>          | 54.57±0.18 <b>a</b> | 3.081±0.066 <b>a</b>         | 19.66±0.42 <b>a</b>         | 4.53±0.12 <b>a</b>        | 3.09±0.06 <b>a</b>        | 5.49±0.04 <b>a</b>        | 4.84±0.21 <b>a</b>            | 0.972±0.015 <b>a</b>            |
| Treatment average | 30                   | $56.43{\pm}0.84\mathbf{a}$ | $30.81{\pm}0.52 \textbf{ab}$ | 54.61±0.12 <b>a</b> | $3.100{\pm}0.059\mathbf{b}$  | $19.78{\pm}0.38\textbf{b}$  | 4.57±0.12 <b>ab</b>       | $3.12{\pm}0.06ab$         | 5.52±0.03 <b>ab</b>       | 4.84±0.06 <b>a</b>            | 1.112±0.059 <b>b</b>            |
|                   | 60                   | $56.63{\pm}0.78\textbf{b}$ | $31.00{\pm}0.46\textbf{b}$   | 54.74±0.19 <b>a</b> | 3.117±0.060 <b>c</b>         | 19.89±0.38 <b>c</b>         | $4.61{\pm}0.11\textbf{b}$ | $3.15{\pm}0.06\mathbf{b}$ | 5.55±0.04 <b>ab</b>       | $4.77{\pm}0.09\boldsymbol{b}$ | 1.220±0.070 <b>c</b>            |
|                   | 90                   | $56.92{\pm}0.86\mathbf{c}$ | 31.19±0.52 <b>c</b>          | 54.79±0.08 <b>a</b> | $3.139{\pm}0.055 d$          | $20.03{\pm}0.35 \textbf{d}$ | 4.66±0.12 <b>c</b>        | $3.18{\pm}0.06\mathbf{c}$ | $5.58{\pm}0.04\mathbf{b}$ | 4.82±0.23 <b>a</b>            | $1.337{\pm}0.070\boldsymbol{d}$ |
|                   | Average              | 56.55±0.79                 | 30.92±0.49                   | 54.68±0.16          | $3.109 \pm 0.060$            | 19.84±0.37                  | 4.59±0.12                 | 3.13±0.06                 | $5.53 {\pm} 0.05$         | 4.82±0.15                     | $1.160 \pm 0.148$               |

Table 1. The changes in some components of Erzincan Tulum cheese samples during the ripening period

C: The tulum cheese sample produced from raw Akkaraman sheep's milk; P: The tulum cheese sample produced from pasteurized Akkaraman sheep's milk. \*Averages of the same column values (each section separately) by the same letter did not differ significantly from Duncan's multiple range tests at 5% significance.

(a-d) Mean  $\pm$  SD, values followed by the same letters within a column are significantly different at P<0.05.

Table 2. The variance analysis results of some components of Erzincan Tulum cheese samples during the ripening period

| Resources of variation | df | Dry matter (F) | Fat (F)     | Fat in dry matter (F) | Total nitrogen (F) | Total protein (F) | Ash (F)   | Salt (F)    | Salt in dry matter (F) | pH (F)    | Acidity (F) |
|------------------------|----|----------------|-------------|-----------------------|--------------------|-------------------|-----------|-------------|------------------------|-----------|-------------|
| Treatment (T)          | 1  | 553.815**      | 145.800**   | 1.878                 | 443.196**          | 439.593**         | 111.534** | 36.000**    | 0.753                  | 597.232** | 89.471**    |
| Ripening period (R)    | 3  | 23.466**       | $9.800^{*}$ | 1.726                 | 24.795**           | 24.878**          | 8.837*    | $6.667^{*}$ | 3.031                  | 11.029**  | 280.373**   |
| TxR                    | 3  | 0.202          | 0.200       | 0.136                 | 0.545              | 0.567             | 0.029     | 0.000       | 0.008                  | 51.261**  | $6.490^{*}$ |
| Error                  | 8  |                |             |                       |                    |                   |           |             |                        |           |             |
| General                | 16 |                |             |                       |                    |                   |           |             |                        |           |             |

\*P<0.05, \*\*P<0.01 significance levels

### REFERENCES

AOAC, 1995. Official Methods of Analysis, Vol. 2., 16th ed. AOAC International, Arlington, VA, pp. 503-515.

- Cakir, Y., Cakmakci, S., and Hayaloglu, A. A., 2016. The effect of addition of black cumin (Nigella sativa L.) and ripening period on proteolysis, sensory properties and volatile profiles of Erzincan Tulum (Şavak) cheese made from raw Akkaraman sheep's milk. Small Ruminant Research, 134: 65-73.
- Cakir, Y., and Cakmakci, S., 2018. Some microbiological, physicochemical and ripening properties of Erzincan Tulum cheese produced with added black cumin (Nigella sativa L.). Journal of Food Science and Technology, 55(4): 1435-1443.
- Çakmakçı, S., 2011. Türkiye Peynirleri (19. Bölüm: 585-614). In: Peynir Biliminin Temelleri. (Editörler: A. A. Hayaloğlu ve B. Özer), ISBN: 978-605-87976-1-1, SİDAS Medya Ltd. Şti., İzmir, 643 s.
- International Dairy Federation (IDF), 1993. Determination of the nitrogen content (Kjeldahl method) and calculation of the crude protein content. In: Standard Method 20B: Milk, IDF, Brussels, Belgium.
- IDF, 1982. Determination of the total solid content (cheese and processed cheese). In: IDF Standard 4A. IDF, Brussels, Belgium.
- Kurt, A., Cakmakci, S., and Caglar, A., 2007. Süt ve Mamülleri Muayene Analiz Metotları Rehberi. Atatürk Universitesi Ziraat Fakültesi Yayınları No: 257, 398 p, Erzurum, Turkey (In Turkish).
- Sert, D., Akin, N., and Aktumsek, A., 2014. Lipolysis in Tulum cheese produced from raw and pasteurized goats' milk during ripening. Small Ruminant Research, 121: 351-360.



# Effect of Cryoprotectant on the Viability of *Lactobacillus brevis* ED25 during Freezing and Freeze Drying and Stability of Freeze Dried Cells under Accelerated Storage Conditions

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**Abstract:** This study investigated the survival of *Lactobacillus brevis* ED25, isolated from Turkish sourdough, after freezing and freeze drying using various kinds of cryoprotectants (skim milk, lactose and sucrose) formulations. A three-level-three-factor Box-Behnken experimental design was used to determine optimum cryoprotectant formulation and the highest cell survival during freezing and freeze-drying was observed with the 17.28% skim milk, 2.12% lactose and 10% sucrose cryoprotectant as the optimum condition with the desirability of these conditions was 0.971 which considered as the highest value. Importantly, no significant difference between the experimental and predicted values was observed revealing model adequacy. The structural and physicochemical properties of the freeze dried *L. brevis* ED25 powder produced at the optimized conditions were acceptable for application with regards to particle surface morphology, moisture and a<sub>w</sub>, glass transition temperature, FTIR spectra, X-ray structure, and also storage stability under the refrigeration and room temperature conditions. The storage stability studies demonstrated that the viability of freeze dried cells was quite stable during storage at 4 °C because the viability loss of 0.14 and 1.17 log units was determined during storage at 4 °C and 25 °C, respectively. Accelerated storage tests using temperatures of 50, 60 and 70 °C were used to develop a model system in order to predict the viability of freeze-dried cells. Accelerated storage test based on Arrhenius equation could be used to predict the shelf life of freeze dried *L. brevis* ED25 in optimum cryoprotectant formulation but only with a certain degree of predictability for long-term storage.

Keywords: Lactic acid bacteria, freeze drying, Box-Behnken experimental design, storage stability, cryoprotectants





# POSTER PRESENTATION

# Marine-derived Poly-oligosaccharides as Prebiotics

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Abstract: The marine polysaccharides have been described as emerging prebiotics due to their stimulating gut microbiota fermentation via colonic microflora, selective stimulation of the growth of colonic bifidobacteria, enhancing immune response through production of short chain fatty acids (SCFA) and potential substitutes for antibiotics. Based on current literature data, we summarized the prebiotic properties of poly-oligosaccharides extracting from marine sources. We also exhibited the content of dietary fiber in marine algae ( $\approx 33 - 75$  % DW as prebiotics) and the distribution of soluble and insoluble dietary fiber contents of red, brown and green algae, this content of dietary fiber in the marine algae, varies depending on the season, age, species, and habitat of algae. Furthermore, this work emphasizes the recent advances in the microwave-assisted, ultrasound-assisted extraction and enzymatic production of prebiotic polychemical, oligosaccharides and by this means it has been intended mention that as it generates high yield and favorable biological properties the enzyme-assisted extraction is considered as one of the best ones. Because of numerous benefits of seaweedderived polysaccharides and their important bioactive molecules such as  $\omega$ -3 fatty acids, essential amino acids, and vitamins A, B, C, and E, they can be applied in many fields such as the dietary supplementation, food functional, drug, cosmetic, and food industries. However, other investigations are needed to modify or improve the activities of the carbohydrates extraction from seaweeds that can be considered suitable and safe for foodstuffs, environment-friendly and cost-effective. Further clinical trials, must be carried out in vivo, will be mandatory to achieve definite evidence of the preventive and curative role of marine-derived prebiotics.

Keywords: Marine algae, prebiotics, polysaccharides, oligosaccharides, seaweeds

# **1. INTRODUCTION**

Prebiotics are usually characterized as nondigestible polysaccharides and oligosaccharides that stimulate the growth of beneficial intestinal bacteria within the gastrointestinal tract (GIT), exert growth – promoting, health improving effects (Vidanarachchi et al., 2009) and apply antagonism to pathogenic bacteria by limiting their proliferation that confers advantages upon host health (Roberfroid et al., 2008). A combination of prebiotics and probiotic is also called a synbiotic. Seaweeds and marine microalgae are one of the most critical sources of polysaccharides or oligosaccharides resultants thereof that are not decomposed by the enzymes of the upper part of GIT unique biochemical and fermenting characteristics (Francisco et al., 2017). Polysaccharides and oligosaccharides algae derivatives showed a vast spectrum of bioactive properties such as virucidal, antibacterial, antifungal, anti-inflammatory, immunomodulatory, anticoagulant or antithrombotic, antiproliferative, tumour suppressor, apoptotic, antilipidemic, hypoglycaemic, hypotensive, antiaging and antioxidant activities (Wijesekara et al., 2011, Koneva, 2009, Raposo et al., 2016).

# **Marine-Derived Polysaccharides as Prebiotics**

Due to their high polysaccharide content seaweed (marine macroalgae) can be utilized as prebiotic functional ingredients (O'Sullivan et al., 2010). This polysaccharide found in the cell wall of seaweeds which are not decomposed by the enzymes of the upper GIT and serve as an essential source of dietary fibers is paid significant attention to use as prebiotics. Table (1) shows the main marine-derived polysaccharides and their molecular weight.

Table 1. Marine-derived polysaccharides and their molecular weight

| Source                  | Polysaccharide     | Molecular Weight (kDa) | References |
|-------------------------|--------------------|------------------------|------------|
| Red algae               |                    |                        |            |
| Furcellaria lumbricalis | Sulphated galactan | 290                    | (22)       |
| Furcellaria lumbricalis | Sulphated galactan | 428                    | (2)        |
| Hypnea musciformis      | Sulphated galactan | 147–155                | (18)       |
| Kappaphycus alvarezii   | Carrageenan        | 390                    | (1)        |
| Mastocarpus stellatus   | Carrageenan        | 1248                   | (8)        |
| Brown algae             |                    |                        |            |
| Macrocystis pyrifera    | Alginate           | 297                    | (3)        |
| Sargassum fusiforme     | Laminarin          | 27.6                   | (32)       |
| Cystoseira barbata      | Alginate           | 204                    | (27)       |
| Cystoseira compressa    | Fucoidan           | 545                    | (10)       |
| Ascophyllum nodosum     | Fucoidan           | 40.23                  | (36)       |
| Laminaria hyperborea    | Laminarin          | 3242–5052              | (29)       |
| Saccharina cichorioides | Fucoidan           | 30                     | (28)       |
| Green algae             |                    |                        |            |
| Ulva armoricana         | Ulvan              | 140–500                | (13)       |

Besides the lack of lignin and a content of the polysaccharide of seaweeds is very high (20 - 76% of dry weight) (Wang et al., 2012), the carbohydrate fraction, in seaweeds includes polysaccharides such as cellulose and starch (25 - 50% DW, 30 - 60% DW in green and red, respectively), and cellulose and laminarin in Brown (30 - 50%) (Husemann, 1968). Marine algae are rich sources of dietary fiber. The composition and content of dietary fibers in the marine algae, varies depending on the season, age, species, and habitat of algae like fucoidans in carrageenans in Rhodophyceae (red algae), Phaeophyceae (brown algae), and ulvans in Chlorophyceae (green algae) with comparison the content of fiber in algae which is from 33 to 50 g per 100 g of wet weight it's higher than that in most fruits and vegetables. The content of dietary fiber in marine algae and the distribution of soluble and insoluble dietary fiber contents of red, brown and green algae shows in Fig.1.



Figure 1. Marine algae as prebiotics and the dietary fiber contents of red, brown and green algae (Daniela G et al., 2019)

Oligosaccharides obtained from alginate confirmed selective growth of bifido, lactobacteria, a decrease in the higher level of enterobacteria and enterococci in the feces and gut in Wistar rats (Wang et al., 2006). *Grateloupia filicina* and *Eucheuma spinosum* showed a strong prebiotics activity by promoting the growth of Bifidobacterium (Chen et al., 2018). The same notable growth of populations of lactobacilli and bifidobacteria in the intestine was shown for mice which consumed 2.5% agarose hydrolysate in their diet for a week (Hu, 2006). Sulfated polysaccharides from marine seaweeds have an anti-inflammatory and antimicrobial effect (such as H.pylori) (Shibata et al., 2003). Fucoidan from *F. Evanescens* showed a hypolipidemic effect and detoxifying action (Maistrovsky et al., 2009; Kuznetsova., 2009). Polysaccharides

isolated from *Grateloupia filicina* and *Eucheuma spinosum* could be utilized as prebiotics due to significant promotion in the bifidobacterium proliferation and 0.1% *E. spinosum* and 0.4% *G. filicina* resulted in the highest proliferation rates of beneficial bacteria (Chen., 2017). Dietary marine polysaccharides cannot degrade by gastric and intestinal enzymes, for this reason they are fermented by specific microbes in the gut in lower GIT and produce a significant amount of oligosaccharides and short-chain fatty acids and other metabolites which could be absorbed and exert a therapeutic effect in addition to stimulate the activity of a limited number of indigenous bacteria (Fig. 2) (Qingsen et al., 2018).



**Figure 2.** Gut microbiota degradation of marine polysaccharides and its effects on intestinal ecology. (Qingsen S., et al., 2018)

# **Marine-Poly-Oligosaccharides Extraction**

There are different methods employed in the extraction poly and oligosaccharides. These are briefly summarized below.

# **Chemical extraction**

This method includes two steps. The first one is pre-extracted by treatment seaweeds with ethanol, chloroform, or formaldehyde to remove proteins, pigments, terpenes, and the other compounds. The second is extraction the polysaccharides under heating and mild acid or water, calcium chloride or sodium carbonate used to precipitate and extract alginate from residual seaweeds. According to solubility the types of solvents should take attention. For example, brown seaweed is insoluble in polar solvents, ethanol and acetone are commonly used in the precipitation step (Chigozie et al., 2017).

## Microwave-assisted extraction (MAE)

Microwave-assisted extraction is approved as a faster and more effective method that may use to extract fucoidans (Rodriguez-Jasso., 2011), used shorter time (pressure of 120 psi for 1 min) and microwave heating to extract fucoidans from *F. Vesiculosis* and got comparable yields (18.2%) of fucoidan. Fucoidan was obtained from *F. Vesiculosis* using microwave heating in a shorter time (120 psi pressure for 1 minute) and a comparable yield (18.2%).

#### Ultrasound-assisted extraction (UAE)

To obtain localized pressure and heat which helps to extract polysaccharides the method uses sound waves migrating through a medium inducing pressure variations and creating small vacuum bubbles or voids which collapse violently. During the experiment, it was observed that the extraction with ultrasound-assisted technology (10.79%) gave better yield when compared to MAE (9.56%) and chemical extraction (4.67%) in lesser time (Kadam et al., 2015). UAE and MAE could be used high energy input and pressure required; however, they are not toxic.

## **Enzyme-assisted extraction**

This method has been described as one of the best as it generates high yield and favorable biological properties. To degrade cell, wall to release polysaccharides can use carbohydrases of desirable bioactivities at optimal temperature and pH like Viscozyme, pH 4.5), Celluclast, pH 4.5, 508C, Ultraflo, pH 7.0, 608C (Rodrigues et al., 2015).

# 2. CONCLUSION

Marine seaweeds have a wide range of applications in functional food products. Seaweeds represent demands grow for special biological activities with modulating the gut microbiota, beneficial effects, and potential medicinal values. In addition to their current status as a source of soluble and insoluble dietary fibers that approved to act as prebiotics. Regarding extraction methods, active research has been ongoing to modify or improve the activities of the carbohydrates extraction from seaweeds that can be considered suitable and safe for foodstuffs, environment-friendly and cost-effective.

# REFERENCES

- A.A. Kalitnik, A.O. Byankina Barabanova, V.P. Nagorskaya, A.V. Reunov, V.P. Glazunov, T.F. Solov'eva, I.M. Yermak, Low molecular weight derivatives of different carrageenan types and their antiviral activity, J. Appl. Phycol. 25 (2013) 65–72 https://doi.org/10.1007/s10811-012-9839-8.
- B. Yang, G. Yu, X. Zhao, W. Ren, G. Jiao, L. Fang, Y. Wang, G. Du, C. Tiller, G. Girouard, C.J. Barrow, S. Ewart, J. Zhang, Structural characterisation and bioactivities of hybrid carrageenan-like sulphated galactan from red alga Furcellaria lumbricalis, Food Chem. 124 (2011) 50–57 https://doi.org/10.1016/j. foodchem.2010.05.102.
- C.G. Gomez, M.V.P. Lambrecht, J.E. Lozano, M. Rinaudo, M.A. Villar, Influence of the extraction-purification conditions on fnal properties of alginates obtained from brown algae (Macrocystis pyrifera), Int. J. Biol. Macromol. 44 (2009) 365–371 https://doi.org/10.1016/j.ijbiomac.2009.02.005.
- 4. Chen X, Sun Y, Hu L, Liu S, Yu H, Li R, Wang X, Li P (2018) In vitro prebiotic effects of seaweed polysaccharides. J Oceanol Limno 36(3):926–932.
- CHEN, X., SUN Y., HU, L., LIU S., YU H., XING, R., LI, R., WANG X., LI P.,2017. In vitro prebiotic effects of seaweed polysaccharides. Journal of Oceanology and Limnology Vol. 36 No. 3, P. 926-932, 2018 https://doi.org/10.1007/s00343-018-6330-7.
- Chigozie L, O., Subin R. C. K. R., Chibuike C. U., Alberta N. A., Beth M. 2017. Prospects of brown seaweed polysaccharides (BSP) asprebiotics and potential immunomodulators. Wiley journal of food chemistry. DOI: 10.1111/jfbc.12392.
- Daniela de Borba Gurpilhares, Leonardo Paes Cinelli, Naomi Kato Simas, Adalberto Pessoa Jr., Lara Durães Sette.2019. Review: Marine prebiotics: Polysaccharides and oligosaccharides obtained by using microbial enzymes. Food Chemistry 280 (2019) 175–186.
- 8. E.Gomez-Ordóñez, A.Jiménez-Escrig, P. Rupérez, Bioactivity of sulfated polysaccharides from the edible red seaweed Mastocarpus stellatus, Bioact. Carbohydr. Diet. Fibre.3(2014) 29–40 https://doi.org/10.1016/j.bcdf.2014.01.002.
- 9. Francisco J. M., Nieves C., Antonia M., Mar V., Agusti'n O., 2017. Current state and latest advances in the concept, production, and functionality of prebiotics oligosaccharides. Current Opinion in Food Science 13:50–55.
- H.H. Ammar, S. Lajili, R.B. Said, D. Le Cerf, A. Bouraoui, H. Majdoub, Physicochemical characterization and pharmacological evaluation of sulfated polysaccharides from three species of Mediterranean brown algae of the genus Cystoseira, DARU J. Pharm. Sci. 23 (1) (2015), https://doi.org/10.1186/s40199-015-0089-6.
- 11. Hu, B., Gong, Q.N., Wang Y., et al., Prebiotic effects of neoagarooligosaccharides prepared by enzymatic hydrolysis of agarose, *Anaerobe*, 2006, vol. 12, pp. 260–266
- 12. Husemann, E., 1968. In: Percival, Von E., McDowell, R.H. (Eds.), Chemistry and Enzymology of Marine Algal Polysaccharides. 80. Academic Press, London-New York, pp. 856. 1967. 1. Aufl., XII, 219 S., mehrere Abb. u. Tab., geb. 60s. Angew. Chemie. https://doi.org/10.1002/ange.19680802022.

- K. Hardouin, G. Bedoux, A.S. Burlot, C. Donnay-Moreno, J.P. Bergé, P. NyvallCollén, N. Bourgougnon, Enzyme-assisted extraction (EAE) for the production of antiviral and antioxidant extracts from the green seaweed Ulva armoricana (Ulvales, Ulvophyceae), Algal Res 16 (2016) 233–239 https://doi.org/10.1016/j. Algal .2016.03.013.
- 14. Kadam, S. U., Tiwari, B. K., & O'donnell, C. P. (2015). Extraction, structure and biofunctional activities of laminarin from brown algae. International Journal of Food Science & Technology, 50, 24–31.
- Kannan M., Samuthirapandian R., Thirunavukkarasu M., Venkatachalam U., Ramachandran C., Palaniappan S., Ramu G, A., Durairaj K, R. Application of marine-derived polysaccharides as immunostimulants in aquaculture: A review of current knowledge and further perspectives. Fish and Shellfish Immunology 86 (2019) 1177–1193.
- 16. Koneva, E.L., Substantiation, and development of tech nologies for alginatecontaining functional products Extended Abstract of Cand. Sci. (Tech.) Dissertation, Vladivostok, 2009.
- 17. Kuznetsova, T.A., Correction of immunity and haemostasis disorders with biopolymers from marine hydrobionts (experimental and clinical aspects), *Extended Abstract of Cand. Sci. (Med.) Dissertation*, Moscow, 2009.
- M, Alves. C.M.P.G. Dore, A.J.G. Castro, M.S. do Nascimento, A.K.M. Cruz, E.M. Soriano, N.M.B. Benevides, E.L. Leite, Antioxidant, cytotoxic and hemolytic effects of sulfated galactans from edible red alga Hypnea musciformis, J. Appl. Phycol. 24 (2012) 1217–1227 https://doi.org/10.1007/s10811-011-9763-3.
- Maistrovsky, K.V., Zaporozhets, T.S., Fedyanina, L.N., et al., The effect of fucoidan immunomodulator from the brown algae *Fucus evanescens* on parameters of the antioxidant system and lipid and carbohydrate exchange in mice, *Tikhookean. Med. J.*, 2009, no. 3, pp. 97–100.
- 20. O'Sullivan L, Murphy B, McLoughlin P, Duggan P, Lawlor PG, Hughes H, Gardiner GE (2010) Prebiotics from marine macroalgae for human and animal health applications. Mar Drugs 8(7):2038–2064
- 21. Qingsen S., Hao J., Chao C., Jiejie H., Guoyun L., Guangli Y.,2018. Gut microbiota fermentation of marine polysaccharides and its effects on intestinal ecology: An overview. Carbohydrate Polymers 179 (2018) 173–185.
- R. Tuvikene, K. Truus, M. Robal, O. Volobujeva, E. Mellikov, T. Pehk, A. Kollist, T. Kailas, M. Vaher, The extraction, structure, and gelling properties of hybrid galactan from the red alga Furcellaria lumbricalis (Baltic Sea, Estonia), J. Appl. Phycol. 22 (2010) 51–63 https://doi.org/10.1007/s10811-009-9425-x.
- 23. Raposo MFJ, Bernardo de Morais AMM, Costa de Morais RMS: Emergent sources of prebiotics: seaweeds and microalgae. Mar Drugs 2016, 14:E27.
- 24. Roberfroid M, Gibson GR, Roberfroid M (2008) Prebiotics: concept, definition, criteria, methodologies, and products. In: Handbook of prebiotics. CRC Press, Boca Raton, pp 39–69.
- Rodrigues, D., Sousa, S., Silva, A., Amorim, M., Pereira, L., Rocha-Santos, T. A. P., . . . Freitas, A. C. (2015). Impact of enzyme- and ultrasound-assisted extraction methods on biological properties of red, brown, and green seaweeds from the Central West Coast of Portugal. Journal of Agricultural and Food Chemistry, 63, 3177–3188.
- 26. Rodriguez-Jasso, R. M., Mussatto, S. I., Pastrana, L., Aguilar, C. N., & Teixeira, J. A. (2011). Microwave-assisted extraction of sulfated polysaccharides (fucoidan) from brown seaweed. Carbohydrate Polymers, 86, 1137–1144.
- S. Sellimi, I. Younes, H.B. Ayed, H. Maalej, V. Montero, M. Rinaudo, M. Dahia, T. Mechichi, M. Hajji, M. Nasri, Structural, physicochemical and antioxidant properties of sodium alginate isolated from a Tunisian brown seaweed, Int. J. Biol. Macromol. 72 (2015) 1358–1367 https://doi.org/10.1016/j.ijbiomac.2014.10.016.
- S.D. Anastyuk, N.M. Shevchenko, R.V. Usoltseva Menshova, A.S. Silchenko, P.A. Zadorozhny, P.S. Dmitrenok, S.P. Ermakova, Structural features and anticancer activity in vitro of fucoidan derivatives from brown alga Saccharina cichorioides, Carbohydr. Polym. 157 (2017) 503–1510 https://doi.org/10.1016/j.carbpol.2016.11.031.
- S.U. Kadam, C.P. O'Donnell, D.K. Rai, M.B. Hossain, C.M. Burgess, D. Walsh, B.K. Tiwari, Laminarin from Irish brown seaweeds Ascophyllum nodosum and Laminaria Hyperborea: ultrasound-assisted extraction, characterization and bioactivity, Mar. Drugs 13 (2015) 4270–4280, https://doi.org/10.3390/md13074270.

- Shibata, H., Iimuro, M., Uchiya, N., et al., Preventive effects of *Cladosiphon* fucoidan against *Helicobacter pylori* infection in Mongolian gerbils, *Helicobacter*, 2003, vol. 8, pp. 59–65.
- Vidanarachchi JK, Iji PA, Mikkelsen LL, Sims I, Choct M (2009) Isolation and characterization of water-soluble prebiotic compounds from Australian and New Zealand plants. Carbohydr Polym 77:670–676. https://doi. org/10.1016/j.carbpol.2009.02.009.
- 32. W. Jin, W. Zhang, J. Wang, S. Ren, N. Song, D. Duan, Q. Zhang, Characterization of laminarin and a highly sulfated polysaccharide from Sargassum fusiforme, Carbohydr. Res. 385 (2014) 58–64 https://doi.org/10.1016/j.carres.2013.12.009.
- 33. Wang, W., Wang, S.-X., & Guan, H.-S. (2012). The antiviral activities and mechanisms of marine polysaccharides: An overview. Marine Drugs, 10(12), 2795–2816.
- 34. Wang, Y., Han, F., Hu, B., et al., *In vivo* prebiotic properties of alginate oligosaccharides prepared through enzymatic hydrolysis of alginate, *Nutr. Res.*, 2006, vol. 8, pp. 597–608.
- 35. Wijesekara I, Pangestuti R, Kim SK (2011) Biological activities and potential health benefits of sulfated polysaccharides derived from marine algae. Carbohydr Polym 84(1):14–21. https://doi.org/10.1016/j. carbpol.2010.10.062.
- 36. Y. Yuan, D. Macquarrie, Microwave-assisted extraction of sulfated polysaccharides (fucoidan) from Ascophyllum nodosum and its antioxidant activity, Carbohydr. Polym. 129 (2015) 101–107 https://doi.org/10.1016/j.carbpol.2015.04.057.





# POSTER PRESENTATION

# Efficacy and Safety Considerations of Pre-probiotics

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Abstract: Probiotics, defined as live microorganisms with beneficially effect on the intestinal microflora, whereas a prebiotic, is a non-viable food component that confers a health benefit on the host. The applications based on this health benefit of pre-probiotics have been expanding in the last decades. Within this scope, we will focus mainly on three viewpoints of pre-probiotics in this review: Consumer health consideration, the safety perspectives of probiotics, finally the commonly used probiotics products and their characteristics. While numerous studies have reported that pre-probiotics have clinical effectiveness in-maintaining the balance of gastrointestinal microbiota to improve health promoting functions like prevents intestinal tract infections, improves lactose metabolism, reduces serum cholesterol level, enhance immunity, detoxify the ingested carcinogens, reducing in the bowel syndrome and inflammatory bowel disease, a critical emphasize is still made with respect the safety considerations and potential risks of probiotics including systemic infections, gastrointestinal side effects, deleterious metabolic activities, excessive immune stimulation in susceptible individuals and gene transfer. Based on current literature data, and unlike most researches we focused on efficacy and safety of probiotics specifically horizontal transfer of antibiotic resistance genes from probiotics to pathogenic gut bacteria that might lead to the development of antibiotic resistant pathogens and the ability of some probiotic microorganisms to cross the intestinal mucosa and enter into bloodstream causing systemic infections. Further research is needed in the case of human probiotics. The safety requirements could be more rigorous, considering the necessities for demarcating the regulation of probiotics for the general dietary and toward clinical interventions.

Keywords: Bifidobacteria; Lactobacilli; prebiotics; probiotics; micro-organisms; antibiotic resistance

# **1. INTRODUCTION**

The expression "probiotic" was probably first defined by Kollath in 1953 (Danuta K-K.., 2012). Probiotics ("pro" (in favor) and "biotic" (life)) are live microorganisms with beneficially effect on the intestinal microflora, the definition restricted the use of the word probiotic to products which contain live microorganisms and indicate the needed to provide an adequate dose of probiotic bacteria in order to exert the desirable effects (Joint FAO/WHO., 2001). A prebiotic is a non-viable food component that confers a health benefit on the host associated with modulation of the microbiota according to the FAO/WHO, Prebiotic should neither be hydrolyzed nor absorbed by mammalian enzymes or tissues, selectively enrich for one or a limited number of beneficial bacteria, beneficially alter the intestinal microbiota and their activities and beneficially alter luminal or systemic aspects of the host defense system (Simmering R., 2001). Commonly used prebiotics." This combination of prebiotics show therapeutic effects against diseases, like gastrointestinal diseases, respiratory infections hypercholesterolemia, atopic dermatitis, allergy, diabetes, liver diseases and cancer (Debapriya, M, et al., 2018). The concept of synbiotics has been proposed recently to characterize colonic foods with prebiotic and probiotic properties as health-enhancing functional foods (Gibson, G.R., 1995).

# **Main Pre-Probiotics**

The micro-organisms that are most commonly used as probiotics belong to the group of lactic acid bacteria (Lactobacillus) and Bifidobacterium(Juan J. S.D., 2017). The primary sources of probiotics and prebiotics appear in the Fig.1. Breast milk naturally contains prebiotics (oligosaccharides) at a level of 10-12 g/l (Rao et al., 2009). According to FAO " a prebiotic can be fiber, but a fiber need not be prebiotics."



Figure 1. Main Pre-probiotics (*a mainly applied in animals*).

# The Characteristics of Pre-Probiotics

The attributes of beneficial prebiotics included resistant to gastric acidity and hydrolysis by mammalian digestive enzymes, metabolized by the host in addition to have a beneficial effect to the host health, selectively stimulate the growth and or activity of health-promoting intestinal bacteria and probiotic strains to become metabolically active, carbohydrates comprising of a mixture of oligosaccharides/ and polysaccharides.

For a probiotic to be effective, it should survive the acidic environment of the stomach and transit through the bowel (Juan J. S.D., 2017). Beside the ability to survive gastric conditions, colonize the intestine, and adhere to intestinal epithelium according to the WHO/FAO definition, the main criterion for a probiotic strain should be the fact that it confers a health benefit on the host such as production of essential amino acids, antitumor activity, and vitamins and food protective activities (growth inhibition of food spoilage or poisoning bacteria (Danuta K-K., 2012). Probiotic should be of host origin, non-pathogenic, withstand processing and storage, resist gastric acid and bile, adhere to epithelium or mucous, persist in the intestinal tract, produce inhibitory compounds and modulate immune response (Simmering R., 2001).

# **The Beneficial Effects of Pre-Probiotics**

Prebiotics are usually characterized as nondigestible polysaccharides and oligosaccharides that stimulate gut microbiota fermentation via colonic microflora, selective stimulation of the growth of colonic bifidobacteria within the gastrointestinal tract (GIT), exert growth – promoting, health improving effects, enhancing immune response through production of short chain fatty acids (SCFA) and potential substitutes for antibiotics (Vidanarachchi et al., 2009) and apply antagonism to pathogenic bacteria by limiting their proliferation that confers advantages upon host health (Roberfroid et al., 2008).

Probiotics, nutraceutical, and functional foods belong to such diet category. Probiotics are selective viable microorganisms administered in an adequate amount to confer health benefits beyond inherent general nutrition. According to the consumer demand that has changed from energy providing diet to the diet with a balanced nutrient profile along with metabolic, physiological, health and functional benefits. These microorganisms have various health-promoting functions like prevents intestinal tract infections, improves lactose metabolism, reduces serum cholesterol level, enhance immunity, stimulates calcium absorption, improves protein digestibility, synthesis of vitamins (vitamin B, nicotinic acid and folic acid), and counteracts the effects of food-borne pathogens (Anil P., 2018). Examples of possible probiotic mechanisms of action, in the control of intestinal pathogens include Antimicrobial substance production, Competitive exclusion of pathogen binding, Competition for nutrients and Modulation of the immune system (Joint FAO/WHO., 2001). The mechanisms of action from fermenting bacteria shown in (Fig.2).



Figure 2. The mechanisms of action from fermenting bacteria.

The beneficial effects of probiotics on GI function are attributed to normalization of permeability, restoration of the microbiota, improvement of barrier immune function, down-regulation of the pro-inflammatory immune response and rebalancing of pro-inflammatory and anti-inflammatory cytokines, those on which probiotics have possible beneficial effects include chronic idiopathic constipation (Girardin M., 2011). There is good evidence that specific strains of probiotics are safe for human use and able to confer some health benefits on the host. The health benefits for which probiotics can be applied include conditions such as gastrointestinal infections, certain bowel disorders, allergy, and urogenital infections.

Effect of pre-probiotics on health including, Hyper cholesteromia and cardiovascular disease: some strains of Probiotics approved to have positive effect in reducing the dietary cholesterol absorption such as *Enterococcus faecium*, Lactobacillus Plantarum, Lactobacillus spp., Bifidobacterium spp., Propionibacterium freudenreichii, Lactobacillus plantarum (Anil P., 2018). (AlSheraji et al., 2013) reported that fructo-oligosaccharides (FOS) has effective against hyperlipidaemia. Diarrhoea : Lb. rhamnosus, Lb. casei, Bf. lactis, Bf. Bifidum, Sc. Thermophilus, Lactobacillus casei observed by (Parvez, S. et al., 2006) to have positive competition with pathogenic bacteria on epithelial cells. S. boulardii appeared to provide significant protection in 'Travellers' diarrhea. (Passariello et al., 2011) reported that a zinc and prebiotics combination reduced the duration of diarrhea in patients by stimulating water and electrolyte re-absorption across gut mucosa and inhibited the pathogens. Lactobacillus rhamnosus GG and Bifidobacterium lactis have rotavirus infections in order to prevent diarrhea (Saavedra et al., 1994; Szajewska et al., 2001, Isolauri et al., 1991; Guarino et al., 1997; Majamaa et al., 1995; Shornikova et al., 1997; Perdone et al., 1999; Guandalini et al., 2000). Antibiotic therapy: Sanders et al. (2005) reported a positive effect of Lb. salivarius, Lb. acidophilus, Lb. johnsonii, Enterococcus mundtii, Lactobacillus plantarum, Lactobacillus brevis, Lactobacillus strains, Bifidobacterium strains, in minimizing the disruptive effect of antibiotics to normal bacterial flora. Kidney Stones: Lactobacillus RC-14, Lactobacillus GR-1, Lactobacillus B-54, approved to degrade or reduce the oxalate excretion (Sanders et al., 2005). Immunity: Some strains like Lb. Casei, Lb. rhamnosus, Lb. acidophilus, Bf. lactis, Bacillus circulans, Lactobacillus Plantarum can enhance the level of reactive immune cells (Sanders et al., 2005). Lactose intolerance: Lb. rhamnosus, Lb. Plantarum, Lb. delberukii, Bf. Lactis, Lactobacillus acidophulus can digest lactose. Probiotic bacteria containing  $\beta$ -galactosidase can be added to food to improve lactose maldigestion (Kim and Gilliland., 1983). Cancer: Bifidobacterium sp., Lb. casei, Lb. acidophilus, Propionibacterium sp. have positive in detoxify the ingested carcinogens (Sanders et al., 2005). In addition, the ability of some strians to produce carcinogens such as nitrosamines that help to prevent or delay the onset of certain cancers. (Hosada et al., 1996) theoretically admistrated the ability of lactobacilli and bifidobacteria to modify the flora in order to decreased  $\beta$ -glucuronidase and carcinogen levels, probiotics including L. casei Shirota instilled in intestinal observed to

reduce cancer recurrences at urinary bladder (Aso et al., 1995) also L. rhamnosus GG and bifidobacteria and L. Rhamnosus strains GG and LC-705 as well as Propionibacterium sp. decreased in availability of carcinogenic aflatoxin in the lumen was noticed by (El-Nezami et al., 2000; Oatley et al., 2000). Butyrate suppressed the expression of transcription factor NF-jB in HT-29 cell lines, in cancer patients (Macfarlane, Steed, and Macfarlane, 2008; Patel, and Goyal., 2012). Hypertension: Lb. Rhamnosus and Lb. Lactis reduce the blood pressure according to Sanders et al. (2005). Pancreatitis: Lb. rhamnosus GG and Bf. lactis BB-12 show decreasing in the occurrence of pancreatic infection (Pezzilli and Fantini., 2006). Tooth problems Lactobacillus: Bifidobacterium decreases teeth problems (Darwazeh and Darwazeh., 2011). Anemia: due to the production of propionic acid Lactobacillus lactobacilli increases the expression of iron transporters in the caecum (Balamurugan et al., 2010). Eczema: Eczema can be cured with probiotics such as Escherichia coli, Bifidobacterium bifidum, Bifidobacterium lactis and Lactococcus lactis (Anil P., 2018). Food allergies: Escherichia coli, Lactobacillus, Bifidobacterium improves the immunity of the body and reduces food allergies (Soh et al., 2009). As a means to prevent certain diseases and modulate host immunity L. rhamnosus GG noticed to decrease the risk of allergy in pregnant women for four weeks prior, newborns (Kalliomaki et al., 2001). and infants allergic to cow's milk (Majamaa and Isolauri., 1996; Isolauri et al., 2000). Nonalcoholic fatty liver disease (NAFLD) : probiotics have many of their effects have proved to be beneficial in NAFLD, including the modulation of the intestinal microbiota, an antibacterial substance production, an improved epithelial barrier function and reduced intestinal inflammation(Anna I., 2011). Inflammatory Bowel diseases: E.coli, Saccharomyces boulardii, Bifidobacterium longum, B. breve, B. infantis, Lactobacillus casei, L. plantarum, L. acidophilus, L. delbrueki subsp. bulgaricus and Streptococcus showed reducing in bowel syndrome and inflammatory bowel disease (Ventura and Perozzi ., 2011). L.plantarum has been used with beneficial effects in the handling of some symptoms in patients with Irritable bowel syndrome (IBS). L.rhamnosus GG and a mixture of probiotics consisting of L. acidophilus, Lactobacillus bulgaricus (L.bulgaricus), B.bifidum and Streptococcus thermophilus (S. thermophilus) also had a significant protective effect versus placebo(Juan J. S.D., 2017). Urinary tract infection: to use Lactobacillus rhamnosus, Lactobacillus reuteri, Lacidophilus urinary tract diseases problems are reduced (Anukam, K.C., 2009). Celiac Disease: therapeutic trials aimed at altering the intestinal microbiome have looked to probiotics as a possible treatment for CD, further research is required to understand the mechanism, and at this point, can't be recommended (S. Krishnareddy., 2017). Pouchitis: The probiotic mixtures (L. acidophilus, L. casei, L. plantarum, L. bulgaricus; B. longum, B. breve, B. infantis, and S. thermophilus) rilac, which contains L. acidophilus, L. delbrueckii ssp. Bulgaricus and B. bifidum showed Maintaining remission of chronic pouchitis and preventing the onset of pouchitis(Juan J. S.D., 2017). Antimicrobial: Studies indicated the activity of some probiotic bacteria against salmonella (Ogawa et al., 2001; Shu et al., 2000) and Helicobacter pylori, such as L. johnsonii La1(Midolo et al., 1995).

#### Safety Considerations and Potential Risks

(FAO/WHO .2002), mentioned that probiotics might theoretically be linked to four specific types of side effects in patients with underlying medical conditions: " (1) systemic infections; (2) deleterious metabolic activities; (3) excessive immune stimulation in susceptible individuals; and (4) gene transfer". Most researches have been focused on efficacy not the safety of probiotics in animal feed to enter the human food chain without considered the including potential for infection of animals or people, transfer of antibiotic resistance genes, and hyperstimulation of the immune system (FAO/WHO). More Research is needed to examine host-probiotic interaction and explore the mode of action to reach to understand the relationship between specific probiotic products and animal species at different ages and points in development and establish minimum required doses (FAO/WHO). Although LAB and Bifidobacteria are regarded as GRAS and have a long history of safe use However, extensive use of antibiotics for treatment and prophylaxis of microbial infections in humans has led to an increase of antibiotic resistance in bacteria, specifically horizontal transfer of antibiotic resistance genes from probiotics to pathogenic gut bacteria might lead to the development of antibiotic resistance plasmids from lactococcus species (Dessart SR., 1991), likewise conjugation transfer from enterococci to lactococci can take place in the gut of animals also in vitro; in the other side it is quite rare the transferring to lactobacilli (Morelli L., 1988). *Lactococcus lactis* has been reported to transfer these ABR genes to *Enterococcus faecalis* (Fig.3) (H. Aarts., 2014).



Figure 3. Probiotics and transfer of antibiotic resistance

Most studies assumed that normal inhabitants of the intestines of animals and humans, also in the environment and food *Enterococcus* genus should be in concerning due to frequent opportunistic pathogens known for their resistance to antibiotics and the ability to transfer genetic information. Some enterococcal strains have shown resistance to vancomycin is an antibiotic used to treat patients with infections caused by methicillin-resistant staphylococci (e.g., *E. faecium*) is acquired and plasmid-mediated (Courvalin., 2006), with the capacity to transfer this type of resistance to other bacterial species. (Danielsen and Wind., 2003) considered vancomycin resistance is an explicative feature of some Lactobacilli species. (Rosander et al., 2008) Demonstrated potentially transferable resistance traits for antibiotics tetracycline and lincomycin by *L. reuteri* ATCC 55730 a commercially available probiotic strain. Certain microorganisms may be problematic, particularly the *Streptococcus* and *Enterococci* (*E. faecium* and *E. faecalis*) (Arturo A., 2016. Salminen and von Wright., 1998).

Information acquired to date shows that lactobacilli have a long history of use as probiotics without established risk to humans, and this remains the best proof of their safety (Naidu et al., 1999). Also, no pathogenic or virulence properties have been found for lactobacilli, bifidobacteria or lactococci (Aguirre and Collins., 1993). However It is also acknowledged that some members of lactic acid bacteria, such as enterococci may possess virulence characteristics, based upon Enterococcus (vancomycin-resistant *Enterococcus* strains) can display a high level of resistance to vancomycin (Eaton and Gasson., 2001 ., Anadón et al., 2006), or can acquire such resistance. Certain strains of vancomycin-resistant enterococci are commonly associated with nosocomial infections in hospitals (Leclercq and Courvalin., 1997). Not be referred to as a probiotic for human use. Lactic acid bacteria possess plasmids containing genes conferring resistance to tetracycline, erythromycin, chloramphenicol or lincosamide, macrolide, streptomycin, and streptogramin (Lin CF., 1996. Tannock GW., 1991).

The normal level of prebiotics that can be consumed approximately 20 g/day more that it will have some side effect such as abdominal bloating, diarrhea or gas production (Tuohy et al., 2003). the Nordic Working Group on Food Toxicology and Risk Evaluation (NNT) (Nordic Committee of Ministers, 2000) mentioned that consumption of 20 g/day or less of FOS by a person weighing 60 kg for the 1 to 3 weeks had adverse effects like flatulence, abdominal pain, bloating, cramps, or diarrhea . During active diarrhea prebiotics also have disadvantages because they may exacerbate the adverse effects of simple sugar absorption. Daily intakes more than 8 g/day of inulin and oligofructose are considered to be with side effects and Health-relevant effects . In a study made by De Vrese and Schrezenmeir, 2008. 0.04–0.06 g/kg body weight oligofructose had ingested at least one of the tested symptoms (headache, belching, flatulence, bowel contractions, or liquid stools) had been observed (De Vrese and Schrezenmeir., 2008). (Clausen et al., 1998) admonstrated that 80 g/day of oligofructose caused diarrhea in 4 of 12 test subjects . Because of fermentation in the large intestine, ingestion of higher quantities of prebiotics may lead to flatulence, abdominal disorders, and diarrhea. (Manning & Gibson., 2004) suggested that to increaes the bifdobacteria levels in the human gut 2 g/day of XOS, 10 g/day of GOS and at least 4 g FOS/day are required. The safety requirements could be more meticulous in the case of human probiotics according to variable health status of humans . (Zunic et al., 1991) reported that pneumonia and nonsymptomatic fungal infections

occurred under the influence of *Lactobacillus* bacteria, *Saccharomyces boulardii* respectively. *Enterococci* (*E. faecium* and *E. faecalis*), which have emerged as opportunistic pathogens in hospital environments, causing nosocomial infections such as endocarditis, bacteraemia, and intra-abdominal, urinary tract, and central nervous system (CNS) infections. (Theodorakopoulou et al., 2013) Administrated that the most important concern about probiotic use is the risk of bacteremia, fungemia, and sepsis. *Lactobacilli* have been reported in adults to cause cases of sepsis, meningitis, and infections localized in different organs (Mackay et al., 1999). Thus, the use of probiotics in immunocompromised patients could generate serious health risks (e.g., *S. boulardii* and *L. rhamnosus*) but it still the risk to benefit ratio appears favorable (Marteau., 2001).

(Damini K., 2019), administrated that *Saccharomyces cerevisiae*, *Lactobacillus*, and *Bifidobacterium* have the ability to cross the intestinal mucosa and enter into bloodstream causing systemic infections such as human sepsis (lactobacilli, *L. rhamnosus* strains) (M.T. Liong., 2008), and *E. coli* NISSLE 1917 probiotic strain in low-birth-weight infants (K. Guenther., 2010), IE, such as *L. rhamnosus*, *L. casei*, *L. acidophilus*, *L. jensenii*, *L. Plantarum and L. paracasei*. Virulence factors and harmful metabolites of probiotics (*Enterococcus*, *Bacillus*, *Lactobacillus*, *and Streptococcus*) might lead to opportunistic infections and metabolic disturbances, respectively; in addition to exaggerated immune response through triggered cytokine production and neurotransmitters BAs (e.g., cadaverine, histamine, tyramine, agmatine, and putrescine) lead to autoimmune disease and trigger headaches in people sensitive to these substances (V.T. Martin., 2016). In experimental probiotic studies, immunobiological activities and immunostimulatory effects of probiotics from the group of Gram-negative (*E. coli* Nissle 1917) and Gram-positive (*Lactobacillus casei* DN-114001) bacteria on the cytokine secretion have been recognized. *Enterococcus faecium*, a normal inhabitant of human gut, was reported to produce a hemolysin (H.A. Elsner ., 2000).

One study observed that after infection onset with *Lactobacillus* bacteremia the mortality rate of 26% in 1 month and 48% in 1 year (M.K. Salminen., 2002). Some studies reported that in fermented foods such as dairy products some probiotic strains (e.g., *Lactobacillus buchneri*, *Streptococcus thermophiles, Lactobacillus hilgardii*, *Lactobacillus helveticus*) can cause reactions in regarded to produce histamine, also *L. lactis* might cause a significant accumulation of cadaverine over a period of consumption (H.M.L.J. Joosten., 1989, Y. Gezginc., 2013, E. Kuley., 2012).



Figure 4. Systemic infections and excessive immune stimulation of Probiotics

## **Commonly Used Probiotics Products and thier Characteristics**

The trend today is the development of novel food for special health use, called functional food, to promote human health and well-being of consumers. Probiotic products are the fastest growing area of novel food product development. Probiotic bacteria strains are successfully used in the production of processed milk products, cereal products, bread-making and vegetable and fruit juices (Danuta K-K., 2012). FAO suggested the following standardizations regarding evaluate and substantiate that a product is a prebiotics.



Figure 3. Guidelines fort the evaluation and substantiation of prebiotics (FAO ., 2007).

Development of the probiotic products is dependent on the pH, sharpness in taste, oxygen content, salt concentration, the temperature of processing and storage, shelf-life and number of viable microorganisms during consumption. Use of probiotic bacteria in the non-dairy product is challenging and important to research and industry for commercialization of healthy beverage (Anil P., 2018). Some of the probiotics used in food production such as, Meat production: probiotics increased the carcass output and water holding capacity and decreased the meat hardness (Paulius M ., 1994), in addition, to reduce morbidity and mortality of growing rabbits during the fattening periods.Several probiotic strains have been utilized for fermented sausages such as lactic acid producing bacteria, mainly lactobacillus, pedococus, and streptococcus . Also in Egg production: probiotics supplementation in feed increased egg quality and egg production, reduced triglycerides and plasma cholesterol and decreased egg contamination (Haddadin MSY. 1996. Kurtoglu V .2004), prebiotic supplementation also increased egg shell weight shell thickness and serum calcium (Panda AK.., 2007).

Table 1. Explains the various types of products including probiotics.

| The nam | e of the | product |
|---------|----------|---------|
|---------|----------|---------|

Fruit based probiotic food products

Grapes probiotic (Hardaliye) : lactic acid bacteria (L. plantarum, L. delbrueckii, and L. rahamnosus), L. casei subsp. pseudoplantarum, L. Paracasei, L. brevis, L. pontis, L. acetotolerans, L. Vaccinostercus, L. Sanfrancisco. (Anil P., 2018).

Blackcurrant probiotic juice : L. plantarum (Luckow., 2004).

Peach Probiotics : L. delbrueckii, L. brevis, L. lactis, Weissella cibaria, W. paramesenteroides, W. minor, Leuconostoc mesenteroides, and E.faecalis (Anil P., 2018).

live probiotic drink : L. pentosus, L. plantarum, Pediococcus cerevisiae, L. mesenteroides, L.brevis, L. mali, L.brevis, L. mesenteroides and L. fermentum (Argyri et al., 2013).

Durian fruit probiotic : L.casei. (Anil P., 2018).

Mango probiotic juice : L. acidophilus, L. delbrueckii, L.plantarum and L. casei (Reddy., 2015).

Sweet lime is probiotic: L. plantarum (Khatoon ., 2015).

Orange probiotic juice : L. bulgaricus and L. Plantarum (Anil P., 2018).

#### The name of the product

Vegetables based probiotic juice or beverages

Carrot and beetroot(Shalgam, Beetroot juice, Kanji): Saccharomyces cerevisiae, Lactobacillus, Leuconostoc, Pediococcus, L. Plantarum, L. acidophilus, L. casei (Anil P., 2018).

Moringa leave : L. plantarum, Enterococcus (Vanajakshi., 2015).

Broccoli/cabbage : L. plantarum, Enterococcus sulphurous, W. paramesenteroides, W. minor, Leuconostoc mesenteroide W. Cibaria, L. mesenteroides, L. plantarum, L. brevis, L. rhamnosus and L. Plantarum. (Anil P., 2018).

Kombucha: Saccharomyces ludwigii, S. cerevisiae, S. bisporus, Torulopsis sp. and Zygo saccharomyces (Jayabalan, M., 2014).

Cucumber(Khalpi, Jiang-gua) : L. plantarum, Pediococcus pentosaceus. W. hellenica, L. plantarum, Leuconostoc lactis, and Enterococcus (Dahal et al., 2005, Chen et al., 2013).

Mustard leaves (Inziangsang) : L. brevis, L. plantarum, and Pediococcus acidilactici (Tamang., 2009; Yan et al., 2008).

Eggplant (Camuoi) : L. fermentum, L. pentosus and L. Brevis (Nguyen ., 2013).

Turnip and radish leaves (Nozawana-Zuke) : L. curvatus (Kawahara ., 2006).

Cereal and legumes based food products

Bushera : L. brevis, Enterococcus, Streptococcus, and Lactococcus (Muyanja et al., 2003).

Mahewu : Lactococcus lactis (Blandino., 2003).

Togwa : Lactobacillus, Streptococcus, and L. Planetarium. (Mugula., 2003).

Kefir Soy : L. brevis, L. kefir, L. mesenteroides and L. helveticus, Kluyveromyces, marxianus, Kluyveromyces lactis (Anil P., 2018).

Velli : B. bifidum, and L. acidophilus (Anil P., 2018).

Boza : L. coprophilous, L. plantarum, L. acidophilus, Leuconostoc reffinolactis, L. brevis, L. fermentum, Leuconostoc mesenteroides, and S. uvarum (Moncheva et al., 2003).

Koozh : W. paramesenteroides, L. plantarum and L. fermentum (Anil P., 2018).

#### Mea products

Dry sausage: L. rhamnosus GG, LC-705, and E-97800 (Erkkilä et al., 2001).

Hungarian salami : NTMS cultures, including Lactobacillus acidophilus LAFTIkL10, L. Paracasei LAFTIkL26, L. paracasei 5119, Lactobacillus sp. L24 and Bifidobacterium lactis LAFTIkB94 (Pidcock et al., 2002).

Fermented sausages : Potential probiotic 20 strains Lactobacillus (brevis, plantarum, casei, paracsei, pentosus, reuteri, curvatus, sakei, zeae) (Pennacchia et al., 2004).

Scandinavian-type fermented sausage : Potential probiotic L. plantarum/pentosus (MF1291, MF1298, MF1300) (Klingberg et al., 2004).

Dry-fermented Iberian Sausages : Potential L. fermentum HL57, L. Reuteri PL519, and L. reuteri PL542 (Ruiz-Moyano et al., 2007).

Dairy-based probiotic foods

Drinkable fresh milk and fermented milk: L. rhamnosus GG, L. acidophilus, L. casei, L. rhamnosus, L. Plantarum (Aragon-Alegro LC., 2007).

Yogurt: L. delbrueckii subsp. bulgaricus, Streptococcus salivarius subsp., and Thermophilus (Danfeng, S, et al., 2012).

Chocolate mousse : L. paracasei subsp. paracasei LBC 82 (Özer BH., 2010).

#### 2. CONCLUSION

The use of probiotics and prebiotics applications have shown a tremendous increase in the last years. This review presents the micro-organisms that are most commonly used as probiotics, the characteristics of beneficial prebiotics included resistant to gastric acidity and hydrolysis by mammalian digestive enzymes, metabolized by the host in addition to have a beneficial effect to the host health, selectively stimulate the growth and or activity of health-promoting intestine, the beneficiary of prebiotics and probiotics, showing their efficacy in the systemic health, metabolism and immune system. Potential advantages of using pre-probiotics from a health standpoint, including hyper cholesterol and cardiovascular disease, diarrhea, antibiotic therapy, Kidney Stones, Lactose intolerance, hypertension, pancreatitis, tooth problems *Lactobacillus*, anemia. Eczema, food allergies, NAFLD, inflammatory bowel diseases, urinary tract infection, celiac disease, and pouchitis. Probiotics are selective viable micro-organisms administered in an adequate amount to confer health benefits beyond inherent general nutrition. Safety Considerations and Potential risks also take place in this

review. Commonly Fruit, Vegetables, Meat, Cereal, legumes, and dairy-based probiotic food products and their characteristics were mentioned with giving examples for the included probiotics.

#### REFERENCES

Aguirre M, Collins, MD (1993): Lactic acid bacteria and human clinical infection. J Appl Bacteriol, 75: 95-107.

- Al-Sheraji, S. H., Ismail, A., Manap, M. Y., Mustafa, S., Yusof, R. M., & Hassan, F. A. (2013). Prebiotics as functional foods: A review. Journal of Functional Foods, 5, 1542–1553.
- Anadón, A., Martinez-Larrañaga, M.R., Martinez, M.A., 2006. Probiotics for animal nutrition in the European Union. Regulation and safety assessment. Regul. Toxicol. Pharmacol. 45, 91–95
- Anil P., Sandeep J., Kiran V., Yogesh G., Vikas K., Navnidhi C.2018.Potential non-dairy probiotic products A healthy approach. Food Bioscience 21 (2018) 80–89.
- Anil Panghala, Sandeep Janghub, Kiran Virkara, Yogesh Gata, Vikas Kumara, Navnidhi Chhikara..2018 .Potential non-dairy probiotic products A healthy approach. Food Bioscience 21 (2018) 80–89.
- Anna I., Giuseppina M. R., Roberto B. C., Antonio C., Rosaria M. Probiotics as an emerging therapeutic strategy to treat NAFLD: focus on molecular and biochemical mechanisms. Journal of Nutritional Biochemistry 22 (2011) 699–711.
- Anna I., Giuseppina M. R., Roberto B. C., Antonio C., Rosaria M. Probiotics as an emerging therapeutic strategy to treat NAFLD: focus on molecular and biochemical mechanisms. Journal of Nutritional Biochemistry 22 (2011) 699–711.
- Anukam, K. C., Hayes, K., Summers, K., & Reid, G. (2009). Probiotic Lactobacillus rhamnosus GR-1 and Lactobacillus reuteri RC-14 may help downregulate TNF-Alpha, IL-6, IL-8, IL-10, and IL-12 (p70) in the neurogenic bladder of spinal cord injured patient with urinary tract infections: A two-case study. Advances in Urology
- Aragon-Alegro LC, Alarcon Alegro JH, Roberta Cardarelli H, Chih Chiu M, Isay Saad SM (2007) Potentially Probiotic and Synbiotic Chocolate Mousse. LWT-Food Sci. Technol. 40:669-675
- Argyri, A. A., Zoumpopoulou, G., Karatzas, K. A. G., Tsakalidou, E., Nychas, G. J. E., Panagou, E. Z., & Tassou, C. C. (2013). Selection of potential probiotic lactic acid bacteria from fermented olives by in vitro tests. Food Microbiology, 33(2), 282–291
- Arturo A., María R. M. L., Irma A., María A. M. Chapter 55 Probiotics: Safety and Toxicity Considerations. Nutraceuticals, Efficacy, Safety and Toxicity .2016, Pages 777-798.
- Aso Y, Akaza H, Kotake T, Tsukamoto T, Imai K, Naito S (1995): Preventive effect of a Lactobacillus casei preparation on the recurrence of superficial bladder cancer in a double-blind trial. The BLP Study Group Eur Urol, 27: 104-9
- Balamurugan, R., Mary, R. R., Chittaranjan, S., Jancy, H., Devi, R. S., & Ramakrishn Ventura, M., & Perozzi, G. (2011). Introduction to the special issue "probiotic bacteria and human gut microbiota". Genes & Nutrition, 6(3), 203.
- Balamurugan, R., Mary, R. R., Chittaranjan, S., Jancy, H., Devi, R. S., & Ramakrishna, B.S. (2010). Low levels of faecal lactobacilli in women with iron-deficiency anaemia in south India. British Journal of Nutrition, 104(7), 931–934
- Blandino, A., Al-Aseeri, M. E., Pandiella, S. S., Cantero, D., & Webb, C. (2003). Cerealbased fermented foods and beverages. Food Research International, 36(6), 527–543(36:527–543).
- Clausen, M.R., Jorgensen, J., Mortensen, P.B., 1998. Comparison of diarrhea induced by ingestion of fructooligosaccharide Idolax and disaccharide lactulose: the role of osmolarity versus fermentation of malabsorbed carbohydrate. Dig. Dis. Sci. 43, 2696–2707.
- Courvalin, P., 2006. Vancomycin resistance in gram-positive cocci. Clin. Infect. Dis. 42 (Suppl. 1), S25-S34
- Dahal, N. R., Karki, T. B., Swamylingappa, B., Li, Q., & Gu, G. (2005). Traditional foods and beverages of Nepal—A review. Food Reviews International, 21(1), 1–25
- Damini K. Seema P., Soo-Ki K. Probiotic supplements might not be universally-effective and safe: A review. Biomedicine & Pharmacotherapy 111 (2019) 537–547.
- Danfeng S., Salam I., Saeed H.2012.Open access peer-reviewed chapter Recent Application of Probiotics in Food and Agricultural Science.DOI: 10.5772/50121. https://www.intechopen.com/books/probiotics/recent-application-of-probiotics-in-food-andagricultural-science.

Danielsen, M., Wind, A., 2003. Susceptibility of Lactobacillus spp. To antimicrobial agents. Int. J. Food Microbiol. 82, 1-11

Danuta K-K., Zbigniew J. D. Probiotic meat products, and human nutrition: Review Process Biochemistry 47 (2012) 1761–1772.

De Vrese, M., Schrezenmeir, J., 2008. Probiotics, prebiotics, and synbiotics. Adv. Biochem. Eng. Biotechnol. 111, 1–66.

- Debapriya M., Snigdha M., Swati M., Priyadarshi S, S.2018. Prebiotics and synbiotics: Recent concepts in nutrition. Food Bioscience 26 (2018) 152–160.
- Dessart SR, Steenson LR. High frequency intergeneric and intrageneric transfer conjugal transfer of drug resistance plasmids in Leuconostoc mesenteroides ssp. cremoris. J Dairy Sci 1991; 74:2912–9.
- Dubos, R., Schaedler, R.W., and Costello, R. (1963). Composition, alteration, and effects of the intestinal flora. Fed. Proc. 22, 1322–1323.
- E. Kuley, E. Balıkcı, I. Özoğul, S. Gökdogan, F. Ozoğul, Stimulation of cadaverine production by foodborne pathogens in the presence of Lactobacillus, Lactococcus, and Streptococcus spp, J. Food Sci. 77 (2012) M650–M658, https://doi.org/10.1111/j.1750-3841.2012.02825.x.
- Eaton TJ, Gasson MJ (2001): Molecular screening of Enterococcus virulence determinant potential for genetic exchange between food and medical isolates. Appl Environ Microbiol, 67: 1628-1635.
- El-Nezami H, Mykkänen H, Kankaanpää P, Salminen S, Ahokas J (2000): Ability of Lactobacillus and Propionibacterium strains to remove aflatoxin B1 from chicken duodenum. J Food Protect, 63: 549-552
- Erkkilä S, Petäjä E, Eerola S, Lilleberg L, Mattila-Sandholm T, Suihko ML. Flavor profiles of dry sausages fermented by selected novel meat starter cultures. Meat Sci 2001;58:111–6.
- FAO technical meeting on prebiotics.2007. www.fao.org/ag/agn/agns/index\_en.stm. September 15-16, 2007.
- FAO/WHO, 2002. Joint FAO/WHO Group Report on Drafting Guidelines for the Evaluation of Probiotics in Food, London Ontario, Canada, April 30 and May 1, 2002, pp. 1–11.
- Gibson, G.R. and Roberfroid, M.B. (1995). Dietary modulation of the human colonic microbiota: Introducing the concept of prebiotics.J. Nutr. 125(6), 1401–1412
- Girardin M, Seidman EG. Indications for the use of probiotics in gastrointestinal diseases. Dig Dis. 2011;29:574---87.
- Goldin, B. and Gorbach, S.L. (1984). The effect of milk and Lactobacillus feeding on human intestinal bacterial enzyme activity.Am. J. Clin. Nutr. 33, 15–18
- Guandalini S, Pensabene L, Zikri MA, Dias JA, Casali LG, Hoekstra H, Kolacek S, Massar K, Micetic-Turk D, Papadopoulou A, de Sousa JS, Sandhu B, Szajewska H, Weizman Z (2000): Lactobacillus GG administered in oral rehydration solution to children with acute diarrhea: A multicenter European trial. J Pediatr Gastroenterol Nutr, 30: 54-60.
- Guarino A, Berni Canani R, Spagnuolo MI, Albano F, Di Benedetto L (1997): Oral bacterial therapy reduces the duration of symptoms and viral excretion in children with mild diarrhea. J Pediatr Gastroenterol Nutr, 25: 516-519
- H. Aarts, A. Margolles, Antibiotic resistance genes in food and gut (nonpathogenic) bacteria. Bad genes in good bugs, Front. Microbiol. 5 (2014) 754, https://doi.org/10.3389/fmicb.2014.00754.
- H.A. Elsner, I. Sobottka, D. Mack, M. Claussen, R. Laufs, R. Wirth, Virulence factors of Enterococcus faecalis and Enterococcus faecium blood culture isolates, Eur. J. Clin. Microbiol. Infect. Dis. 19 (2000) 39–42, https://doi.org/10.1007/s100960050007.
- H.M.L.J. Joosten, M.D. Northolt, Detection, growth, and amine-producing capacity of Lactobacilli in cheese, Appl. Environ. Microbiol. 55 (1989) 2356–2359.
- Haddadin MSY, Abdul Rahim SM, Robbinson RK (1996) The effect of *Lactobacillus acidophilus* on the production and chemical composition of hens egg. J Poult Sci 75: 491–494.
- Hibberd, P.L., Davidson, L.E., 2008. Safety of probiotics. AgroFOOD Ind. Hi-Tech. 19, 30-33
- Hosada M, Hashimoto H, He D, Morita H, Hosono (1996): Effect А of with milk Lactobacillus LA-2 administration of fermented acidophilus on faecal mutagenicity and microflora in the human intestine. J Dairy Sci, 79: 745-749.
- Islam SU. Clinical uses of probiotics. Medicine (Baltimore).2016;95:e2658

- Isolauri E, Juntunen M, Rautanen T, Sillanaukee P, Koivula T (1991): A human Lactobacillus strain (Lactobacillus casei sp. strain GG) promotes recovery from acute diarrhea in children. Pediatrics, 88: 90-97.
- Jayabalan, R., Malbaša, R. V., Lončar, E. S., Vitas, J. S., & Sathishkumar, M. (2014). A review of kombucha tea-microbiology, composition, fermentation, beneficial effects, toxicity, and tea fungus. Comprehensive Reviews in Food Science and Food Safety, 13(4), 538–550
- Joint FAO/WHO Expert Consultation on Evaluation of Health and Nutritional Properties of Probiotics in *Food including Powder Milk* with Live Lactic Acid Bacteria, 1-4 October 2001.
- Juan J. S.D, Review of the role of probiotics in gastrointestinal diseases in adults. Gastroenterol Hepatol. 2017;40(6):417---429
- K. Guenther, E. Straube, W. Pfister, A. Guenther, A. Huebler, Sever sepsis after probiotic treatment with Escherichia coli NISSLE 1917, Pediatr. Infect. Dis. J. 29 (2010) 188–189, https://doi.org/10.1097/INF.0b013e3181c36eb9.
- K. Guenther, E. Straube, W. Pfister, A. Guenther, A. Huebler, Sever sepsis after probiotic treatment with Escherichia coli NISSLE 1917, Pediatr. Infect. Dis. J. 29 (2010) 188–189, https://doi.org/10.1097/INF.0b013e3181c36eb9.
- Kalliomaki M. Salminen S, Arvilommi H, Kero P, Koskinen Ρ, Isolauri Ε (2001): **Probiotics** Α placebo-controlled in primary prevention of atopic disease: randomized Trial. Lancet. 357: 1076-9.
- Kawahara, T., & Otani, H. (2006). The stimulatory effect of lactic acid bacteria from commercially available Nozawana-zuke pickle on cytokine expression by mouse spleen cells. Bioscience, Biotechnology, and Biochemistry, 70(2), 411–417.
- Khatoon, N., & Gupta, R. K. (2015). Probiotics beverages of sweet lime and sugarcane juices and its physiochemical, microbiological & shelf-life studies. Journal of Pharmacognosy and Phytochemistry, 4(3).
- Kim HS, Gilliland SE (1983): Lactobacillus acidophilus as a dietary adjunct for milk to aid lactose digestion in humans. J Dairy Sci, 66: 959-966.
- Klingberg TD, Axelsson L, Naterstad K, Elsser D, Budde BB. Identification of potential probiotic starter cultures for Scandinaviantype Sausages. Int J Food Microbiol 2005;105:419–31
- Kurtoglu V, Kurtoglu F, Seker E, Coskun B, Balevi T, Polat ES (2004) Effect of probiotic supplementation on laying hen on yield performance and serum and egg yolk cholesterol. J Food Additives and Contaminants, pp. 817–823.
- Leclercq R, Courvalin P (1997): Resistance to glycopeptides in enterococci. Clin Infect Dis, 24: 545-54.
- Lin CF, Fung ZF, Wu CL, et al. Molecular characterization of a plasmid-borne (pTC82) chloramphenicol resistance determinant (cat-TC) fromLactobacillus reuteri G4. Plasmid 1996; 36:116–24.
- Luckow, T., & Delahunty, C. (2004). Which juice is 'healthier'? A consumer study of probiotic non-dairy juice drinks. Food Quality and Preference, 15(7), 751–759.
- M.T. Liong, Safety of probiotics: translocation and infection, Nutr. Rev. 66 (2008)192–202, https://doi.org/10.1111/j.1753-4887.2008.00024.x.
- Macfarlane, G. T., Steed, H., & Macfarlane, S. (2008). Bacterial metabolism and health-related effects of galacto-oligosaccharides and other prebiotics. *Journal of Applied Microbiology*, *104*, 305–344.
- Mackay, A.D., Taylor, M.B., Kibbler, C.C., et al., 1999. Lactobacillus endocarditis caused by a probiotic organism. Clin. Microbiol.Infec. 5 (290), 292.
- Majamaa H, Isolauri E, Saxelin M, Vesikari T (1995): Lactic acid bacteria in the treatment of acute rotavirus gastroenteritis. J Pediatr Gastroent Nutr, 20: 333-338.
- Manning, T. S., & Gibson, G. R. (2004). Prebiotics. Best Practice & Research Clinical Gastroenterology, 18, 287–298.
- Marteau, P., 2001. Safety aspects of probiotic products. Scand J. Nutr.45, 22-24.
- Lambert (1995): Midolo PD, JR, Hull R. Luo F, Grayson ML In vitro inhibition of pylori NCTCorganic Helicobacter 11637 by acids and lactic acid bacteria. J Appl Bacteriol, 79: 475-479.

- Moncheva, P., Chipeva, V., Kujumdzieva, A., Ivanova, I., Dousset, X., & Gocheva, B. (2003). The composition of the microflora of boza, an original Bulgarian beverage. Biotechnology & Biotechnological Equipment, 17(1), 164–168.
- Morelli L, Sarra PG, Bottazzi V. In vivo transfer of pAM beta one from Lactobacillus reuteri to Enterococcus faecalis. J Appl Bacteriol 1988; 65:371–5.
- Mugula, J. K., Nnko, S. A. M., Narvhus, J. A., & Sørhaug, T. (2003). Microbiological and fermentation characteristics of togwa, a Tanzanian fermented food. International Journal of Food Microbiology, 80(3), 187–199.
- Muyanja, C. M. B. K., Narvhus, J. A., Treimo, J., & Langsrud, T. (2003). Isolation, characterization, and identification of lactic acid bacteria from bushera: A Ugandan traditional fermented beverage. International Journal of Food Microbiology, 80(3),201–210.
- Naidu AS, Biblack WR, Clemens RA (1999): Probiotic spectra of lactic acid Bacteria (LAB). Crit Revs Food Sci & Nutr, 39: 13-126.
- Napolitano, A., Costabile, A., Martin-Pelaez, S., Vitaglione, P., Klinder, A., Gibson, G. R., & Fogliano, V. (2009). The potential prebiotic activity of oligosaccharides obtained by enzymatic conversion of durum wheat insoluble dietary fiber into the soluble dietary fiber. *Nutrition, Metabolism and Cardiovascular Diseases*, 19, 283–290.

Needed to substantiate a health effect for probiotics and prebiotics. European Journal of Nutrition, 44(5), 303–310.

- Nguyen, D. T. L., Van Hoorde, K., Cnockaert, M., De Brandt, E., Aerts, M., & Vandamme, P. (2013). A description of the lactic acid bacteria microbiota associated with the production of traditionally fermented vegetables in Vietnam. International Journal of Food Microbiology, 163(1), 19–27.
- Niers, L., Martín, R., Rijkers, G., Sengers, F., Timmerman, H., Van Uden, N., ... Hoekstra, PandA study). The effects of selected probiotic strains on the development of eczema (the Allergy, 64(9), 1349–1358 M. (2009).
- Nordic Committee of Ministers, 2000. Nordic Committee of Senior Officials for Food Issues. Nordic Working Group on Food Toxicology and Risk Evaluation. Safety Evaluation of Fructans. TemaNord, Copenhagen, pp. 523
- Oatley JT, Rarick MD, Ji GE, Linz JE (2000): Binding of aflatoxin B1 to bifidobacteria in vitro. J Food Prot, 63: 1133-6.
- K, T. Ogawa M. Shimizu K. Nomoto Takahashi Μ, Watanuki M, Tanaka R. Tanaka Takeda (2001): Hamabata Τ, Yamasaki S. Υ Protective effect Lactobacillus casei of strain Shirota onShiga toxin-producing Escherichia coli 0157:H7 infection infant in rabbits. Infect Immun, 69: 1101-8.
- Özer BH, Kirmaci HA (2010) Functional Milks and Dairy Beverages. Int. j. Dairy technol. 63:1-15.
- Panda AK, Ramarao SS, Raju MV, Sharma SS (2007) Effect of probiotic (*Lactobacillus sporogenes*) feeding on egg production and quality, yolk cholesterol and humoral immune response of whiteleghorn layer breeders. J Sci Food and Agri 88: 43—47.
- Parvez, S., Malik, K. A., Ah Kang, S., & Kim, H. Y. (2006). Probiotics and their fermented food products are beneficial for health. Journal of Applied Microbiology, 100(6),1171–1185.
- Passariello, A., Terrin, G., De Marco, G., Cecere, G., Ruotolo, S., Marino, A., ... Canani, R. B. (2011). Efficacy of a new hypotonic oral rehydration solution containing zinc and prebiotics in the treatment of acute childhood diarrhea: A randomized controlled trial. *Journal of Pediatrics*, 158, 288–292.
- Patel, S., & Goyal, A. (2012). The current trends and future perspectives of prebiotics research: A review. 3 Biotech, 2, 115–125.
- Paulius M, Lina A, Ana Z, Andrzej G, Manfred OL, Areta H (2006) Effect of probiotic Bioplus 2B on the performance of growing rabbit. Arija Zootechnica 36: 54—59.17- Hammes WP, Knauf HJ (1994) Starters in the processing of meat products. J Meat Sci 36: 155—168
- Pennacchia C, Ercolini D, Blaiotta G, Pepe O, Mauriello G, Villani F. Selection of Lactobacillus strains from fermented sausages for their potential use as probiotics. Meat Sci 2004;67:309–17.
- Perdone CA, Bernabeu AO, Postaire ER. Bouley CF. Reinert Р (1999): The effect of DN-114 supplementation by Lactobacillus casei (strain 001) on accute diarrhea in children attending day care centers. Int J Clin Pract, 53: 179-184.

Pidcock K, Heard GM, Henriksson A. Application of nontraditional meat starter cultures in production of Hulgarian salami. Int J Food Microbiol 2002;76:75–81.

Pezzilli, R., & Fantini, L. (2006). Probiotics and severe acute pancreatitis. JOP, 7(1),93-92.

- Probiotic supplementation in the first 6 months of life in at risk Asian infants–effects on eczema and atopic sensitization at the age of 1 year. Clinical & Experimental Allergy, 39(4), 571–578.
- Rao, S., Srinivasjois, R., & Patole, S. (2009). Prebiotic supplementation in full-term neonates: A systematic review of randomized controlled trials. *Archives of Pediatrics and Adolescent Medicine*, *163*, 755–764.
- Reddy, L. V., Min, J. H., & Wee, Y. J. (2015). Production of probiotic mango juice by fermentation of lactic acid bacteria. Microbiology and Biotechnology Letters, 43, 120–125.
- Roberfroid M, Gibson GR, Roberfroid M (2008) Prebiotics: concept, definition, criteria, methodologies, and products. In: Handbook of prebiotics. CRC Press, Boca Raton, pp 39–69.
- Rosander, A., Connolly, E., Roos, S., 2008. Removal of antibiotic resistance gene-carrying plasmids from Lactobacillus reuteri ATCC55730 and characterization of the resulting daughter strain, L. reuteri DSM 17938. Appl. Environ. Microbiol. 74, 6032– 6040.
- Ruiz-Moyano S, Martín A, Benito MJ, Hernández A, Casquete R, Serradilla MJ, et al. Safety and functional aspects of pre-selected lactobacilli for probiotic use in Iberian dry-fermented sausages. Meat Sci 2009;83:460–7.
- S. Krishnareddy, P.H.R. Green.2017. The Microbiota in Gastrointestinal Pathophysiology Implications for Human Health, Prebiotics, Probiotics, and Dysbiosis 2017, Pages 365-371.
- Saavedra JM, Bauman NA, Oung I, Perman JA, Yolken RH (1994): Feeding of Bifidobacterium bifidum and Streptococcus thermophilus to infants in hospital for prevention of diarrhea and shedding of rotavirus. Lancet. 344: 1046-9.
- Salminen, S., von Wright, A., 1998. Current probiotic-safety assured? Microb. Ecol. Health Dis. 10, 68-77.
- Sanders, M. E., Tompkins, T., Heimbach, J. T., & Kolida, S. (2005). Weight of evidence.
- Shornikova AV, Isolauri E, Burkanova L, Lukovnikova S, Vesikari T (1997). A trial in the Karelian Republic of oral rehydration and Lactobacillus GG for treatment of acute diarrhea. Acta Paediatr, 86: 460-5.
- Shu Q, Lin H, Rutherfurd KJ, Fenwick SG, Prasad J, Gopal PK, Gill HS (2000): Dietary Bifidobacterium lactis HN019 enhances resistance to oral Salmonella typhimurium infection in mice. Microbiol. Immunol, 44: 213 222.
- Simmering R, Blaut M (2001) Pro- and prebiotics-the tasty guardian angles? Applied Microbiol and Biotechnol 55 : 19-28
- Soh, S. E., Aw, M., Gerez, I., Chong, Y. S., Rauff, M., Ng, Y. P. M., ... Shek, L. C. (2009).
- Spindler-Vesel, A., Bengmark, S., Vovk, I., Cerovic, O., & Kompan, L. (2007). Synbiotics, prebiotics, glutamine, or peptide in early enteral nutrition: A randomized study in trauma patients. Journal of Parenteral and Enteral Nutrition, 31, 119–126.
- Szajewska H, Kotowska M, Mrukowicz JZ, Armanska M, Mikolajczyk W (2001): Efficacy of Lactobacillus GG in prevention of nosocomial diarrhea in infants. J Pediatr, 138(3): 361-365.
- Tamang, J. P. (2009). Himalayan fermented foods: Microbiology. Nutrition, and ethnic values. CRC Press.
- Tannock GW, Luchansky JB, Miller L, et al. Molecular characterization of a plasmid-borne (pGT633) erythromycin resistance determinant (ermGT) from Lactobacillus reuteri 100–63. Plasmid 1994; 31:60–71.
- Theodorakopoulou, M., Perros, E., Giamarellos-Bourboulis, E.J., et al., 2013. Controversies in the management of the critically ill: theroleofprobiotics. Int. J. Antimicrob. Agents 42 (Suppl.), S41–S44
- Tuohy, K. M., Probert, H. M., Smejkal, C. W., & Gibson, G. R. (2003). Using Probiotics and Prebiotics to improve gut health. Drug Discovery Today, 8, 692–700.
- V.T. Martin, B. Vij, Diet, and headache: part 1, Headache 56 (2016) 1543-1552, https://doi.org/10.1111/head.12953.
- Vanajakshi, V., Vijayendra, S. V. N., Varadaraj, M. C., Venkateswaran, G., & Agrawal, R. (2015). Optimization of a probiotic beverage based on Moringa leaves and beetroot. LWT-Food Science and Technology, 63(2), 1268–1273.

Venter, C. S. (2007). Prebiotics: An update. Journal for Family Ecology and Consumer Sciences, 35, 17-25.

Vidanarachchi JK, Iji PA, Mikkelsen LL, Sims I, Choct M (2009) Isolation and characterization of water-soluble prebiotic compounds from Australian and New Zealand plants. Carbohydr Polym 77:670–676. https://doi.org/10.1016/j.carbpol.2009.02.009.

- Y. Gezginc, I. Akyol, E. Kuley, F. Özogul, Biogenic amines formation in Streptococcus thermophilus isolated from home-made natural yogurt, Food Chem. 138 (2013) 655–662, https://doi.org/10.1016/j.foodchem.2012.10.138.
- Yu P, Huber JT, Theurer CB, Chen KH, Nussio LG, Wu Z (1997) Effect of steam flaked or steam rolled corn with or without Aspergillus oryzae in the diet on performance of dairy cows fed during hot weather. J Dairy Sci 80 : 3293—3297.
- Zunic, P., Lacotte, J., Pegoix, M., Buteux, G., Leroy, G., Mosquet, B., et al., 1991. Fongemie a Saccharomyces boulardi. Therapie 45, 497–501.





# ORAL PRESENTATION

# Kurut, Forgotten Traditional Product

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**Abstract:** In addition to the production of foods, it is also important to preserve foods, to extend their shelf life. With the increase in the population of the world and the migration from the village to the city, it has become important to deliver food products from the places of production to the mass consumption centers. From this point, various methods have been used to make food products more durable and maintain them for a long time.

Drying is a method widely used in food preservation and known since ancient times. Different types of drying such as freeze drying, spray drying, microwave drying, drying under vacuum are known. Among these methods, drying foods under the sun is the cheapest, very easy to apply, and require less labor and less equipment. Different products with limited shelf life can be dried and made more durable. In Central Asian countries and in some regions of Turkey, various methods such as draining, and sun drying are used in order to extend the shelf life of yogurt. Kurut is one of the traditional fermented products for which drying technologies can be applied. It is a fermented milk product, which made from yogurt or buttermilk by draining water, and addition of additives such as salt, cream and drying. Kurut is made in Southeastern Anatolia and Eastern Anatolia regions of Turkey, usually in summer, during periods of plenty of milk to meet the needs of yogurt in winter times. Despite of the use as a traditional food in many regions of Turkey, kurut is not popularly known among the people. It is also produced in Central Asian countries such as Kyrgyzstan, Kazakhstan and Uzbekistan and in Middle East countries such as Iran, Lebanon, Syria, Iraqi both by people in rural areas as well as commercial.

Long-term storage without degradation and loss of nutritional value, high protein content, the best use of the residual matter made kurut a nutrient to be emphasized. It is known that kurut is also rich in terms of nutrients and minerals. However, the nutrient content, mineral matter content and microbiological properties of the kurut vary according to the method of the production, the tools and containers that were used and the diet of the animal.

It is thought that kurut can provide a significant contribution to the national economy if it is produced technologically. In this review, studies on kurut, which is started to be forgotten are determined and it is expected that it will contribute positively to the researchers in the next studies.

Keywords: Fermented dairy products, Yoghurt, Kurut, Physicochemical properties, Microbiological properties.

# **1. INTRODUCTION**

Drying is one of the oldest food preservation methods (Patır and Ateş, 2002). Drying is used for the preservation of foods as well as the production processes of some products. Among the drying methods sun drying is one of the cheapest and very easy to maintain (Say et al, 2015). Drying process is used in the dairy industry, in production of milk powder, yogurt powder and some cereal-yogurt mixtures such as kishk, tarhana and in production of traditional products such as kurut, that is made by making yoghurt or buttermilk more durable (Patır and Ateş, 2002).

Kurut is made in Eastern and Southeastern Anatolia Region of Turkey by local people. They make kurut in the summer periods to meet their needs in winter. It is a delicious dairy product, that has sour taste and it becomes like yogurt when it is diluted (Çetinkaya, 2004; Soltani and Güzeler, 2009; Say et al., 2015). Kurut is one of the main winter foods of the local people with low income (Öksüztepe et al., 2013). People in the countryside, produce kurut because of the ease of storage for the winter season when protein sources are scarce. It can be stored in the village conditions for years without deterioration and loss of nutritional value. The high rate of protein, the best use of the residual buttermilk in the current conditions, meeting the protein needs of the people in the period when animal protein sources are scarce and providing

additional income to families makes kurut a dairy product that needs attention (Akyüz and Gülümser, 1987). It is stated that the nutrient value of the kurut is quite high and it contains important amounts of animal protein and calcium, potassium and phosphorus which are necessary for healthy living and development of the person (Öküztepe et al., 2013). The most important source of nitrogenous substances among human foods are; eggs, meat and fish. There is about 13% in egg, 20% in meat and fish and there is about 52.35% protein in the composition of kurut (Eralp, 1953; Patır and Ateş, 2002). According to the Turkish Food Codex Fermented Milk Communique, kurut, is included in the class of concentrated fermented milk products, which protein content have been increased to at least 5.6% before or after fermentation (Say et al., 2015).

Kurut is a milk product that is traditionally produced and consumed by Turks for many years (Kamber, 2008). "Kurut" is comes from root of Turkish word "drying". Kurut is known by different names in different regions of Turkey. In Bolu it is called as "Kesh", in Siirt villages as "Geshk", in Bingol it known as "Keshk", "Chorten", "Torak", "Terne" and as "Chortan" in Mardin areas (Atasever, 2007). This dried milk product is known in Iran as Kashk, in Lebanon Kishk, Jub-Jub in Syria and as Kushuk in Iraq and manufactured using similar methods. It is also known that kurut is made by Kyrgyz, Kazak, Tatar and Uzbek Turks in Central Asia (Say et al., 2015). Besides being a durable yogurt type, it has a longer life than yoghurt. As it is prepared in small pieces, it is easier and economical for consumption. Fat content of kurut varies according to the taste and pleasure of person and is also made in some regions from skimmed milk. The advantage of being made from oily milk; it can easily melt in water, leave a fuller flavor in the mouth and it does not tarnish during the sun drying. In good condition it can last for several years (Çetinkaya, 2004).

The chemical composition results that have been obtained by the researchers in their studies on kurut are given in Table 1, the mineral material and heavy metal contents are given in Table 2 and the values containing the microbiological properties are given in Table 3.

|                | Eralp,<br>(1953) | Akyüz<br>and<br>Gülümser,<br>(1987) | Akyüz<br>et al.,<br>(1993) | Patır<br>and<br>Ateş,<br>(2002) | Atasever,<br>(2007) | Karabulut<br>et al.,<br>(2007) | Kamber,<br>(2008) | Güven<br>and<br>Karaca,<br>(2009) | Soltani<br>and<br>Güzeler,<br>(2009) |
|----------------|------------------|-------------------------------------|----------------------------|---------------------------------|---------------------|--------------------------------|-------------------|-----------------------------------|--------------------------------------|
| pH             | -                | -                                   | 4.26                       | 4.15                            | 4.09                | 3.92                           | 4.2               | 4.28                              | 4.27                                 |
| Asidity(%la)   | -                | -                                   | 1.18                       | 2.40                            | 1.84                | -                              | 2.9               | -                                 | 1.40                                 |
| Dry matter (%) | 80.03            | 79.69                               | 85.51                      | 89.06                           | 86.62               | 84.25                          | 87.9              | 86.86                             | 85.79                                |
| Fat (%)        | 11.07            | 10.58                               | 8.52                       | 32.90                           | 15.48               | 8.57                           | 45.9              | 8.44                              | 9.17                                 |
| Protein(%)     | 52.35            | 52.89                               | 54.64                      | -                               | 49.67               | 53.60                          | 25.5              | 53.41                             | 51.74                                |
| Total ash (%)  | -                | -                                   | 14.89                      | 11.79                           | 11.89               | 11.08                          | 9.98              | -                                 | 12.25                                |
| Salt (%)       | 9.11             | 9.66                                | 12.18                      | 12.85                           | 10.15               | 9.95                           | 6.65              | 10.44                             | 9.77                                 |
| Sample amount  | 42               | 13                                  | 20                         | 25                              | 43                  | -                              | 50                | 22                                | 20                                   |

**Table 1.** The chemical composition of kurut samples

Table 2. Amount of mineral material and heavy metals in kurut samples

|               | Atasever (2007), % | Öküztepe et al., (2013), (mg/kg) |
|---------------|--------------------|----------------------------------|
| Ca            | 7,03±2,22          | 13968.52±10215.11                |
| Р             | 4,10±0,96          | $1060.47 \pm 990.00$             |
| Mg            | 0,31±0,11          | 432.42±90.52                     |
| Na            | 18,87±4,00         | 9782.45±1123.10                  |
| K             | 8,33±2,60          | 7012.45±813.00                   |
| Al            | $0,07{\pm}0,06$    | $1.07\pm0.06$                    |
| Fe            | 0,19±0,18          | $6.57\pm4.04$                    |
| Cu            | $0,02{\pm}0,02$    | $2.44\pm0.15$                    |
| Zn            | 0,30±0,46          | $9.66 \pm 3.04$                  |
| Sample amount | 43                 | 25                               |

|                    |                                   |                        |                  | Kamber       |
|--------------------|-----------------------------------|------------------------|------------------|--------------|
|                    | Akyüz and Gülümser                | Patır and Ateş (2002), | Atasever (2007), | (2008),      |
|                    | (1987), (kob/g)                   | (kob/g)                | (kob/g)          | $(\log_{10}$ |
|                    |                                   |                        |                  | kob/g)       |
| Aerobic mesophilic | 8 41 <sub>m</sub> 10 <sup>3</sup> | $2.40 \times 10^4$     | 2 2-104          | 4.50         |
| bacteria           | 6.41X10 <sup>-</sup>              | 5.40x10                | 5,2X10           | 4.52         |
| Mould and yeast    | 5.01x10 <sup>3</sup>              | $1.12 \times 10^4$     | $1,6x10^5$       | 3.94         |
| Coliform           | -                                 | $2.79 \times 10^2$     | $3,7x10^{2}$     | -            |
| Staphyloccocus     |                                   | $2.40-10^{3}$          | 2 1 103          | 1.01         |
| -Micrococcus       | -                                 | 2.40X10 <sup>e</sup>   | 2,1810           | 1.81         |
| Lactobacillus-     |                                   |                        |                  |              |
| Leuconostoc-       | -                                 | 2.23x10 <sup>4</sup>   | -                | 3.60         |
| Pediococcus        |                                   |                        |                  |              |
| Lactococcus        | -                                 | 1.13x10 <sup>4</sup>   | $1,9x10^4$       | 3.60         |
| Lactobacillus      | -                                 | -                      | $2,2x10^4$       | -            |
| Sample amount      | 13                                | 25                     | 43               | 50           |
|                    |                                   |                        |                  |              |

# Table 3. Microbiological properties of kurut samples

# 2. PREPARATION OF KURUT

There are different methods are used to prepare kurut in different literatures. Kurut is done under very primitive conditions. It does not have a large-scale production and it is produced only in the family businesses in the villages (Akyüz and Gülümser, 1987).

## Preparation of kurut from yogurt

Kurut is made of cows' milk in the summer months (Figure 1). After milking and filtering, the milk is heated to boiling point for 15-20 minutes and then cooled down to the fermentation temperature. After cooling, 2% homemade yogurt is added to this milk as a starter culture by slow mixing and incubated. The incubation period varies from home to home, but it is generally between 2,5 and 8 hours. The yogurt obtained at the end of this process is placed in the refrigerator and kept for 24 h and then poured onto a cloth bag and filtered for 1 day to remove water. Sometimes the filtration process continues for 10-20 days. A well-drained yogurt is then taken into a large container and optionally salt (1–3%) and cream (5–10%) may be added before the shaping process. The concentrated yogurt is then cut into 20–60 g small pieces to give 4–5 cm in diameter and hand-shaped round, oval or conical shapes. These shaped pieces are then placed on a tray (on clean cloths) and dried in a shady, airy place for 2–3 days. They are then dried for 3–5 days in the sun. At the end of this process, they are again taken to a shady place and dried for a further 2–3 months. After this time, kurut is ready for consumption. It can be softened by immersing in warm water to obtain the required consistency before serving; or after grilling and granulating. In a cool and dry place, it can be stored for several years without losing its properties (Kamber, 2008; Güven and Karaca, 2009; Say et al., 2015). Approximately 15-17 kg of yogurt should be used to obtain 1 kg kurut. That is, the yield is about 6% (Akyüz and Gülümser, 1987).

#### Preparation of kurut from buttermilk

In some regions of Turkey butter is made from yoghurt. Buttermilk remains as nutrient with high nutritional value. Apart from fat, buttermilk contains almost all the nutrients in milk such as lactose, protein and mineral substances (Akyüz & Gülümser, 1987). Waste buttermilk is converted to dry form and evaluated with minimum food loss (Atasever, 2007). The buttermilk is heated in large boilers. In some places, some salt is added during heating. Buttermilk clots with the effect of temperature. Then the boiler is taken from the fire and the clot collapses to the bottom, a clear green water is collected on top. This water is poured, and the clot stuffed in cloth bags for filtration. In this way a significant part of the water is separated. Afterwards, the bags are pressed between the stones to ensure that the remaining water is disposed as much as possible. The resulting precipitate is salted. Sometimes, it is kneaded by adding fresh cream or butter for good taste. Then it is divided into 30-40-gram pieces and shaped round or oval by hand. The shaped product is usually laid on paper or on cloths and dried in the sun. In order acceleration of drying occasionally kuruts are turned upside down. Kuruts are left to dry for 10-15 days under the sun. Kurut is stored in a cool place to be used in winter meals (Akyüz and Gülümser, 1987; Patır and Ateş, 2002).



Figure 1. Preparation procedure of kurut

# **3. KURUT IN THE WORLD**

Kurut (fermented yak milk) is a naturally fermented dairy product and a main source of livelihood for Tibetan households in the Qing-Tibetan highlands. It has long played important economic and dietary roles to the people of Qinghai and Tibet. Kurut is a pure white, viscous, fermented dairy food with unique organoleptic sensations of acidity and alcohol; fermentation usually takes 3 to 6 days at an ambient temperature around 10 to 20°C to achieve the desired properties. Kurut contains 5.37% milk fat and approximately 5.44% total protein, on average, which is about twice as rich in fat and protein as in yogurt. The contents of calcium (140 mg/100 g), phosphorus (146 mg/100 g), magnesium (154 mg/100 g), zinc (5.74 mg/100 g), and B vitamin in kurut are also higher than in yogurt (Liu et al., 2012).

In Iran, nomad ranchers and peasants process cows, sheep and goat's milk to kurut in primitive conditions. This product in Iran is called "Kashk". Kashk is a fermented dairy product with a high nutritional value. In Iran, kashk is produced in three ways: dry kashk, traditional liquid kashk and industrial liquid kashk. Dry kashk is obtained in the traditional conditions by the peasants and nomads by removing the water and drying the yoghurt in the sun. The dry kashk is usually undergo to microbial contamination, because of the production and storage conditions are not suitable for hygienic conditions. In order to solve this problem and to protect consumer health, traditional liquid kashk and industrial liquid kashk are produced in industrial units. Traditional liquid kurut and industrial liquid kurut have a significant share in the production of Iranian dairy products. It is used in the preparation of various dishes lovingly by the people (Say et al., 2015).

Kurut is one of the most valuable products that nomad Kyrgyz people have consumed for centuries. Long-term storage, high protein content have made kurut a very valuable product among the people. Today, the kurut is made in all regions of Kyrgyzstan in summer periods by families in homes. If some families produce kurut to eat in the winter, other families produce it in order to obtain additional income. In Kyrgyzstan, it is possible to buy kurut everywhere because it is widely consumed. It is also a product consumed by children fondly. Almost all of the kuruts sold in Kyrgyzstan are produced by traditional methods. There are only a few companies producing kurut industrially (Figure 2). In Kyrgyzstan, kuruts are produced in different sizes, compositions and figures. There are varieties such as fruity kurut, kurut with red pepper, smoked kurut, kurut with dill.



Figure 2. Types of kurut produced industrially in Kyrgyzstan

## 4. CONCLUSION

It is determined that kurut, which is traditionally produced in Turkey and Central Asian countries has a high nutritional value and mineral matter content. The content of this product, which provide the recycle of the buttermilk, varies from product to product. The difference between these values is dependent on the raw materials and methods that was used in the production of kurut.

According to the researches, it has been determined that the kuruts contain significant pathogenic microorganisms. The main reason is not paying enough attention to hygienic conditions during the production and storage of kuruts. In addition, there are no scientific researches on kuruts produced in Kyrgyzstan.

Despite of being traditionally produced in many regions of Turkey, kurut is not much known among the people. In the process of developing industrial size production techniques of kurut, the developing of controlled storage conditions with standard production methods will prevent fluctuations in the chemical composition of the product as well as increase the microbiological quality. By modernizing kurut production techniques, production of this dairy product will be encouraged not only in rural areas, but also in urban environments, so that it can be offered for larger consumer groups.

#### REFERENCES

Akyüz, N., and Gülümser, S., 1987. Kurutun Yapılışı ve Bileşimi Üzerine Bir Araştırma.

- Akyüz, N., Çoşkun, H., Bakırcı, İ., Çon, A. H., 1993. Van ve yöresinde imal edilen kurutlar üzerinde bir araştırma. GIDA, 18 (4): 253-257.
- Atasever, A., M., 2007. Erzurum Ve Bingöl Yöresinden Toplanan Kurut Örneklerinin Mikrobiyolojik Ve Kimyasal Nitelikleri. Yüksek Lisans Tezi. Atatürk Üniversitesi Sağlık Bilimleri Enstitüsü Gıda Hijyeni Ve Teknolojisi Anabilim Dalı, Erzurum.
- Çetinkaya, A., 2004. Kars Kremalı Kurut. 1. Geleneksel Gıdalar Sempozyumu, 23-24 Eylül, Van, Türkiye.
- Eralp, M., 1953. Kurut Yapılışı ve Terkibi. Ankara Üniversitesi, Ziraat Fakültesi Yıllığı, Fasikül: 3-4, Ankara.
- Güven, M., and Karaca., O. B., 2009. Van Ve Şırnak İllerinden Temin Edilen Kurutulmuş Yoğurtların (Kurut) Bileşim Özellikleri. GIDA 34 (6): 367-372.
- Kamber, U., 2008. The manufacture and some quality characteristics of kurut, a dried dairy product. International Journal of Dairy Technology. No 2, Vol 61.
- Karabulut, I., Hayaloglu, A. A., and Yıldırım, H., 2007. Tin-layer drying characteristics of kurut, a Turkish dried dairy byproduct. International Journal of Food Science and Technology 42:1080-1086.
- Karaçıl, M. Ş., and Acar Tek N., 2013. Dünyada Üretilen Fermente Ürünler: Tarihsel Süreç Ve Sağlık İle İlişkileri. Uludağ Üniversitesi Ziraat Fakültesi Dergisi, Cilt 27, Sayı 2, 163-173.
- L iu, W. J., Sun, Z. H., Zhang, Y. B., Zhang, C. L., Menghebilige, Yang, M., Sun, T. S., Bao, Q. H., Chen, W., and Zhang, H. P., 2012. A survey of the bacterial composition of kurut from Tibet using a culture-independent approach. Journal of Dairy Science 95: 1064–1072.
- Öksüztepe, G., İncili, G. K., and Uysal İ. A., 2013. Elazığ'da Satılan Çökelek Ve Kurutların Mineral Madde Ve Ağır Metal Düzeyleri. E-Journal of New World Sciences Academy. NWSA-Veterinary Sciences, 3B0022, 8, (3), 1-9.
- Patır, B., and Ateş, G., 2002. Kurutun Mikrobiyolojik ve Kimyasal Bazı Nitelikleri Üzerine Araştırmalar. Turkish Journal of Veterinary Animal and Animal Science 26: 785–792.
- Say, D., Soltani, M., and Güzeler, N., 2015. Kurutulmuş Yoğurtlar: Kurut ve Kashk. Pamukkale Univiversitesi Muhendislik Bilimler Dergisi 21(9), 428-432.



# The Effects of Black Pepper and Sodium Ascorbate on Microbiological Properties of Sucuk

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Abstract: Sucuk, a dry-fermented sausage, is a traditional Turkish meat products that are widely consumed. The aim of this research is to determine the effects of levels of black pepper (5 g/kg, 10 g/kg or 15 g/kg) and the use of sodium ascorbate on the microbiological properties of sucuk. Lactobacillus plantarum GM77 (107 cfu/g) + Staphylococcus xylosus GM92 (106 cfu/g) strains isolated and identified from sucuk were used as starter culture. Ripening (fermentation and drying) was carried out under controlled conditions. At the end of ripening, samples were subjected to microbiological analysis (lactic acid bacteri, Micrococcus/Staphylococcus, Enterobacteriaceae and mold-yeast). The use of sodium ascorbate and black pepper level were not significantly effective on the number of lactic acid bacteria (P>0.05). Sodium ascorbate x black pepper interaction had a very significant effect on the number of lactic acid bacteria (P<0.01). The strain of L.plantarum GM77 used as starter culture showed a good growth. The number of Microccocus/Staphylococcus was not significantly affected by the sodium ascorbate factor (P>0.05). In contrast, the black pepper level had a significant effect on the number of Micrococcus/Staphylococcus (P<0.05). The high black pepper level (10 g/kg or 15 g/kg) reduced the number of Microccocus/Staphylococcus (P<0.05). However, the number of Microccocus/Staphylococcus in all sucuk groups was above 1x106 cfu/g. The interaction of sodium ascorbate and black pepper did not affect the number of Micrococcus/Staphylococcus (P>0.05). The use of sodium ascorbate and black pepper level as well as interaction of sodium ascorbate and black pepper had no significant effect on the number of yeast and mould (P>0.05). On the other hand, the number of Enterobacteriaceae was below the detectable level (<2 log cfu/g) in all sucuk groups.

Keywords: Sucuk, black pepper, sodium ascorbate, *Micrococcus/Staphylococcus*, lactic acid bacteria.

# **1. INTRODUCTION**

Sucuk, a traditional Turkish dry-fermented sausage, is made from beef and/or water buffalo and/or mutton meat. The sucuk production process is based on fermentation and drying (ripening) and does not include smoke or heat treatment. Beef fat and/or sheep tail fat is used in the production. Curing agents (nitrate and/or nitrite), salt and spices are also used in the sucuk production (Gökalp et al. 2004). After stuffing the sucuk batter into air-dried bovine small intestines or casings of similar characteristic, ripening is carried out under natural or climatic conditions. In the fermentation stage, temperature vary between 12 °C and 26°C (Ertaş and Göğüş, 1980; Gökalp and Ockerman, 1985; Soyer et al., 2005).

Spices include a wide range of ingredients that comprise 0.5 to 2% of most fermented sausage. The addition of spices is very useful in the production of dry-fermented sausage (Ordonez et al., 1999). The major purpose of adding spices is to provide distinct flavor to the final product (Chi and Wu, 2007). Some spices are also added as coloring agents. However, they are a source of many other substances, such as sugars, nitrates, and metallic ions. It was reported that spices added to meat could accelerate lactic acid production by the lactic starter culture (Verluyten et al. 2004). On the other hand, spices and their extracts have preservative and natural antioxidant properties. The level of spices used in the production depends on the type of sausage. Black pepper is one of the most important spice in the production of sucuk. This spice is usually added to 5-10 g / kg of the sucuk batters (Sallan et al, 2019).

Sodium ascorbate is commonly used in meat product. It increases the rate of some of chemical reactions to expedite the development of cured color. Therefore, the use of ascorbate results in a more rapid depletion of residual nitrite during the curing process (King et al, 2016). There is no study on the effect of black pepper and sodium ascorbate on the microbiological properties of fermented sausages. The aim of the study was to determine the effects of black pepper and sodium ascorbate on the microbiological properties of sucuk. Sucuk batters was prepared by adding black pepper and / or sodium ascorbate at different levels and the effects of black pepper and ascorbate factors on microbiological properties of the sucuk were investigated.

# 2. MATERIALS AND METHODS

# Materials

Beef meat from the shoulder and beef fat were obtained from Erzurum Meat Processing Plant of General Directory of Meat and Milk Board. Raw meat was trimmed of visible fat and connective tissue. Lean meat and beef fat were separately cut into small pieces. These small pieces were mixed by hand and divided into six batches. Then, these batches were vacuum-packaged and stored at -18 °C until production. All treatments, 5 kg each (4 kg lean meat+1 kg beef fat), were repeated two times from separate sources of raw materials at two different times. *Lactobacillus plantarum* GM77+*Staphylococcus xylosus* GM92 were used as starter cultures (Kaban and Kaya, 2009).

# Sucuk formulation and preparation

The following ingredients were used per kg of beef meat and beef fat (80:20) : 20 g salt, 10 g garlic, 4 g sucrose, 7 g red pepper, 9 g cumin, 2.5 g allspice and 0.15 g sodium nitrite. Considering this formulation, 6 sucuk batters were prepared using different levels of black pepper (5, 10 or 15 g kg<sup>-1</sup>) and sodium ascorbate (0 or 568 mg kg<sup>-1</sup>). The batters were prepared in a laboratory-type bowl cutter (MTK 662; MADO GmbH, Dornhan, Germany). *Lactobacillus plantarum* GM77 ( $10^7$  cfu g<sup>-1</sup>) + *Staphylococcus xylosus* GM92 ( $10^6$  cfu g<sup>-1</sup>) were added to all sucuk batters as starter culture. Each group weighing 200 ± 10 g was stuffed into collagen casings (38 mm; Naturin GmbH & Co., Weinheim Germany) with a laboratory-type filling machine (MTK 591, MADO GmbH, Dornhan, Germany). Prepared sausages were ripened in an automatic climate unit (REICH Thermoprozesstechnik GmbH, Schechingen, Germany). Ripening programs were as follows:  $22 \pm 1$  °C for 2 days,  $90 \pm 2\%$  for relative humidity (RH), 2 days at  $20 \pm 1$  °C and RH 88 ±2%, 3 days at  $18 \pm 1$  °C and RH 80 ± 2%. The air flow was maintained between 0.5ms–1 in the first 3 days of ripening and gradually decreased to 0.1ms<sup>-1</sup> over the following days.

# Methods

For microbiological analysis, 25 g of the sample was taken into a sterile bag and added to 225 ml of sterile physiological saline and homogenized in stochmacher (Lab Stomacher Blander 400 - BA 7021). MRS Agar (de Man Rogosa Sharpe Agar, Merck) was used for the lactic acid bacteria count in sausage samples. The lactic acid number was determined by considering catalase (-) colonies after anaerobic incubation (Anaerocoult A, Merck) for 2 days at 30 ° C (Baumgart *et al.* 1993).MSA (Mannitol Salt Phenol Red Agar, Merck) was used for *Micrococcus / Staphylococcus* counting. The number of *Micrococcus / Staphylococcus* was determined by considering catalase (+) colonies after incubation for 2 days at 30 ° C (Baumgart *et al.* 1993). RBC (Rose Bengal Chloroamphenicol, Merck) agar was used for the mould-yeast enumeration. The plates were incubated at 25° C for 5 days. Enterobacteriacea were determined on VRBD (Violet Red Bile Dextrose, Merck) agar, incubation was carried out at 30°C for 48 h in anaerobic conditions (Anaerocoult A, Merck) (Baumgart *et al.* 1993).

#### 3. RESULTS AND DISCUSSION

The overall effects of samples manufactured in different amounts of black pepper and ascorbate on the numbers of lactic acid bacteria, *Micrococcus Staphylococcus*, yeast-mould and Enterobacteriacea were showed in Table 1.The use of sodium ascorbate and black pepper level were not significantly effective on the number of lactic acid bacteria (P>0.05). In contrast, sodium ascorbate x black pepper interaction had a very significant effect on the number of lactic acid bacteria (P<0.01). In the study, it can be said that the *L.plantarum* GM77 strain used as starter culture showed a good growth and it was not significantly affected by black pepper and ascorbate (p>0.05) (Table 1). Lactic acid bacteria in fermented meat products such as sucuk, salami and Rohwurst are microorganisms with technological properties that are important for product safety. The decrease in pH occurs due to acidification during the fermentation phase, which contributes to product safety. In addition, the drop of pH to the isoelectric point accelerates the drying. Acidification as a result of the activity of lactic acid bacteria is also effective in the development of sensory characteristics of the product (Lücke 1985; Kaya and Kaban 2014).

*Microccocus/Staphylococcus* are an important microorganisms in fermented meat products. They play an important role in the color formation and stabilization, aroma development and delaying the lipid oxidation. The number of *Microccocus/Staphylococcus* was not significantly affected by the sodium ascorbate factor (P>0.05) (Table 1). However, the black pepper level had a significant effect on the number of *Microccocus/Staphylococcus* (P<0.05). The number of *Microccocus/Staphylococcus* decreased in the present of black pepper level of 10 g/kg (P<0.05). The similar result was observed in the group with black pepper level of 15 g/kg. However, the number of *Microccocus/Staphylococcus* in all

sucuk groups was above  $1 \times 10^6$  cfu/g. Moreover, the interaction of sodium ascorbate and black pepper did not affect the number of *Micrococcus/Staphylococcus* (P>0.05) (Table 1).

The effect of sodium ascorbate, black pepper level and sodium ascorbate x black pepper level on yeast and mold were not found significantly (p > 0.05). As can be seen from the results given in Table 1, the averages for each group were similar. Enterobacteriaceae are sensitive to acid and water activity (Lücke, 1985). In this study, the number of Enterobacteriaceae in fermented sausages produced by using sodium ascorbate and starter culture were found to be less than  $10^2$  cfu / g in all groups. It has been usually determined that Enterobacteriaceae family members in fermented sausage has been detected below the limit in many research (Kaya and Gökalp 2004a,b; Kaban and Kaya 2006; Gençcelep *et al.* 2007).

**Table 1.** Overall effect of sodium ascorbate and black pepper level on the number of lactic acid bacteria ,*Microccocus/ Staphylococcus*, mould-yeast and *Enterobacteriaceae* of sucuk samples (mean  $\pm$ SD) (log cfu/g)

|   | n  | Lactic acid<br>bacteria | Microccocus/<br>Staphylococcus | Mould-yeast | Enterobacteriaceae |
|---|----|-------------------------|--------------------------------|-------------|--------------------|
| Sodium ascorbate (mg kg <sup>-1</sup> )(SA) |    |                         |                                |             |                    |
| without ascorbate                           | 12 | 8,68±0,26a              | 6,62±0,28a                     | 2,84±0,43a  | <2                 |
| with ascorbate                              | 12 | 8,68±0,18a              | 6,62±0,14a                     | 2,66±0,52a  | <2                 |
| Significance                                |    | NS                      | NS                             | NS          |                    |
| Black pepper (g kg <sup>-1</sup> ) (BP)     |    |                         |                                |             |                    |
| 5   | 8  | $8{,}65\pm0.16a$        | 6,78±0,16a                     | 2,79±0,42a  | <2                 |
| 10  | 8  | $8{,}60\pm0.28a$        | 6,52±0,20b                     | 2,81±0,58a  | <2                 |
| 15  | 8  | $8,80 \pm 0.16a$        | 6,56±0,22b                     | 2,65±0,46a  | <2                 |
| Significance                                |    | NS                      | *                              | NS          |                    |
| SA x BP                                     |    | **                      | NS                             | NS          |                    |

a–b: Any two means in the same column having the same letters in the same section are not significantly different at p > 0.05, \*\*p < 0.01, \*p < 0.05, NS: not significant; SD: standard deviation.

# 4. CONCLUSION

In conclusion, black pepper is an important spice for the sucuk formulation. It is added to sucuk batter at the different levels. According to the results of this study, the level of black pepper used in sucuk production did not show any effect on lactic acid bacteria which are technologically important microorganisms. However, the black pepper level had a significant effect on the number of Micrococcus/Staphylococcus. On the other hand, use of sodium ascorbate in sucuk had no a significant effect on the microbiological properties of the product.

#### REFERENCES

- Baumgart J, Eigener V, Firnhaber J, Hildebrant G, Reenen Hoekstra E S, Samson R A, Spicher G, Timm F, Yarrow D & Zschaler R (1993). Mikrobilogische Unterschung von Lebensmitteln, (3., aktualisierte und erw. Aufl.), Hamburg, Germany.
- Chi S P & Wu Y C (2007). Spices and Seasonings.Fermented Meat and Poultry. Editor Fidel Toldra. ISBN 13:978-0-8138-1477-3, Black Publishing, USA.
- Ertaş A H & Göğüş A K (1980). Değişik Oranlarda Kuyruk Yağı ve Farklı Starter Kullanılmış olan Sucuklar Üzerine Araştırmalar. Doğa Bilim Dergisi- Veterinerlik ve Hayvancılık/Tarım ve Ormancılık, 4(3): 48–53.
- Gençcelep H, Kaban G & Kaya M (2007). Effects of Starter Cultures and Nitrite Levels on Formation of Biogenic Amines in Sucuk. Meat Science, 77(3): 424-430.
- Gökalp H Y, Kaya M & Zorba Ö (2004). Et Ürünleri İşleme Mühendisliği. Atatürk Üniversitesi Ziraat Fakültesi Yayınları No:320, 468 s, Erzurum.
- Gökalp H Y & Ockerman H W (1985). Herstellung von Rohwurst Türkischer Art (soud-jouk) mit Hilfe verschiedener Starterkulturen und unter verschiedenen Reifung-stemreraturen. I. Wachstum der Gesamtkeimzahlen sowie der psychrophilen, pro-teoltischen und lipolytischen Keime. Fleischwirtschaft 66(10): 1248–54.
- Kaban G & Kaya M (2006). Effect of starter culture on growth of Staphylococcus aureus in sucuk. Food Control, 17: 797-801.

- Kaban G & Kaya M (2009). Effects of Lactobacillus plantarum and Staphylococcus xylosus on the Quality Characteristics of dry fermented sausage "sucuk". Journal of Food Science, 74(1): S58-S63.
- Kaya M & Gökalp H Y (2004a). The Behavior of Listeria monocytogenes in Sucuks Produced with Different Lactic Starter Cultures. Turkish Journal of Veterinary and Animal Science, 28 (6): 1113-1120.
- Kaya M & Gökalp H Y (2004b). The Effects of Starter Cultures and Different Nitrite Doses on the Growth of Listeria monocytogenes in sucuk production. Turkish Journal of Veterinary and Animal Science, 28 (6): 1121-1127.
- Kaya M & Kaban M (2014). Fermente Et Ürünleri. Gıda Biyoteknolojisi. Editör Necla ARAN, ss. 157-195, 5. Basım, ISBN: 978-605-133-134-8, Nobel Yayıncılık, İstanbul.
- King A M, Glass K A, Milkowski A L, Seman D L & Sindelar J J (2016). Modeling the Impact of Ingoing Sodium Nitrite, Sodium Ascorbate, and Residual Nitrite Concentrations on Growth Parameters of Listeria monocytogenes in Cooked, Cured Pork Sausage. Journal of Food Protection. 79(2): 184-193.
- Lücke F K (1985). Mikrobiologische Vorgange bei der Herstellung von Rohwurst und Rohschinken. In: Mikrobiologie und Qualitat von Rohwurst und Rohschinken. Bundesanstalt für Fleischforschung, Kulmbach, 85-102, Germany.
- Ordonez J A, Hierro E M, Bruna J M & De Lahoz L (1999). Changes in the components of dry-fermented sausages during ripening. Critical Reviews in Food Science and Nutrition, 39(4): 329-367.
- Sallan S, Kaban G & Kaya M (2019). Nitrosamines in sucuk: Effects of black pepper, sodium ascorbate and cooking level, Food Chemistry, 288: 341-346.
- Soyer A, Ertaş A H & Üzümcüoğlu Ü (2005). Effect of Processing Conditions on The Qualityof Naturally Fermented Turkish Sausages (Sucuks). Meat Science, 69, 135–41.
- Verluyten J, Leroy F & De Vuyst L (2014). Effects of Different Spices Used in Production of Fermented Sausages on Growth of and Curvacin A Production by Lactobacillus curvatus LTH 1174. Applied and Environmental Microbiology. 70(8): 4807-4813.



# The Toxic Properties and Bioactive Components of Sambucus ebulus L.

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**Abstract:** In this paper, it is aimed to give information about toxic properties and bioactive components of Sambucus ebulus L. (dwarf elder) a part of natural vegetation of Kastamonu.

Sambucus ebulus L. is a folk medicine widely used plant which is known as 'cüce mürver' in Anatolia. The leaves of the cüce mürver applied externally are used to for healing various types of inflammatory disorders (edema, urticarial, eczema, burns and abscess) in Turkish folk medicine. In addition, one of the remedies of open wounds is cüce mürver has been used. Also, Khamaan is the name of cüce mürver in traditional persian medicine. Khamaan an analgesic therapy for various painful diseas such as bone/joint disorders and joint pain. The raw berries of cüce mürver are known as poisonous while high-dose consumption of the other parts may lead to toxicity. The vomitory toxicity might occure due to excessive consumption of cüce mürver fruits, particularly in children. Hepatotoxicity and nephrotoxicity have also been seemed to be induced by ethyl acetate extract of cüce mürver in rat model. There are several bioactive compounds in cüce mürver, for instance: steroids, tannins, caffeic acid derivatives, flavonoids, glycosides, cardiacglycosides, lectins, and volatile substances. It is thought that the responsible anti-ulcerogenic and anti-inflammatory properties of cüce mürver, is its flavonoids. In addition, the positive effects of fruit infusion on lipid metabolism have been showed. Lectins are known as cell-agglutinating proteins with carbohydrate binding ability found in all organisms, and its special types are irreversibly inhibits protein synthesis. Cüce mürver is an plant that contain lectins in several tissues such as rhizome, fruit and leave. The palmitic acid, oleic acid, a-linolenic and linoleic acid as essential polyunsaturated fatty acids were founded in seeds of cüce mürver.

Keywords: Sambucus ebulus, toxicity, lectins, Kastamonu.

# **1. INTRODUCTION**

Sambucus ebulus is a perennial plant which its leaves have 5 to 9 leaflets with fetid smell. The leaves are pinnate, and opposite to each other. Plant has an underground stem rhizome and unbranched erect stems grow in large groups near each other. The stems end in a corymb, 10 to 15 cm in diameter, with white (occasionally pink) flat-topped flowers. Sambucus ebulus fruit is dark blue to violet in color. Sambucus ebulus L. is widely used plant in folk medicine. The plant is a member of Caprifoliaceae family which commonly grows in north of Türkiye and known as 'cüce mürver' in Anatolia. The leaves of the cüce mürver are used to for healing various types of inflammatory disorders (urticarial, edema, eczema, burns and abscess) and applies externally in Turkish folk medicine (Balkan et al., 2017). In addition, the cüce mürver has been used as one of the remedies of open wounds. (Ghabaee et al., 2017). A pilot study demonstrated that topical treatment of gel containing extracts of cüce mürver can be recommended for patients with knee osteoarthritis (Jabbari et al., 2016). Also, Khamaan is the name of cüce mürver in traditional persian medicine. Khamaan an analgesic therapy for various painful diseas such as bone/joint disorders and joint pain (Jabbari et al., 2016). In this paper, it is aimed to give information about toxic properties and bioactive components of Sambucus ebulus L. (dwarf elder) which is a member of natural vegetation of Kastamonu city.

# 2. MATERIALS AND METHODS

In the preparation of the paper, the compilation of the information from the literature was used.

# 3. RESULTS AND DISCUSSION

# Toxicity of Sambucus ebulus

The raw berries of cuce murver are known as poisonous while high-dose consumption of the other parts may lead to toxicity. The vomitory toxicity might occure due to excessive consumption of cuce murver fruits, particularly in children (Suntar et al. 2010). Stomach upset also could happen by the fruit of some sambucus species. But, the toxicity of berries of cuce murver is at such a low level that could be prevented by heating process such as cooking (Shokrzadeh et al. 2010). The short-term heating process could eliminate toxic lectins of cuce murver and any significant reduction in polyphenol and antioxidant contents is not seen (Jimenez 2014).

Hepatotoxicity and nephrotoxicity have also been seemed to be induced by ethyl acetate extract of cüce mürver in rat model (Saravi and Shokrzadeh, 2009). Lethal dose 50% (LD50) of rhizome methanol extract from cüce mürver is 600 mg kg<sup>-1</sup> in rats (Ahmadiani et al. 1998). Cüce mürver fruits contain the ebulin f structurally similar to ricin. It has been found that intraperitoneal toxicity of ebulin f leads to specific regulation of the intestines (Jiménez et al., 2013). Intraperitoneal administration of ebulin f was significantly less toxic than ricin in mice and targets were intestinal cryptic cells with apoptosis. The sensitivity of small intestine crypts were more higher than large intestine crypts (Jiménez et al., 2013).

# **Bioactive components of Sambucus ebulus**

The elderberries have important nutritional value due to compounds such as vitamins, sugars, fibers and minerals in their compositions (Vlachojannis et al., 2010). There are several bioactive compounds in cüce mürver, for instance: steroids, tannins, caffeic acid derivatives, flavonoids, glycosides, cardiacglycosides, lectins, and volatile substances (Balkan et al. 2017). It is thought that the responsible anti-ulcerogenic and anti-inflammatory properties of cüce mürver, is its flavonoids (Ebrahimzadeh et al., 2014). In addition, the positive effects of fruit infusion on lipid metabolism have been showed (Ivanova et al., 2014). After isolating the active ingredients (isorhamnetin-3-O-glucoside, quercetin-3-O-glucoside, and ursolic acid) from the leaves of its, the wound healing potential, anti-inflammatory effect and anti-ulcerogenic activity of cüce mürver extracts have been mentioned in some previous studies. (Süntar et al., 2010; Schwaiger et al., 2011; Yesilada et al., 2014). Newly, the chemical composition of leaves essential oil of the cüce mürver was extensively investigated and demonstrated that presence of more than sixty mono- and sesqui- terpenoids (Feizbakhsh et al., 2014).

The ripe cüce mürver fruit's colur is dark blue to violet due to several anthocyanidins such as cyanidin-3-sambubioside-5-glucoside, cyanidin-3-O-sambubioside, cyanidin-3,5-diglucoside and cyanidin-3-O-glucoside (Mikulic-Petkovsek et al., 2014). In addition, it was revealed that cyanidin-3-O-glucoside inhibits the proliferation of cancer cells and tumor xenographs in immune-deficient mice (Ding et al., 2006).

Two iridoids were recently isolated (sambulin A, sambulin B) from the leaves of cuce murver (Atay et al., 2015). Iridoids have cyclopentanopyranoid skeletons and are monoterpenic compounds have been shown to exert hepatoprotective, choleretic, antihypertensive, hypolipidemic, hypoglycaemic, anti-spasmodic, immunomodulator, anti-inflammatory and anti-viral activities (Ghisalberti, 1998). The in vitro anti-inflammatory activities sambulin A and sambulin B, which is two iridoids isolated from the leaves of cuce murver was investigated by Balkan et al. (2017) for the first time. In their study, Sambulin B significantly inhibited extracellular signal-regulated kinases (ERK)-1 and -2, c-Jun N-terminal kinase (JNK) phosphorylations suggested as a possible mechanism for the in vitro anti-inflammatory activity of sambulin B. Lectins are known as cell-agglutinating proteins with carbohydrate binding ability found in all organisms, and its special types are irreversibly inhibiting protein synthesis (Schrot et al., 2015; Di Maro et al., 2014). Thus its are called the ribosome-inactivating proteins (RIPs). RIPs is a member of enzymes class EC 3.2.2.22 which has rRNA N-glycosidase activity. RIPs causes to the release of a adenine residue from sarcinricin loop of the large rRNA responsible for the interaction of the ribosome with the elongation factor. Hence, the irreversible inhibition of protein synthesis takes place (Endo and Tsurugi, 1988; Iglesias et al. 2018). There are two types RIPs. If there is a lectin chain in structure of RIP, it is a type 2 RIPs or there is no any lectin chain in structure, it is a type 1 RIPs. Hence RIPs which consist of a single enzymatic active (A) chain are type 1 RIPs and type 2 RIPs are composed of an A chain linked by a disulphide bond to a (B) chain showed lectin activity. A particular RIP may differ and this may be activating to apoptosis. Besides, there are several strongly toxic RIPs on cancer cells and these RIPs exhibit low toxicity towards normal cells. Moreover, they lead to apoptosis and impede or inhibit tumor growth by this way (Zheng et al., 2015). Therefore, RIPs or its conjugates are promises hope for the production of selective anticancer and antiviral pharmaceutics (Xiong et al. 2009; Fang et al., 2011). RIPs have been tested in experimental therapies against different malignancies as the toxic part of several conjugates (Polito et al., 2011; Polito et al., 2016). It was shown that, the presence of some RIPs and lectins in several tissues of cuce
mürver. There are type 2 RIP ebulin 1 (A-B type); a monomeric lectin SELIm and a dimeric lectin SELId in its leaves. The rhizome contained two type 2 RIPs (ebulin r1 and r2), tetrameric type 2 RIP SEA and the monomeric lectin SEAII. Ebulin f and the lectin SELfd are lectins which found in fruits. The SEA is specific for sialic acid and the other hololectins and B chains of ebulins are galactose specific lectins (Iglesias et al., 2018). Monossaccharides or oligossacharides which inhibit lectin-dependent agglutination or precipitation reactions mainly drive specificity of lectin. Lectins are strong candidates for use in the construction of the third generation coated nanosystems specific to target as a carrier for drug delivery due to its non-immunogenic properties and multivalent properties (Kompella and Lee, 2011). Carbohydrate binding properties are the reason for the use of lectins as biomarkers in cancer research (Dai et al., 2009). In the construction of conjugates and immunotoxins for specific targets, RIPs of some bacterial and plant species stand out as toxic parts (Gilabert-Oriol et al., 2014). Also, RIPs have been exhibited to increase resistance against virus and other parasites in transgenic plants, (Di and Tumer 2015). Moreover, some essential polyunsaturated fatty acids such as palmitic acid, oleic acid, a-linolenic and linoleic acid as were founded in seeds of cüce mürver (Dulf et al., 2013).

#### 4. CONCLUSION

Consequently, the bioactive properties of cuce murver above described make it a good candidate for developing standardized preparations as drugs for the several health disorders and the pharmaceutical industry. Hence, it is thought that it can contribute to the economy of Kastamonu city by using the cuce murver a part of the natural vegetation of the city of Kastamonu, for pharmaceutical industry.

#### REFERENCES

- Ahmadiani, A., Fereidoni, M., Semnanian, S., Kamalinejad, M., Saremi S., 1998. Antinociceptive and anti- inflammatory effects of Sambucus ebulus rhizome extract in rats. Journal of Ethnopharmacology 61: 229-235.
- Atay, İ., Kırmızıbekmez, H., Gören, A.C., Yesilada, E., 2015. Secondary metabolites from Sambucus ebulus. Turkish Journal of Chemistry 39: 34-41.
- Balkan, İ.A., Akülke, A.Z.İ., Bağatur, Y., Telci, D., Gören, A.C., Kırmızıbekmez, H., Yesilada, E., 2017 Sambulin A and B, nonglycosidic iridoids from Sambucus ebulus, exert significant in vitro anti-inflammatory activity in LPS-induced RAW 264.7 macrophages via inhibition of MAPKs's phosphorylation. Journal of ethnopharmacology 206: 347-352.
- Dai, Z., Zhou, J., Qiu, S.J., Liu, Y.K., Fan, J., 2009. Lectin-based glycoproteomics to explore and analyze hepatocellular carcinomarelated glycoprotein markers. Electrophoresis 30: 2957-2966.
- Di Maro, A., Citores, L., Russo, R., Iglesias, R., Ferreras, J.M., 2014. Sequence comparison and phylogenetic analysis by the maximum likelihood method of ribosome-inactivating proteins from angiosperms, Plant Mol. Biol. 85: 575-588.
- Ding, M., Feng, R., Wang, S.Y., Bowman, L., Lu, Y., Qian, Y., Castranova, V., Jiang, B.H., Shi, X., 2006. Cyanidin-3-glucoside, a natural product derived from blackberry, exhibits chemopreventive and chemotherapeutic activity. The Journal of Biological Chemistry 281: 17359-17368.
- Ebrahimzadeh, M.A., Rafati, M.R., Damchi, M., Golpur, M., Fathiazad, F., 2014. Treatment of paederus dermatitis with Sambucus ebulus lotion. Iranian Journal of Pharmacology Research 13: 1065-1071
- Endo, Y., Tsurugi, K., 1988. The RNA N-glycosidase activity of ricin A-chain. The characteristics of the enzymatic activity of ricin A-chain with ribosomes and with rRNA, The Journal of Biological Chemistry. 263: 8735-8739.
- F Fang, E., B Ng, T., C Shaw, P., & NS Wong, R., 2011. Recent progress in medicinal investigations on trichosanthin and other ribosome inactivating proteins from the plant genus Trichosanthes. Current medicinal chemistry 18(28): 4410-4417.
- Feizbakhsh, A., Pazoki, H., Mohammadrezaei, V., Ebrahimzadeh, M.A., 2014. Effect of phytochromones on the composition of Sambucus ebulus leaf essential oil. Tropical Journal of Pharmaceutical Research 13: 581-586.
- Ghabaee, D. N. Z., Ebrahimzadeh, M. A., Akbari, J., & Amiri, F. T., 2017. Wound healing activity of Sambucus ebulus. International Journal of Pharmaceutical Sciences and Research, 8(1): 132-135.
- Ghisalberti, E.L., 1998. Biological and pharmacological activity of naturally occurring iridoids and secoiridoids. Phytomedicine 5: 147-163.
- Gilabert-Oriol, R., Weng A., von Mallinckrodt, B., Melzig, M. F., Fuchs, H., & Thakur, M., 2014. Immunotoxins constructed with ribosome-inactivating proteins and their enhancers: A lethal cocktail with tumor specific efficacy. Current Pharmaceutical Design 20: 6584–6643.
- Iglesias, R., Ferreras, J. M., Di Maro, A., & Citores, L., 2018. Ebulin-RP, a novel member of the Ebulin gene family with low cytotoxicity as a result of deficient sugar binding domains. Biochimica et Biophysica Acta (BBA)-General Subjects 1862(3): 460-473.

- Ivanova, D., Tasinov, O., Kiselova-Kaneva, Y., 2014. Improved lipid profile and increased serum antioxidant capacity in healthy volunteers after Sambucus ebulus L. fruit infusion consumption, International Journal of Food Sciences and Nutrition 65:6, 740-744.
- Jabbari, M., Hashempur, M. H., Razavi, S. Z. E., Shahraki, H. R., Kamalinejad, M., & Emtiazy, M., 2016. Efficacy and short-term safety of topical Dwarf Elder (Sambucus ebulus L.) versus diclofenac for knee osteoarthritis: a randomized, double-blind, active-controlled trial. Journal of ethnopharmacology 188: 80-86.
- Jiménez, P., Gayoso, M., Tejero, J., Cabrero, P., Córdoba-Díaz, D., Basterrechea, J. E., & Girbés, T., 2013. Toxicity in mice of lectin ebulin f present in dwarf elderberry (Sambucus ebulus L.). Toxicon 61: 26-29.
- Jimenez P, Cabrero P, Basterrechea JE, et al., 2014. Effects of short-term heating on total polyphenols, anthocyanins, antioxidant activity and lectins of different parts of dwarf elder (Sambucus ebulus L.). Plant Foods for Human Nutrition 69: 168-174.
- Kompella, U.B., Lee, V.H., 2001. Delivery systems for penetration enhancement of peptide and protein drugs: design considerations. Advanced drug delivery reviews 46(1-3): 211-245.
- Mikulic-Petkovsek, M., Schmitzer, V., Slatnar, A., Todorovic, B., Veberic, R., Stampar, F., Ivancic, A., 2014. Investigation of anthocyanin profile of four elderberry species and interspecific hybrids. Journal of Agricultural and Food Chemistry 62: 5573-5580.
- Polito, L., Bortolotti, M., Pedrazzi, M., & Bolognesi, A,. 2011. Immunotoxins and other conjugates containing saporin-s6 for cancer therapy. Toxins 3(6): 697-720.
- Polito, L., Djemil, A., & Bortolotti, M., 2016. Plant toxin-based immunotoxins for cancer therapy: A short overview. Biomedicines 4(2): 12.
- Saravi, S.S, Shokrzadeh, M., 2009. Anti-inflammatory, toxic effects, biochemical and pathological analysis in presence or lack of vitamins C and E, and cytotoxicity of n-hexane, methanolic and ethyl acetate extracts of Sambucus ebulus. Toxicology Letters 189: 166-167.
- Schrot, J., Weng, A., Melzig, M.F., 2015. Ribosome-inactivating and related proteins, Toxins 7: 1556-1615.
- Shokrzadeh, M., Saeedi Saravi, S., 2010. The chemistry, pharmacology and clinical properties of Sambucus ebulus: a review. Journal of Medicinal Plants Research 4: 95-103.
- Süntar, I.P., Akkol, E.K., Yalçın, F.N., Koca, U., Keles, H., Yesilada, E., 2010. Wound healing potential of Sambucus ebulus L. leaves and isolation of an active component, quercetin 3-O-glucoside. Journal of Ethnopharmacology 129: 106-114.
- Schwaiger, S., Zeller, I., Pölzelbauer, P., Frotschning, S., Laufer, G., Messner, B., Pieri, V., Stuppner, H., Bernhard, D., 2011. Identification and pharmacological characterization of the anti-inflammatory principal of the leaves of dwarf elder (Sambucus ebulus L.). Journal of Ethnopharmacology 133: 704-709.
- Vlachojannis J, Cameron M, Chrubasik S., 2010. A systematic review on the sambuci fructus effect and efficacy profiles. Phytotherapy Research. 24: 1-8.
- Xiong, S. D., Yu, K., Liu, X. H., Yin, L. H., Kirschenbaum, A., Yao, S., ... & Ho, S. M., 2009. Ribosome-inactivating proteins isolated from dietary bitter melon induce apoptosis and inhibit histone deacetylase-1 selectively in premalignant and malignant prostate cancer cells. International Journal of Cancer 125(4): 774-782.
- Yesilada, E., Gürbüz, I., Toker, G., 2014. Anti-ulcerogenic activity and isolation of the active principle from Sambucus ebulus L. leaves. Journal of Ethnopharmacology 153: 478-483.
- Zeng, M., Zheng, M., Lu, D., Wang, J., Jiang, W., & Sha, O., 2015. Anti-tumor activities and apoptotic mechanism of ribosomeinactivating proteins. Chinese journal of cancer 34(3): 30-40.



# The Levels of Patulin Contamination in Apple and Apple Products in Türkiye

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Abstract: The purpose of this report is to provide information about the level of contamination toxic patulin in apple and apple products in Türkiye. The Penicillium, Aspergillus and Byssochlamys produces Patulin and apple products such as apple juice, apple cider and apple past may include these molds. The patulin firstly isolated as an antibiotic in 1940s, but later demonstrated that patulin is toxic to plants and animals plants in addition fungi and bacteria. There are several effects of patulin exposure on human such as acute (vomiting, edema, agitation and convulsion), chronic (neurotoxic, immunotoxic, genotoxic), and cellular (inhibition of DNA and protein synthesis). 50 µg mL-1 is fixed a limit value for apple juice in "The Communiqué on Maximum Limits of Contaminants in Foodstuffs" in Turkey. The World Health Organization has been announced that this limit is safe. Consequently, all studies conducted in Türkiye have showed that Patulin is a potential contaminant of apples under conditions in Türkiye. That's why, the authorities and manufacturers should immediately take action to reduce the contamination of Patulin.

Keywords: Patulin, toxicity, apple, Türkiye.

#### **1. INTRODUCTION**

Patulin is a toxic secondary metabolite produced by Aspergillus, Byssochlamys and Penicillium. the most common patulin producing molds growing in apples, peaches, pears, apricots, grapes, cherries and tomatoes is Penicillium expansum and it leads to brown decay (Moake et al., 2005). Apple juice, apple cider and apple past are apple products that can contain this molds.



Figure 1. Structure of patulin (Stott and Bullerman 1975)

4-hydroxy-4H-furo [3,2c] pyran-2[6H]-one (Patulin) is firstly isolated as an antibiotic which is a water-soluble lactone, in the 1940s (Stott and Bullerman, 1975) (Figure 1). On the other hand, various studies revealed that the patulin also toxic to higher plants and animals. These studies showed that symptoms of acute patulin exposure can include vomiting, intestinal inflammation, intestinal hemorrhage, GI tract distension, epithelial cell degeneration, dysponea, agitation, convulsions, pulmonary congestion, ulceration, hyperemia, edema, and other GI and kidney problems (Abu-Bakar et al., 2014).

Neurotoxic, immunosuppressive, immunotoxic, teratogenic, genotoxic and carcinogenic effects are the risks of chronic patulin exposure, in terms of health (Alves, 2000; Schumacher et al., 2006; Selmanoglu, 2006; Wu et al., 2005). The one of these studies has been reported that patulin affects the gastrointestinal tract of the rats feeding with contaminated water (295 mg L-1) for four weeks and and causes ulcers (Speijers et al., 1988). The patulin cell infiltration ranging from 28 to

41 mg kg-1 produced lesions in mice gastrointestinal tissues and these lesions are degeneration, ulceration and bleeding of the gastric mucosa (Mckinley et al., 1982). In another study, it was showed that patulin at micromolar concentrations leads to dramatic and rapid damage in human epithelial intestinal cells (Mahfoud et al., 2002). In addition, patulin can react with terminal sulfhydryl groups of polypeptides and proteins of foods (Fliege and Metzler, 2000).

There are teratogenic effects of patulin on chickens (Ciegler et al., 1977) and immunotoxic, mutagenic, genotoxic and neurotoxic effects in rodents (Ritieni, 2003). Exposure through the consumption of contaminated products with patulin may lead to neurotoxic, hepatotoxic, nephrotoxic, teratogenic, mutagenic, and genotoxic effects in human. Nausea, vomiting and other gastrointestinal trauma associated with kidney injury are several acute effects of patuline (Mahfoud et al., 2002; Ritieni, 2003; Speijers, 2004; Selmanoglu, 2006). The high doses exposure of patulin causes immunosuppressive properties (Moake et al., 2005). Besides, digestion of high patulin may cause serious hazards to human health, in particular for children (Prieta et al., 1994).

#### 2. MATERIALS AND METHODS

The method of the study has been selected as to investigation of researchs conducted in Türkiye and compilation of informations.

#### **3. RESULTS AND DISCUSSION**

#### Studies on Patulin Contamination in Apple Products in Türkiye

In the literature review, it is seen that very few studies have been conducted investigating patulin levels in apple products in our country. 463 apple juice concentrate samples from Turkey was analysed during 1993–94 and 1994–95 in the studies of Karadeniz and Eksi (1995). They was demonstrated that 23.1% of samples has patulin content higher than 25  $\mu$ g L<sup>-1</sup> while 4.5% had higher than 50  $\mu$ g L<sup>-1</sup>. In a study, Patulin was determined in the range of 7 to 376  $\mu$ g L<sup>-1</sup> in all apple juice concentrate samples. The patulin levels of 46% of all analysed samples exceeded 50  $\mu$ g L<sup>-1</sup>(Gökmen and Acar 1998). The patulin contents of apple juice concentrates produced through 1996-99 were analysed by Gökmen and Acar (2000). The patulin content in 48% of apple juice concentrates produced in 1996 was higher than 50 µg L<sup>-1</sup>. The patulin contamination levels was determined 34% of the samples exceeded the limit level with a slight decrease, in 1997. The patulin levels of samples in years of 1998 and 1999 were significantly better and only 8% of samples produced in these years exceeded 50 μg L<sup>-1</sup>. The highest concentrations of patulin were 376, 153, 103 and 119 μg L<sup>-1</sup> in 1996, 1997, 1998 and 1999, respectively, in their study. Patulin was found in range of 19.1 to 732.8  $\mu$ g L<sup>-1</sup> in the 60% of apple juices in another study, and the patulin levels in 44% of the samples were exceeded 50 µg L<sup>-1</sup> (Yurdun, Omurtag, and Ersoy, 2001). Effects of thiamine hydrochloride, pyridoxine hydrochloride and calcium-d-pantothenate on the patulin content of apple juice concentrate were investigated in a study (Yazıcı and Velioğlu 2002). In this study, patulin level of the clear apple juice concentrate which used in assays was 304 µg kg<sup>-1</sup>. The worrying and surprising situation was that this highly contaminated juice concentrate a commercial product obtained directly from a producer in Turkey in 1999. Another study in Turkey showed that patulin levels in 46% of all apple juice concentrates from three different companies were higher than 50  $\mu$ g L<sup>-1</sup> (Akta et al. 2004). Patulin concentrations in all samples were higher than 50  $\mu$ g L<sup>-1</sup> in the study investigated to effect of heat treatment and evaporation on patulin (Kadakal and Nas 2003). The heat treatment was founded more effective in reducing the patulin level according to comparison with the evaporation process. Patulin was detected in all of the fruit juice concentrates analysed in a study in Greece (23 apple juices originated from Turkey; n = 90), and the most contaminated samples were imported from Turkey (Moukas et al. 2008). Karaköse et al. (2015) analysed 6 fruitbased baby foods. In this study, the patulin level determined in all samples was  $\leq 3 \ \mu g \ kg^{-1}$ . Hence, all of the analysed samples may be considered as safe in terms of patulin exposure. Finally, in the study conducted by us, all of the examined apple sour samples was contaminated with Patulin (İçli, 2019). The patulin levels detected in our study was in range of 20 to 1416 mg kg<sup>-1</sup> and mean patulin amount was  $284 \pm 307 \,\mu$ g kg<sup>-1</sup>. The Patulin content in 94.9% of samples was found to be equal to or greater than 50  $\mu$ g kg<sup>-1</sup>.

#### 4. CONCLUSION

As a result, it can be seen that there is a widespread contamination problem from a few studies examining the levels of patulin contamination in apple products in our country. Producers should be warned to use strong apples, and work should be done by authorities to prevent contamination.Regular monitoring of contamination will be beneficial in terms of public health protection.

#### REFERENCES

- Abu-Bakar, N.B., Makahleh, A., Saad, B., 2014. Vortex-assisted liquid–liquid microextraction coupled with high performance liquid chromatography for the determination of furfurals and patulin in fruit juices. Talanta. 120: 47-54.
- Akta, A.H., Yılmazer, M., Demirci, S., 2004. Determination of patulin in apple juice produced in Isparta, Turkey by HPLC with diode array detection. Journal of Food and Drug Analysis 12(3): 228-231
- Alves, I., Oliveira, N. G., Laires, A., Rodrigues, A. S., & Rueff, J., 2000. Induction of micronuclei and chromosomal aberrations by the mycotoxin patulin in mammalian cells: role of ascorbic acid as a modulator of patulin clastogenicity. Mutagenesis 15(3): 229-234.
- Ciegler, A., Vesonder, R.F. and Jackson, L.K., 1977. Production and biological activity of patulin and citrinin from Penicillium expansum. Applied Environmental Microbiology 33: 1004-1006.
- Fliege, R., Metzler, M., 2000. Electrophilic properties of patulin: adduct structures and reaction pathways with 4-bromothiophenol and other model nucleophiles, Chemical Research in Toxicology 13: 363-372.
- Gökmen, V., Acar, J., 1998. Incidence of patulin in apple juice concentrates produced in Turkey. Journal of Chromatography A 815(1): 99-102.
- Gökmen, V., & Acar, J. 2000. Long-term survey of patulin in apple juice concentrates produced in Turkey. Food additives & contaminants 17(11): 933-936.
- İçli, N., 2019. Occurrence of patulin and 5-hydroxymethylfurfural in apple sour, which is a traditional product of Kastamonu, Turkey. Food Additives & Contaminants: Part A DOI: 10.1080/19440049.2019.1605207.
- Karadeniz, F., and Ekşi, A., 1995. Elma suyu konsantrelerinde patulin miktarı ve değişkenliği. Gıda Sanayii Dergisi 39: 14-18.
- Karakose, A., Sanli, S., Sanli, N., & Bulduk, I., 2015. Evaluation of patulin in commercial baby foods by solid phase extraction and liquid chromatography PDA detection. Czech Journal of Food Sciences 33(1): 52-57.
- Kadakal, C., & Nas, S., 2003. Effect of heat treatment and evaporation on patulin and some other properties of apple juice. Journal of the Science of Food and Agriculture 83(9): 987-990.
- Mckinley, E.R., Carlton, W.W., Boon, G.D., 1982. Patulin mycotoxicosis in the rat: toxicology, pathology and clinical pathology. Food and Chemical Toxicology 20(3): 289-300.
- Mahfoud, R., Maresca, M., Garmy, N., & Fantini, J., 2002. The mycotoxin patulin alters the barrier function of the intestinal epithelium: mechanism of action of the toxin and protective effects of glutathione. Toxicology and applied pharmacology 181(3): 209-218.
- Moake, M.M., Padilla-Zakour, O.I., Worobo, R.W., 2005. Comprehensive review of patulin control methods in foods, Comprehensive Reviews in Food Science and Food Safety 4: 8-21.
- Moukas, A., Panagiotopoulou, V., Markaki, P., 2008. Determination of patulin in fruit juices using HPLC-DAD and GC-MSD techniques. Food Chemistry 109(4): 860-867.
- Ritieni, A., 2003. Patulin in Italian commercial apple products. Journal of Agricultural and Food Chemistry 51: 6086-6090.
- Schumacher, D. M., Müller, C., Metzler, M., & Lehmann, L., 2006. DNA–DNA cross-links contribute to the mutagenic potential of the mycotoxin patulin. Toxicology letters 166(3): 268-275.
- Selmanoğlu, G., 2006. Evaluation of the reproductive toxicity of patulin in growing male rats. Food and Chemical Toxicology 44(12): 2019-2024.
- Speijers, G. J. A., Franken, M. A. M., & Van Leeuwen, F. X. R., 1988. Subacute toxicity study of patulin in the rat: effects on the kidney and the gastro-intestinal tract. Food and chemical toxicology 26(1): 23-30.
- Wu, T. S., Yu, F. Y., Su, C. C., Kan, J. C., Chung, C. P., & Liu, B. H., 2005. Activation of ERK mitogen-activated protein kinase in human cells by the mycotoxin patulin. Toxicology and applied pharmacology 207(2): 103-111.
- Yazici, S., & Velioglu, Y. S., 2002. Effect of thiamine hydrochloride, pyridoxine hydrochloride and calcium-d-pantothenate on the patulin content of apple juice concentrate. Food/Nahrung 46(4): 256-257.
- Yurdun, T., Omurtag, G. Z., & Ersoy, Ö., 2001. Incidence of patulin in apple juices marketed in Turkey. Journal of Food Protection 64(11): 1851-1853.





# POSTER PRESENTATION

# **An Overview of Probiotics**

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**Abstract:** Probiotics are live microorganisms that are very beneficial for host health when taken in sufficient quantities. Some of the commonly used probiotic bacteria include *Lactobacillus, Bifidobacteria*, and the yeast *Saccharomyces boulardii*. Generally, a probiotic product should contain more than 10<sup>6</sup>-10<sup>8</sup> cfu of bacteria per gram or millilitre They are used in foods, especially in fermented dairy products, but also in pharmaceutical preparations. Probiotics are widely used to prepare fermented dairy products such as yogurt or freeze-dried cultures. Several health-related effects associated with the probiotics, including alleviation of lactose intolerance, immune enhancement have been reported in human studies. Some evidences suggest a role for probiotics in reducing the risk of rotavirus induced diarrhoea and colon cancer. Also, other areas of application include probiotic effects against *Helicobacter pylori* infections in the stomach, alcoholic liver disease, small bowel bacterial overgrowth, ulcerative colitis, allergy to milk protein, antioxidative effects and asthma. Studies suggest that probiotics may be useful in treatment of patients with hypertension, urogenital infections, lactose intolerance, and elevated levels of cholesterol. Present review focuses on probiotics.

Keywords: Probiotic, prebiotic, health, food



# Effects of Different Nitrite Levels on Volatile Compounds and Physico-Chemical Properties of Pastırma

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**Abstract:** Pastrma is a traditional Turkish dry-cured meat product. In the curing process, the first stage of pastrma production, nitrate is usually used as a curing agent. However, nitrite may be used alone or with nitrate. In this study, in the presence of nitrate, the effects of different nitrite levels (50, 75, 100 and 150 ppm) on the volatile compounds and physico-chemical properties of pastrma were investigated. The production of pastrma was carried out under controlled conditions and as two replications. After the production, samples were subjected to physico-chemical and volatile compound analyses. In the extraction of volatile compounds, the solid phase microextraction technique (SPME) was used and the identification was performed using GC/MS. The different nitrite levels had no significant effect on pH and aw values as well as content of non-protein nitrogenous substance and residual nitrite (P>0,05). However, this treatment had a significant effect on thiobarbituric acid reactive substances (TBARS). The use of 50 ppm nitrite in curing process showed a higher TBARS value than the group with 150 ppm nitrite. L\* and a\* values were not affected by this factor (P>0.05). A total of 38 volatile compounds were identified in pastrma samples. The use of different nitrite levels affected 2-pentyl-furan, octanal, limonene, propyl hexanoat and 2,4-hexadienoic acid, methyl ester (P<0.05) and di-2-propenyl disulphide (P<0.01). In addition, the group with 50 ppm nitrite generally had higher values in terms of many compounds than other groups.

Keywords: Pastırma, nitrite, SPME, GC/MS, TBARS.

## **1. INTRODUCTION**

Pastirma is the most popular dry-cured meat product in Turkey. Processing of pastirma consist of many stages; raw material preparation, curing, washing, first drying, first pressing, second drying, second pressing, covering with çemen and drying with çemen. Pastirma is traditionally processed under natural conditions where air temperature and relative humidity depend on climate and weather conditions. Period covering late September and October-November, which is called "Pastirma Summer", is preferred for traditional production (Gökalp et al., 1994). Pastirma processing generally takes one month. The stability and safety of pastirma is mainly due to low water activity. In addition, paste containing garlic has a protective effect on mould growth and excess drying of the final product (Kaban 2013).

In pastirma production, salt, nitrate and nitrite or nitrate/nitrite are used. Salt gives the typical taste and play a role to decreasing of water activity. Curing agents contribute both the product properties such as color and flavor and the safety of the product (Kaya and Kaban 2016). However, nitrate becomes an active curing agent only after it reduces to nitrite (Honikel 2008; Kaban 2013). On the other hand, it is also known that nitrite plays a significant role in the formation of carcinogenic nitrosamines (Sallan et al., 2019).

Proteolysis and lipolysis are two important reactions during pasturma processing with consequences for the final sensory quality. Significant changes occur in the amount and numbers of volatile compounds during pasturma production (Kaban 2009). There are a few studies on the effect of curing agents on volatile compounds and qualitative properties of pasturma production (Akköse et al. 2017; Hazar et al. 2017). Akköse et al. (2017) investigated the effects of different nitrate levels (150, 300, 450, and 600 mg/kg) on the changes in the composition of volatile compounds, and thiobarbituric acid reactive substances (TBARS), non-protein nitrogenous matter content, color and residual nitrite in pasturma. In another study, the effects of processing conditions (at two different temperatures (4 or 10 C) with two different curing agents (150 mg/kg NaNO2 or 300 mg/kg KNO3) on biogenic amine content and other qualitative properties (pH, a(w), color, residual nitrite, TBARS, NPN-M, microbiological properties) were determined.

Up to now, the effects of different nitrite levels in the presence of nitrate have not been evaluated. The aim of the study was to determine the effects of different nitrite levels (50, 75, 100 and 150 mg/kg) in the presence of nitrate on volatile compounds and some quality properties (150 mg/kg).

#### 2. MATERIALS AND METHODS

In pastirma production, M. Longissmus dorsi muscles were used as raw material. After removing fat and connective tissue from the surface, the muscle was cut across the centre into two pieces. In this way, four strips were prepared for each experiment, and a total of 8 strips were used for two experiments. The production was conducted under the controlled condition in a climate chamber (Reich, Germany). Meat strips were covered with curing mixture (10% NaCl, 0,3% sucrose, 150 mg/kg KNO3 and 50, 75, 100 or 150 mg/kg NaNO2) and kept at  $4\pm1^{\circ}$ C for 48 h. After that, cured strips were dried, pressed and covered with cemen (Tomaç Yazıcı, 2018).

pH was measured in the homogenised samples using a pH meter (ATI Orion 420, USA). Water activity was determined using aw device (TH-500 aw Sprint, Switzerland) at 25 °C. Residual nitrite in samples was detected according to Tauchmann (1987). Color values (L\*, a\* and b\*) were measured using a color meter (CR-400 Konika Minolta, Japan).TBARS (Thiobarbituric acid-reactive substances) value, NPN-M (non-protein nitrogenous matter) content and volatile compounds were determined according to the methods of Lemon (1975), Anonymus (1989) and Kaban (2009), respectively. Pastrma samples were also subjected to the sensorial analysis.

The experiment was a randomised complete block design with two replicates. The obtained data were subjected to analysis of variance (ANOVA). The difference between means were evaluated by Duncan's multiple range test (SPSS 20.0).

#### 3. RESULTS AND DISCUSSION

The different nitrite levels had no significant effect on pH and aw values as well as content of non-protein nitrogenous substance and residual nitrite (P>0,05). The mean pH value of pastirma samples varied between  $5.91\pm0.09$  and  $5.99\pm0.12$ . The pH values in all samples were not under 5.5 and the similar results were determineted by Kaban (2009),Akköse et al.(2017) and Hazar et al., (2017). The average aw value was under 0.90 in all pastirma samples.

The mean value of residual nitrite was found under 10 mg/kg. In another study on pastirma, residual nitrite was also under 10 mg/kg (Hazar et al., 2017). In dry-cured meat products, proteolysis is one of the most important reactions. Amounts of non-protein nitrogenous substances such as peptides and free amino acids increase as a result of proteolysis (Akköse et al., 2017). In the present study, NPN-M content ranged from  $4.61\pm0.23$  g/100g to  $5.21\pm0.81$  g/100g.

This treatment had a significant effect on thiobarbituric acid reactive substances (TBARS). The use of 50 mg/kg nitrite  $(24.76\pm7.85 \ \mu mol MDA/kg)$  in curing process showed a higher TBARS value than the group with 150 ppm nitrite  $(20.57\pm11.47 \ \mu mol MDA/kg)$ . However, the differences were quite low. Akköse et al.(2017) reported that the lowest TBARS value was found in the group containing 150 ppm KNO3 and this value was also statistically different from mean TBARS values of other groups (P<0.05).

 $L^*$  and a\* values were not affected by this factor (P>0.05). There were no significant differences in the sensory properties (color, taste, odor, texture and general acceptibility) of the pasturma groups produced with different nitrite levels (P>0.05).

A total of 38 compounds consisting of 10 different chemical such as aliphatic hydrocarbons, sulfur compounds, aromatic hydrocarbons, aldehydes, terpenes, ketones, esters, furans, acids and alcohols were identified in pasturma samples produced using different nitrite levels (Table 1). The use of different nitrite levels affected 2-pentyl-furan, octanal, limonene, propyl hexanoat and 2,4-hexadienoic acid, methyl ester (P<0.05) and di-2-propenyl disulphide (P<0.01). In addition, the group with 50 ppm nitrite generally had higher values in terms of many compounds than other groups.

#### 4. CONCLUSION

The use of different nitrite level in the presence of nitrate had no significant effect on pH, aw, residual nitrite and NPN-M content. It was also determined that differences between the pastirma groups with different nitrite levels were very low in term of TBARS value, an indicator of lipid oxidation. Color parameters are very important for the evaluation of pastirma quality. The different nitrite levels did not cause any significant changes in L\* and a\* values. Morever, there were also no significant differences in the pastirma groups in terms of sensory parameters. However, the group with 50 ppm nitrite/150 mg/kg nitrate generally had higher values for many compounds than other groups.

#### REFERENCES

- Akköse, A., Ünal, N., Yalınkılıç, B.,Kaban, G.,Kaya, M.,2017. Volatile compounds and some physico-chemical properties of pastırma produced with different nitrate levels. Asians- Australasian Journal of Animal Sciences, 30(8), 1168–1174.
- Anonymous, 1989. Amtliche Sammlung von Untersuchungsverfahren nach 35 LMBG. Untersuchung von Lebensmitteln. Bestimmung des Gehaltes an Nichtprotein- Stickstaffsubstanz in Fleischerzeugnissen.
- Gökalp, H.Y., Kaya, M., Zorba, O., 1994. Et ürünleri işleme mühendisliği. Atatürk Üniversitesi yayın no:786, Ziraat Fakültesi yayın no: 320, Ders kitapları serisi No:70, Atatürk Üniversitesi Ziraat Fakültesi, Erzurum.
- Hazar, F.Y., Kaban, G., Kaya, M., 2017. The effects of different processing conditions on biogenic amine formation and some qualitative properties in pasturma. Journal of Food Science and Technology ,54(12), 3892-3898.
- Honikel, K.O.,2008. The use and control of nitrate and nitrite fort he processing of meat products. Meat Science, 78,68-76.
- Kaban, G., 2009. Changes in the composition of volatile compounds and in mikrobiological and physicochemical parameters during pasturma processing. Meat Science, 82, 17-23.
- Kaban, G., 2013. Sucuk and pastirma : Microbioolgical changes and formation of volatile compounds. Meat science, 95, 912-918.
- Kaya, M., Kaban, G., 2016. Fermente et ürünleri. Gıda Teknolojisi, Editör Necla ARAN, ss. 157-190, Nobel Yayıncılık, İstanbul.
- Lemon, D.W., 1975. An Improved TBA Test for Rancidity New Series Circular, No:51. Halifax-Laboratory, Halifax, Nova Scotia.
- Sallan, S., Kaban, G., Kaya, M., 2019. Nitrosamines in sucuk: Effects of black pepper, sodium ascorbate and cooking level. Food Chemistry, 288, 341-346.
- Tomaç Yazıcı, D., 2018.Pastırmanın fiziko-kimyasal özellikleri ve uçucu bileşikleri üzerine farklı nitrit seviyelerinin etkileri. Yüksek lisans tezi, Atatürk Üniversitesi Fen Bilimleri Enstitüsü,Erzurum.
- Tauchmann, F., 1987. Methoden der chemischen Analytik von Fleisch und Fleischwaren, Bundensanstalt für Fleischforschung, Kulmbach, 80.







# **ORAL PRESENTATION**

# Is Rice Safe?

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**Abstract:** Food engineer's responsibility start from field to fork. Therefore, in food safety point of view rice has problematic issue that is revealed as water contamination and heavy metal migration from soil which are possible reservoirs of heavy metals whom is grow inside the water. It is well known that rice consumption has been related with increased urinary arsenic (As) amount based on numerous studies. Because rice has an ability to accumulate some of the heavy metal like as and cadmium (Cd) which are presents inside the growing area. Rice of Tosya is famous enough to have geographical identification and this study was aimed to checking the heavy metal accumulation in rice plants of Kastamonu-Tosya region. Devrez stream is the major water source to watering rice seeds in this area therefore was selected to be water collection reservoir.

Consequently, water, soil and rice samples were started to be collect from least 6 (six) pre-determined regions in March and October considering before fertilisation and after seeding periods. Heavy metal levels of the samples will be determined on the ICP-OES device. As a preliminary study, water and soil samples were collected from six different region Yukarıkayı, Dağardı, Çepni, Sevinçören, Sapaca and Ortalıca villages respectively in March 2019. Then, these samples were transferred to the laboratory on the same day in cold chain. Water and soil samples were prepared for ICP-OES. 14 elements in water samples measured by ICP-OES. Results of heavy metal analysis showed that all water samples include aluminium (Al) and iron (Fe). The lowest and the highest average value of these elements of Al and Fe were found ranged between 0.858-1.939 mg/L and 0.982-1.342 mg/L, respectively. In addition, water samples were collected from two region contains copper (Cu) including Al and Fe. The measured copper values are 0.013 and 0.236 µg/L. According to initial preliminary results, it is concluded that water samples do not contain toxic heavy metals (cadmium, chromium, nickel, lead, etc.).

Keywords: Rice, Tosya, Kastamonu, water, heavy metal, ICP-OES.

## **1. INTRODUCTION**

Paddy is uncultivated grains of cultured plants in Oryza sativa L. species of Gramineae (wheat) family (Anonim 2010). There are 25 different types of paddy in the world. The most cultivated paddy species are Oryza sativa and Oryza glaberrima sleuth. Homeland of paddy is referred to as Southeast Asia (Anonim 2017). Paddy grows from 90 to 180 days. It grows inside water until harvest (Anonim 2017). Paddy is the only grain that can grow in water (Şapaloğlu 2015). Paddy is the staple food of more than half of the world's population, especially in South Asia and Far East countries. Most rice is consumed around the world after wheat (Anonim 2017).

Tosya district of Kastamonu province is the region where the first paddy cultivation in Turkey. In the 1500s, paddy farming was started in Tosya. Paddy cultivation is done along the valley formed of Devrez stream (Akay et al.,2018). Thus water is the main source of initial contamination and should be analysed before and after seeding period of rice.

Therefore, in this study it was aimed to measuring the heavy metal concentration in water of Kastamonu-Tosya region. Devrez stream is the major water source to watering rice seeds in this area therefore was selected to be water collection reservour. And all data were compared with defined water quality classes as represented in Table 1.

|   |         | WATER ( | QUALITY ( | CLASSES         |
|---|---------|---------|-----------|-----------------|
| WATER QUALITY PARAMETERS                      | Ι       | II      | III       | IV              |
| A) Physical and inorganic-chemical parameters |         |         |           |                 |
| Temperature (°C)                              | 25      | 25      | 30        | >30             |
| pH  | 6.5-8.5 | 6.5-8.5 | 6.0-9.0   | 6.0-9.0 outside |
| Dissolved O <sub>2</sub> (mg/L)               | 8       | 6       | 3         | <3              |
| B) Inorganic Contamination Parameters         |         |         |           |                 |
| Al (mg Al/L)                                  | 0.3     | 0.3     | 1         | >1              |
| As (µg As/L)                                  | 20      | 50      | 100       | >100            |
| Cd (µg Cd/L)                                  | 3       | 5       | 10        | >10             |
| Cu (µg Cu/L)                                  | 20      | 50      | 200       | >200            |
| Co (µg Co/L)                                  | 10      | 20      | 200       | >200            |
| Cr (total) (µg Cr/L)                          | 20      | 50      | 200       | >200            |
| Fe (µg Fe/L)                                  | 300     | 1000    | 5000      | >5000           |
| Pb (µg Pb/L)                                  | 10      | 20      | 50        | >50             |
| Mn (µg Mn/L)                                  | 100     | 500     | 3000      | >3000           |
| Ni (µg Ni/L)                                  | 20      | 50      | 200       | >200            |
| Zn (µg Zn/L)                                  | 200     | 500     | 2000      | >2000           |

Table 1. Quality criteria according to the classes of water resources (Anonim 2004).

#### 2. MATERIALS AND METHODS

Water samples were collected from six different region Yukarıkayı, Dağardı, Çepni, Sevinçören, Sapaca and Ortalıca villages respectively in March 2019. Some physical and chemical properties were measured when collecting water samples (Table 2). 3 ml of nitric acid (HNO3) was added to water samples. These samples were transferred to the laboratory on the same day in cold chain. Then, water samples were filtered and analysed on the Inductively Coupled Plasma Emission Spectrometry (ICP-OES) device.

## 3. RESULTS AND DISCUSSION

#### Results

Water samples physical and chemical properties of dissolved  $O_2$  amount, pH values, temperatures, salinity percentages and conductivity were recorded while sampling as seen in Table 2. Additionally, 14 elements concentration in collected water samples were measured by ICP-OES and the results are given in Table 3. Results of heavy metal analysis show that all water samples include Al and Fe. The lowest and the highest average value of these elements are found to be ranged between 0.858-1.939 mg/L and 0.982-1.342 mg/L, respectively. In addition, water samples were collected from two region contains copper including Al and Fe. The measured copper values are 0.013 and 0.236  $\mu$ g / L. According to initial analysis, it is concluded that water samples do not contain toxic heavy metals (cadmium, chromium, nickel, lead, etc.).

|          | Dissolved O <sub>2</sub><br>(mg/L) | pН   | Temperature<br>(°C) | Salinity<br>(%) | Conductivity<br>(µs/cm) |
|----------|------------------------------------|------|---------------------|-----------------|-------------------------|
| Sample 1 | 11.43                              | 9.6  | 7.8                 | 11              | 150.1                   |
| Sample 2 | 11.49                              | 8.95 | 8                   | 10              | 149.3                   |
| Sample 3 | 11.41                              | 8.58 | 8                   | 12              | 169.7                   |
| Sample 4 | 11.21                              | 8.71 | 8.7                 | 12              | 178.7                   |
| Sample 5 | 11.35                              | 8.58 | 8.8                 | 14              | 198.7                   |
| Sample 6 | 11.32                              | 8.83 | 9.2                 | 14              | 239.4                   |

Table 2. Some physical and chemical properties of water samples

Table 3. ICP-OES results of water samples

|       |      | Limit    |             |             |             |             |             |             |
|-------|------|----------|-------------|-------------|-------------|-------------|-------------|-------------|
| Metal | Unit | Value*   | Station 1   | Station 2   | Station 3   | Station 4   | Station 5   | Station 6   |
| Al    | mg/L | 5        | 0.898       | 1.939       | 0.929       | 0.994       | 0.858       | 1.005       |
| As    | μg/L | 160      | ND          | ND          | ND          | ND          | ND          | ND          |
| Cd    | μg/L | 8.2      | ND<br>0.236 | ND          | ND          | ND<br>0.013 | ND          | ND          |
| Cu    | μg/L | 200-1000 |             | ND          | ND          |             | ND          | ND          |
| Со    | μg/L | 50       | ND          | ND          | ND          | ND          | ND          | ND          |
| Cr    | μg/L | 8        | ND<br>0.983 | ND<br>0.999 | ND<br>1.119 | ND<br>1.148 | ND<br>1.342 | ND<br>1.227 |
| Fe    | mg/L | 5        |             |             |             |             |             |             |
| Pb    | μg/L | 200      | ND          | ND          | ND          | ND          | ND          | ND          |
| Mn    | μg/L | 200      | ND          | ND          | ND          | ND          | ND          | ND          |
| Мо    | μg/L | 10       | ND          | ND          | ND          | ND          | ND          | ND          |
| Ni    | μg/L | 200      | ND          | ND          | ND          | ND          | ND          | ND          |
| Zn    | mg/L | 1 or 5   | ND          | ND          | ND          | ND          | ND          | ND          |

**ND: Non Detected** 

\*(Anonim 2018)

#### Discussion

Sample 1;  $1^{st}$  quality according to temperature value,  $4^{th}$  quality according to pH value,  $1^{st}$  quality according to dissolved O<sub>2</sub> value according to the Water Pollution Control Regulation.

Sample 2;  $1^{st}$  quality according to temperature value,  $3^{rd}$  quality according to pH value,  $1^{st}$  quality according to dissolved O<sub>2</sub> value according to the Water Pollution Control Regulation.

Sample 3;  $1^{st}$  quality according to temperature value,  $3^{rd}$  quality according to pH value,  $1^{st}$  quality according to dissolved O<sub>2</sub> value according to the Water Pollution Control Regulation.

Sample 4;  $1^{st}$  quality according to temperature value,  $3^{rd}$  quality according to pH value,  $1^{st}$  quality according to dissolved O<sub>2</sub> value according to the Water Pollution Control Regulation.

Sample 5;  $1^{st}$  quality according to temperature value,  $3^{rd}$  quality according to pH value,  $1^{st}$  quality according to dissolved  $O_2$  value according to the Water Pollution Control Regulation.

Sample 6;  $1^{st}$  quality according to temperature value,  $3^{rd}$  quality according to pH value,  $1^{st}$  quality according to dissolved  $O_2$  value according to the Water Pollution Control Regulation. Water quality according to Al values of samples 2 and sample 6 is  $4^{th}$  class. Other samples according to Al values have  $3^{rd}$  grade quality. Copper element was found in two samples (Station1,4).  $1^{st}$  class quality in two samples according to copper content. Water quality according to Fe values of samples 2 is  $2^{nd}$  class. Other samples according to Fe values have  $3^{rd}$  grade quality.

#### 4. CONCLUSION

According to the initial results of the analysis, water samples do not contain risk. All water samples have metal content below the threshold limit values of water quality standards belongs to surface water. The lowest and the highest average value of these elements of Al and Fe were found ranged between 0.858-1.939 mg/L and 0.982-1.342 mg/L, respectively. In addition, water samples were collected from two region contains copper including Al and Fe. Water quality of selected location are between 1st and 3rd quality standards based on temperature and pH values respectively. Some metals were below the detection limit of the ICP-OES device. Our research will be continued with soil and rice samples analysis.

#### Acknowledgements

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#### REFERENCES

- Akay H., Sezer, İ., & Mut Z., (2018). Coğrafi İşaretler ve Tosya Yerel Çeltik Genotipleri Örneği. Adnan Menderes Üniversitesi Ziraat Fakültesi Dergisi, 15(2), 81-85.
- Anonim, 2004. Su Kirliliği Kontrolü Yönetmeliği, (SKKY), Türkiye Cumhuriyeti Resmi Gazete No:25687.
- Anonim, 2010. Türk Gıda Kodeksi Pirinç Tebliği, Türkiye Cumhuriyeti Resmi Gazete No:27808.
- Anonim, 2017. 2017 Hububat Raporu, Toprak Mahsulleri Ofisi Genel Müdürlüğü.
- Anonim, 2018. Environmental quality guidelines for Alberta surface waters.
- Şapaloğlu, A. (2015). Pirinç üretim-tüketim zincirinde pazarlama kanallarının yapısı ve pirinç pazarlama marjları: Edirne ili örneği (Thesis of Master, Namık Kemal Üniversitesi).



# Determination of Food Safety Risks in the Heat Treatment Stage of Emulsified Meat Products Production

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Abstract: The need for preservation techniques has been needed as meat is a basic food and can be deteriorated quickly. The process of emulsifying is a method of preservation, whereby water and oil are held together in a homogeneous manner by means of meat proteins and emulsifiers. In this study, a food safety risk assessment has been established for the heat treatment stage in a facility where emulsified meat products are produced. Heat treatment, the general name of the processes such as cooking, pasteurization, sterilization, is a cooking process between 68-87 °C for 90-180 min depending on the size of the product. For this purpose, five different risk assessment methodologies were used as preliminary risk analysis using checklists, failure mode and critically effects analysis, event tree analysis, hazard and operability study, and the risk assessment matrix. As a result of the study, heat treatment which is one of the most important process steps of a meat product facilities, it was determined that the biological hazard is a high risk because of the microbial activity of the most sensitive point. The source and limit of the hazards are analysed and the process steps before and after the heat treatment process are evaluated and the control measure to be taken against the biological hazard is determined as the critical control point (CCP). The food safety team leader, in each baking process, should carry out all monitoring and controls by checking the product's midpoint temperature and the oven temperature. Regular maintenance of the furnace and emergency repair at the time of failure are the most important preventive action. Corrective action is the re-cooking of the undercooked product or the retrieval and destruction if it is not possible to cook again. No physical, chemical or allergic hazards detected. In conclusion, the biohazard is evaluated with different risk assessment methodologies and integrated with food safety management systems. The methodologies used in this study are recommended to be applied at all stages of the production of emulsified meat products.

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Keywords: Risk analysis, critical control point, assessment matrix, event analysis, decision tree



# Mouldy Civil Cheese: Manufacture, Chemical Properties and Recently Studies

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Abstract: Civil is a traditional cheese produced in eastern of Turkey, especially Erzurum province. It is produced by using skimmed milk and the addition of whey in its manufacture. The cheese is produced by both rennet and acid coagulation and also a combination of heat treatment of milk. It is characterized by a fibrous structure which is formed by continuous stirring of milk during the coagulation period; and it has a low fat content and a plastic texture because of kneading during manufacture. Due to its low fat level, its popularity has increased recently among people with high cholesterol or similar health problems. During ripening, mould may develop on the surface spontaneously. Civil cheese has a Protected Geographical Indication status, and its characteristics are specified in the documents published by Turkish Patent and Trademark Office (TÜRKPATENT 2009). The resultant cheese is evaluated as a different variety and named 'mouldy Civil' cheese. In this situation, mouldy Civil cheese is completely different from regular Civil cheese; extent microbiological and biochemical changes occur during ripening. The ripened cheese has a blue greenish colour, and a stringy structure. Although its popularity has increased recently due to its nutritive value and distinctive flavor; however, the mould growth on the cheese surface may be a potential risk for public health. So, a few comprehensive researches have been carried out to overcome this potential problem. Recently, the popularity of the cheese increased in Turkey and researches which on mould starter culture have increased. In this presentation, the recent works on Moldy Civil Cheese (proteolysis, volatile compounds, non-toxin producing strains of *Penicillium roqueforti*, other changes during ripening) were discussed in detail.

Keywords: Cheeses of Turkey, Civil cheese, Mouldy cheese, cheese properties

## **1. INTRODUCTION**

Many cheese varieties that are produced only in restricted geographic areas of the world are consumed locally in large quantities. More than 100 varieties of cheese are found in Turkey; however, three of them (Beyaz, Kasar, and Tulum cheeses) are the most popular (Çakmakçı, 2011). Civil cheese is greatly consumed cheese variety in Turkey and generally produced in the eastern part of Turkey, especially in Erzurum province. The cheese is manufactured using skim milk, so contains low level of milk fat. Recently, consumers are keeping away from milk fat due to cholesterol and high energy value (Cambaztepe et al. 2009). Consequently, consumers prefer to take low-or reduced-fat foods (especially dairy products) and so, their popularity has increased. It is generally ripened under brine or after dry-salted. Also, a mixture of Civil and Lor cheese (whey cheese) is pressed into goat skins or plastic bags and ripened for 3 months or longer. The dry-salted type is spontaneously mildewed during ripening. The mould-ripened cheese is preferred by some consumers and is called "Göğermiş cheese" in Turkish in local producers and consumers.

## 2. DEFINITION AND MANUFACTURE

This mould-ripened Civil cheese is very popular in the eastern region of Turkey. The ripened cheese has a blue greenish colour, and a stringy structure (Cakmakci et al. 2012; 2013). There is also no study on packaging of Civil cheese. Civil cheese is ripened in different methods (brine salting, dry salting, incorporating with Lor-Turkish whey cheese and vacuum packaging). Civil cheese is generally kept in plastic bags. Civil cheese has a Protected Geographical Indication status, and its characteristics are specified in the documents published by Turkish Patent and Trademark Office (TÜRKPATENT 2009). Due to its low fat level, its popularity has increased recently among people with high cholesterol or similar health problems (Cambaztepe et al. 2009). The resultant cheese is evaluated as a different variety and named "mould-ripened Civil" cheese. In this situation, mould-ripened Civil cheese is completely different from regular Civil cheese; many microbiological and biochemical changes occur during ripening.

The production flow chart of Civil cheese is given in Figure 1 and also some photos of cheese production are also shown in Figures 2-10.

Raw cows' milk J Cream separation Gradual heating to 45-50 °C Addition of acidified whey by indigenous microorganisms (ca. 0.5%, as lactic acid) Addition of rennet at 40-42 °C with gentle stirring Gradual heating to 55-60 °C with stirring Transfer the curd onto a table and kneading Ł Hang the curd on a platform for stretching and to form a fibrous texture Rest for overnight and pressing for about 2 days The following day, the pressed curd was shredded like wire or string shapes Shredded curd was divided into parts (normal or mould inoculation) Add coarse salt ↓ Packaging Ł Ripening (at 4-10 °C)

Figure 1. A flow sheet for the production of Civil cheese (Cakmakci et al. 2014)

The cheese is manufactured using skim milk without starter cultures in small scale dairies and for home production by families. However, due to its importance in the field of nutrition and its growing popularity, it is now being produced in large milk factories. A major part of the skim milk, especially from butter production, in dairy plants is processed into Civil cheese to gain a substantial additional economic profit (Cakmakci, 2011). Civil cheese is either consumed as fresh or generally kept in brine for storage. Sometimes, it may be matured together with Lor cheese. Lor cheese is created with the whey released during the production of white pickled and Kasar cheeses (Cambaztepe et al., 2009). The excessive whey is boiled, and the resulting coagulated matter is broken up into tiny pieces. Lor is an unsalted and cheaper than other types of cheese due to by-product in dairy factories. Also, it has high nutritional value due to serum protein. Due to its low fat level, its popularity has increased recently among people with high cholesterol or similar health problems (Cambaztepe et al. 2009). The resultant cheese is evaluated as a different variety and named 'mould-ripened Civil' cheese. In this situation, mould-ripened Civil cheese is completely different from regular Civil cheese; many microbiological and biochemical changes occur during ripening.



2

3



7



6

5



Figures 2-10. Photos of Civil cheese production

As given in the Figures, the ripened cheese has a blue greenish colour, and a stringy structure. Because of its low-fat and high-protein content, its popularity has increased recently in terms of its nutritive value and distinctive flavour. Recently, the popularity of the cheese increased in Turkey and researches which on mould starter culture have increased. In this presentation, the recent works on Mold-ripened Civil Cheese (proteolysis, volatile compounds, *P. roqueforti* strains that do not produce toxins, other changes during ripening) were given in detail.

## 3. RECENT STUDIES ON MOLD-RIPENED CIVIL CHEES

In a research conducted by our research group, four different types of mould-ripened Civil cheese were manufactured. A defined (non-toxigenic) strain of a *P. roqueforti* was used as the secondary starter with or without addition of the whey cheese (Lor); in parallel, secondary starter-free counterparts were manufactured. Chemical composition, microbiology and proteolysis were studied during the ripening. The incorporation of whey cheese in the manufacture of mould-ripened Civil cheese changed the gross composition and adversely affected proteolysis in the cheeses. The inoculated *P. roqueforti* moulds appeared on the surface and grown slowly. A little proteolysis was evident in all cheeses during the first 90 days of ripening. However, sharp increases in the soluble nitrogen fractions were observed in all cheeses after 90 days. In the other our study, we determined that use of Lor in production caused a slow proteolysis during ripening by

filling the holes among fibrous structure and blocked the growth of P. roqueforti. As a result, it was recommended that only Civil cheese should be put into the cheese package, tulum (goat-skin bag) or plastic material (Cakmakci et al. 2014). Microbiological analysis showed that the microbial counts in the cheeses were at high levels at the beginning of ripening, while their counts decreased approximately 1-2 log cfu/g towards the end of ripening (Cakmakci et al. 2014). A total of 83 compounds were identified in moluld ripened Civil cheeses. Ketones, alcohols and esters were the principal classes of volatile components. Principal component analysis of the headspace volatiles grouped cheeses by age and type and P. roqueforti inoculated cheese was clearly separated from the other cheeses at 180 days of ripening, and these cheeses were characterized with high levels of ketones (e.g., 2-butanone, 2-heptanone). Differences in the panel scores between the cheese samples were not significant during the first stage of ripening (up to 60 days); as ripening proceeded, these differences were become evident and P. roqueforti inoculated cheeses received higher scores than others. Addition of Lor in the manufacture of mould-ripened Civil cheese caused lower points by the sensory panel, and the cheese inoculated with P. roqueforti and Lor-free was the best type of mould-ripened Civil cheese. The results showed that the use of P. roqueforti in the manufacture of mould-ripened Civil cheese has significant impact on the volatile profiles and sensory attributes (Cakmakci et al. 2013). In relation to the same research; a total of 186 mold strains were isolated from 41 samples of mould-ripened Civil cheese, and 165 of these strains were identified as P. roqueforti. Identification and mycotoxicologic analyses were conducted using morphotypic and molecular methods. Of 165 isolates, only 28 isolates do not produce penicillic acid, P. roqueforti toxin, or roquefortine (Cakmakci et al. 2012). In relation to the same research; all strains were capable of mycotoxin production which toxins were analysed by HPLC method. It was established that all the strains (albeit at very low levels) produced roquefortine C, penicillic acid, mycophenolic acid and patulin. The amounts of toxins were in the ranges of 0.4-47.0, 0.2-43.6, 0.1-23.1 and 0.1-2.3 mg/kg, respectively. Patulin levels of the samples were lower than the others. The lowest level and highest total mycotoxin levels were determined as 1.2 and 70.1 mg/kg, respectively. The results of this preliminary study may help in the choice of secondary cultures for mould-ripened civil cheese and similar varieties of cheeses (Cakmakci et al. 2015).

#### 4. CONCLUSION

The popularity of mould-ripened Civil cheese increased recently and it is possible to find it easier when compared to last decade. It can be thought that having the geographical indication may be positive contribution its popularity and/or recognition in Turkey. Mould-ripened Civil cheese was well-characterised by supporting TÜBİTAK project within the last ten years; however, it needs a non-toxigenic *P. roqueforti* strain as a seconder starter for industrial production. In this cheese with high export potential, the last step of the commercial starter development should be completed and appropriate packaging should be developed.

#### REFERENCES

- Çakmakçı, S., 2011. Türkiye Peynirleri. In: A A Hayaloğlu & B Özer (Eds.), Peynir Biliminin Temelleri. ISBN: 978-605-87976-1-1, SİDAS Medya Ltd. Şti., İzmir, 585-614.
- Cakmakci, S., Gundogdu, E., Hayaloglu, A. A, Dagdemir, E., Gurses, M., Cetin, B., and Tahmas-Kahyaoglu, D., 2012. Chemical and microbiological status and volatile profiles of mouldy Civil cheese, a Turkish mould-ripened variety. International Journal Food Science and Technology, 47: 2405-2412.
- Cakmakci, S., Hayaloglu. A. A., Dagdemir, E., Gurses, M., Cetin, B., and Tahmas-Kahyaoglu, D., 2013. Effect of *Penicillium roqueforti* and incorporation of whey cheese on volatile profiles and sensory characteristics of mould-ripened Civil cheese. International Journal of Dairy Technology, 66: 512-526.
- Cakmakci, S., Hayaloglu, A.A., Dagdemir, E., Cetin, B., Gurses, M., and Tahmas-Kahyaoglu, D., 2014. Effect of *Penicillium roqueforti* and whey cheese on gross composition, microbiology and proteolysis of mould-ripened civil cheese during ripening. International Journal of Dairy Technology, 67: 594-603.
- Cakmakci, S., Gürses, M., Hayaloglu, A.A., Cetin, B., Sekerci, P. and Dagdemir, E., 2015. Mycotoxin production capability of *Penicillium roqueforti* in strains isolated from mould-ripened traditional Turkish civil cheese. Food Additives & Contaminants: Part A, 32: 245-249.
- Cambaztepe, F., Cakmakci, S. and Dagdemir, E., 2009. Effect of some technological parameters on microbiological, chemical and sensory qualities of Civil cheese during ripening. International Journal of Dairy Technology, 62: 541-548.
- TÜRKPATENT, 2009. Civil Peynir Coğrafi İşaret Tescil Belgesi. Turkish Patent and Trademark Office, Ankara, Turkey.



# Comprasion of Phosphorus Content of Çankırı Rock Salt with Phosphorus Content of the Others Salt Consumed in Turkey

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**Abstract:** Phosphorus element is the most plentiful element in human body after calcium element. It works with calcium and other nutrients. The main function of phosphorus is in the formation of bones and teeth in the body. It plays an important role in how the body uses carbohydrates and fats and also needed for the body to make protein for the growth, maintenance, and repair of cells and tissues.

A simple chemical compound, sodium chloride, also known as food salt, is a foodstuff that has been of great importance to humans for centuries. Rock salt is one if the major salt sources. Çankırı rock salt region formed in Tertiary period is one of important mine regions of Turkey. In this study, the phosphorus contents of forty-five Çankırı rock salt samples gathered from the salt caves are compared with the phosphorus contents of the lake salts and sea salts consumed in Turkey. Phosphorus contents were measured using energy-dispersed X-ray fluorescence (EDXRF) spectroscopy. The average phosphorus content of Çankırı rock salt, lake salt and sea salt were found as 114 mg/kg, 71 mg/kg and 97 mg/kg, respectively. The average phosphorus content of Çankırı rock salt is 60% higher than the phosphorus content of the lake salt and 17% higher than phosphorus content of the sea salt. Result reveals that Çankırı rock salt is more rich phosphorus than lake and sea salts.

Keywords: Phosphorus, Rock Salt, Sea salt, Lake salt, EDXRF.

## **1. INTRODUCTION**

Salt is a nutrient required for the human body. In our body protein, fat, carbohydrates and water, along with Calcium, Phosphorus, Sodium, Chloride, Potassium, Sulfur and Magnesium, such as ions are found. Sodium, Chloride, Potassium and Calcium ions provide the electrolyte balance of our body. If any of these elements is more or less in the human body, the electrolyte will disrupt the balance and cause some diseases in the body. It is possible to find Sodium and Chloride at the same time in these elements. Approximately 40% of the pure salt contains Sodium and 60% Chloride. If for any reason these two elements decrease in the body, the majority of our body's needs can be obtained from salt. [6]

Phosphorus is especially important for the nervous system and brain which are phosphorus in their structure. Calcium works with phosphorus in the organism as well as with magnesium. The bones of the newborn baby are in cartilage, bendable. The calcium stored in the bones gives them hardness with phosphorus. This event is carried out with the catalysis of vitamin D. Calcium and phosphorus, bone, as well as the formation of teeth, hardening and gain their properties and take their normal shape together. Phosphorus helps the body as a major protein material in all tissues and pores. The phosphorus that works so important prevents the normal work of calcium when taken more than necessary, thus causing calcium deficiency. [10]

The amount of phosphorus you need in your diet depends on age. Adults require less phosphorous than children aged 9-18 years and more phosphorus than children under 8 years of age. The average amount of phosphorus needed by the human body can be sorted as follows. Adults (19 years and older) 700 mg, Children (9-18 years) 1,250 mg, Children (4 to 8 years) 500 mg, Children (1 to 3 years) 460 mg, Infants (7 to 12 months) 275 mg, Babies (0 to 6 months) are 100 mg. [10]

Rarely, some people need nutritional supplements for phosphorus. However, the required amount of phosphorus is taken by mostly consumed foods. Phosphorus height may create problems in terms of our health. Too much phosphorus can be toxic. Mineral excess may cause diarrhea and organs to harden and soft tissue formation. High phosphorus levels can have a negative impact on your body's effective use of other minerals such as iron, calcium, magnesium and zinc. Phosphorus may begin to accumulate in muscles by joining with calcium. Too much phosphorus in the blood is rare. Typically, this problem may occur in patients who have a kidney problem or cannot adjust the calcium level due to discomfort.

Normally, phosphorus degradation is not a very common problem. However, some drugs used may cause phosphorus degradation. For example, insulin, ACE inhibitors, corticosteroids, antacids, anticonvulsants. Symptoms of phosphorus abortion include:

- Joint or bone pain
- Loss of appetite
- Irritability or anxiety
- Fatigue
- Poor bone development in children

In these cases, a specialist doctor should be consulted.

#### 2. MATERIALS AND METHODS

Rock Salt Samples were collected from 3 different salt mine quarries located in Yenidoğan, Ballıbağı Village and Doğantepe Locations within the boundaries of Çankırı province. A total of 45 rock salt samples, each of which were 15 samples from different points of each salt mine, were kept in plastic bags and brought to the laboratory. In addition, 15 different salt samples (Sea salt, Lake salt, Himalayan salt top) were collected in supermarkets. It was then allowed to dry.

Rock salt samples were powdered by physical means in order to have the same geometry as the reference material used for the calibration of the detector in gamma-ray spectrometric measurements. Each sample was coded to include information about the locations and points where they were collected. For gamma-ray spectrometric analyzes, each sample of rock salt was transferred to polyethylene specimens of 5x6 cm size with the same size as the calibration sources and weighed by net weighing (Table 1.). were incubated for 45 days to provide a permanent balance between radium (226Ra) and radon (222Rn).

| Kod         | Net<br>Ağırlık | Kod         | Net<br>Ağırlık | Kod | Net<br>Ağırlık |
|-------------|----------------|-------------|----------------|-----|----------------|
| <b>S</b> 1  | 128,06         | S21         | 131,14         | S41 | 137,61         |
| S2          | 135,05         | S22         | 136,09         | S42 | 125,24         |
| <b>S</b> 3  | 133,06         | S23         | 129,17         | S43 | 125,42         |
| <b>S</b> 4  | 137,06         | S24         | 135,53         | S44 | 123,64         |
| S5          | 143,78         | S25         | 144,03         | S45 | 125,94         |
| <b>S</b> 6  | 121,08         | S26         | 130,69         | M1  | 145,75         |
| <b>S</b> 7  | 142,00         | S27         | 132,68         | M2  | 135,46         |
| <b>S</b> 8  | 132,18         | S28         | 135,48         | M3  | 145,71         |
| <b>S</b> 9  | 130,30         | S29         | 144,37         | M4  | 137,02         |
| <b>S</b> 10 | 127,32         | <b>S</b> 30 | 134,20         | M5  | 141,81         |

| S11 | 136,44 | S31 | 128,92 | M6  | 146,29 |
|-----|--------|-----|--------|-----|--------|
| S12 | 123,16 | S32 | 129,99 | M7  | 154,65 |
| S13 | 130,22 | S33 | 142,73 | M8  | 127,92 |
| S14 | 134,19 | S34 | 132,96 | M9  | 159,98 |
| S15 | 138,17 | S35 | 124,98 | M10 | 131,75 |
| S16 | 141,60 | S36 | 136,46 | M11 | 134,91 |
| S17 | 130,57 | S37 | 125,72 | M12 | 109,75 |
| S18 | 139,47 | S38 | 128,74 | M13 | 119,7  |
| S19 | 139,67 | S39 | 140,31 | M14 | 135,98 |
| S20 | 134,23 | S40 | 144,32 | M15 | 147,78 |

The activity concentrations (levels) of radionuclides (226Ra, 232Th and 40K), which are naturally contained in rock salt samples, were determined using an HPGe detector gamma-ray and (2) main element and / or heavy metal levels by X-ray spectrometric method.

#### 3. RESULTS AND DISCUSSION

As a result of measurements, phosphorus ratios and average value in rock salt samples are given in the graph below (graph 1).



Graphic 1. Phosphorus values in collected rock salt samples

In the chart below, the average phosphorus value of the rock salts and the phosphorus value of sea salt and lake salt samples collected from the markets are given.



Graphic 2. Rock Salt-Lake Salt-Sea Salt Phosphorous Content

The average phosphorus content of Çankırı rock salt, lake salt and sea salt was found to be 114 mg / kg, 71 mg / kg and 97 mg / kg, respectively.

The average phosphorus content of the Çankırı rock salt is 60% higher than the phosphorus content of the lake salt and 17% higher than the phosphorus content of the sea salt. The result shows that Çankırı rock salt has a rich phosphorus content than lake and sea salts.

#### REFERENCES

- [1]Peach, C. J., Spiers, C. J., Tankink, A. J., & Zwart, H. J. (1987). Fluid and ionic transport properties of deformed salt rock (No. EUR--10926). Commission of the European Communities.
- [2]Özşen, H. (2009). Kaya tuzuna ait kısa ve uzun dönemli mekanik özelliklerin belirlenmesi ve matematiksel modellenmesi (Doctoral dissertation, Selçuk Üniversitesi Fen Bilimleri Enstitüsü).
- [3]ICRP (International Commission on Radiological Protection). (1996). Age-depended doses to members of the public from intake of radionuclides. Vol. 72, Part 5: compilations of ingestion and inhalation dose coefficient. Kanada.
- [4]TAEK,2002.Türkiye Atom Enerjisi Kurumu ve Biz.Ankara.
- [5]URL-1, http://physics.isu.edu/radinf/natural.html, 19 Temmuz 2012.
- [6]URL-2, http://www.billurtuz.com.tr/tuz\_nedir.php,2016.
- [7]URL-3, http://www.mta.gov.tr/v3.0/bilgi-merkezi/kaya-tuzu,2018
- [8]URL-4,www.taek.gov.tr/bilgi-koşesi/radyasyon-insan-ve-cevre/81-radyasyonla-birlikte- yasiyoruz/ 233-yapay-radyasyon-kaynaklari.html, 5 Temmuz 2012.
- [9]URL-5, www.nuclearnewcomer.com, Erişim Tarihi: 28 Aralık 2008.
- [10]URL-6,https://www.bilimvesaglik.com/mineraller/fosfor-ve-yararlari.html,2009
- [11]UNSCEAR 2000, United Nations Scientific Committee on the Effects of Atomic Radiation Annex B Exposure from natural radiation sources .



# Potential Use of Pumpkin Seed (*Cucurbita pepo* L.) Flour as Fat Replacer and Functional Ingredient in the Meatball Production

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**Abstract:** Meatball is one of the most important foods of Turkish cuisine, and in Turkey, about 290 types of meatballs are produced. In this study, pumpkin seed flour was used as a fat replacer and functional ingredient in the production of meatball. Five different meatball formulations were prepared where fat was replaced with pumpkin seed flour as 0% (control), 3, 6, 9 and 12. Meatballs were analyzed for moisture, protein, fat, ash, cooking loss, water holding capacity (WHC), pH, color parameters, texture profile analysis, fatty acid profiles and sensory properties. Utilization of pumpkin seed flour increased dry matter, protein, ash and pH, and decreased fat content of meatballs. The addition of pumpkin seed flour improved the cooking loss, but did not affect WHC of meatballs. Incorporation of pumpkin seed flour increased  $L^*$  (lightness),  $b^*$  (yellowness) and cohesiveness values, and decreased  $a^*$  (redness) and hardness values, but  $a^*$ , hardness and cohesiveness values of meatballs containing 3% pumpkin seed flour were close to control without pumpkin seed flour. Linoleic acid (C18:2n-6c) and polyunsaturated fatty acids (PUFA) content of meatballs increased as the level of pumpkin seed flour increased. The PUFA/saturated fatty acids (SFA) ratio increased from 0.05 in the control to 0.26 in meatballs with 12.0% pumpkin seed flour. Also, the n-6/n-3 ratio increased from 3.41 in the control to 17.78 in the meatballs with 12.0% pumpkin seed flour. The results indicate that the nutritional status of meatballs was enhanced, while their fat content decreased with minimal composition and sensory changes with 3% pumpkin seed flour addition.

Keywords: Meatball, pumpkin seed, fat replacer, functional ingredient, nutritional quality



# Potential Use of Natural Additives in Functional Modification and Preservation of Meat Products: A Review

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Abstract: Meat and meat products are a major source of food proteins with high biological value, and some valuable nutrients such as minerals and vitamins. Some of these nutrients (e.g., iron, vitamin B12 and folic acid) are either not present or have inferior bioavailability in other foods. Meat and meat products also contain elements which in certain circumstances and in inappropriate proportions have a negative effect on human health. This unfortunate situation derives mainly from the content of fat, saturated fatty acids, and cholesterol and their association with cardiovascular diseases, some types of cancer and obesity. A major cause of meat products deterioration is lipid oxidation, as they contain a high level of lipid and metals such as iron. In addition, sodium chloride used in meat products production is known to be a prooxidant. The oxidation process greatly reduces the nutritional value of lipids and leads to the development of undesirable rancidity and potentially toxic reaction products. For these reasons, it is very important to retard the oxidation process and achieve healthier meat products for the food industry. Meat product manufacturers in the past few decades have used several synthetic food additives with antioxidant, antimicrobial and functional properties to improve the shelf life and nutritional quality of meat products. The use of synthetic food additives, however, has begun to be restricted because of their toxicity and carcinogenicity. There is, therefore, a growing interest in the identification of new, natural ingredients that would serve as alternatives to the synthetic compounds. Currently, a series of ingredients such as vegetable oils, fish oils and natural extracts with antioxidant properties, vegetable products and fiber are added to meat products to improve the shelf life and functional properties.

Keywords: Meat product, natural additive, preservation, functional modification, shelf life



# The Effect of Using Vegetable Fibers on Phase Separation of Tahini

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Abstract: Tahini, condensed with a colloidal suspension, also has a high protein and fat content. A large part of the structure of tahini is composed of 55-60% fat, 23-27% part of the protein, 6.4-20% of the carbohydrate and 1-3% of the water. A phase separation takes place based on the separation of oil from other components during storage of tahini. Although this event a natural phenomenon encountered in the storage of tahini, it leads to be perceived as corrupt or poor quality products by consumers and it creates a negative image of the product. Also in the advanced stages of sedimentation solidifies the bottom of the packaging and the use of tahini is becoming a difficult task. In addition, oil separation on the tahini is also a problem when using tahini in other products. The purpose of this study is to prevent the oil droplets from joining (coalescence) easily by increasing the viscosity of the tahini phases. With these effects, the size of the oil particles in the product is prevented from growing and a more homogeneous structure is formed. This ensures that the product remains stable for a longer period of time and that the oil separation is delayed. In order to prevent oil decomposition, lecithin as an emulsifier with vegetable fibers (sugar beet fiber and sesame fiber) was added to the tahini samples in different proportions. In the first part of the study, different amounts of sugar beet fiber (0.0%, 0.5%, 1.0% and 2%) were used in the study. It resulted in significant decreases in the amount of oil separated from the product kept at room temperature for 30 days. In the study, the amount of added sugar beet fibers were 0.0%, 0.5%, 1.0% and 2.0%, and was found oil separation from tahini 8.54%, 6.48%, 5.71% and 3.75%, respectively. The results of the study are of great importance in order to prevent oil separation and to provide customer satisfaction.

Keywords: Tahini, vegetable fiber, phase separation, sugar beet fiber, storage periods



# The Determination of Quality Properties of Traditional Kastamonu Pastırma

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Abstract: Pastirma, a traditional Turkish dry-cured meat product, is produced by curing, drying and covering with cemen of meat pieces from certain parts of beef or water buffalo carcasses. This product is included in the intermediate moisture food class. This traditional meat product is widely produced and likely consumed in Turkey. The production period is approximately one month. Many types of pastirma are produced from a carcass having various properties of shape and quality. In pastirma production, 16 and even more types of pastirma can be produced from one carcass. The naming of pastirma types is made according to the part where the muscles and/or muscles are located. According to the classification made in terms of quality, sirt and kusgömü are the first class types of pastirma. Kastamonu pastirma is an inheritance from the ancestors and is transferred from grandfather to grandchild has got an ongoing tradition. The production of pastirma in the Kastamonu started in the 1870s and its production continues to be traditionally intact. The production is usually carried out in 1500-2000 m<sup>2</sup> facilities. Daily production capacity is between 70-100 kg. Kastamonu pastrami consists of 6 or 7 family enterprises with an annual production capacity of 35-36 tons of each. Kastamonu pastırma is produced completely naturally. Drying process is carried out in natural conditions 20-30 days depending on weather conditions. Chemical substances and preservatives are not used in the production of Kastamonu pastırma. In general, antrichot, tenderloin and back muscles of carcasses are used in pastirma production. Their production amounts are approximately the same. Taste of Kastamonu pastirma is the best because of used famous Taşköprü garlic in fenugreek paste. Taşköprü garlic gives characteristic taste and aroma to pastırma, and it also plays a very important role on physicochemical and microbiological profile of the end product. Although there are many studies on the general characteristics of the pastirma produced by different locations, there is no study the pastirma produced from Kastamonu yet. In this study, it was aimed to investigate the quality properties of pasturma samples (kuşgömü and antrikot) traditionally produced in Kastamonu with standardized methods and to evaluate the results in the context of quality properties of pastirma. In this study, two kinds of bacon (kusgömü and antrikot) samples were taken from six different companies and some chemical analyzes were made. All samples were analyzed for pH, water, protein, ash, salt, color (L\*, a\*, b\*), free fatty acid, water activity, TBARS and NPN values. According to the Turkish Food Codex Meat and Meat Products Communique, the pH value and moisture content of pastirma must not exceed 6.00 and 50%, respectively. In the study, although the water values of the majority of the pastirma were below 50%, and pH values were found higher than 6.0. Also, there were differences between pastirma types in terms of the other quality property values (protein, ash, salt, color (L\*,a\*,b\*), free fat acid, water activity, TBARS, and NPN values) (P>0.05).

Keywords: Kastamonu pastırma, quality properties, physicochemical properties, oxidation, water activity



# The Effect of Roasted Hazelnut Skin on the Oxidative Properties of Frankfurter Sausages

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Abstract: Hazelnut skin arises as a by-product in the roasting process of hazelnuts. Hazelnut skin is removed from kernel after roasting process of hazelnut and represents about 2.5 % of total hazelnut kernel weight. It is an extremely rich source of fiber and several different polyphenolic compounds such as derivatives of benzoic acid (gallic acid) and cinnamic acid (p-coumaric, ferulic, sinapic, caffeic acid). The most common phenolic compound in the hazelnut skin is gallic acid and its amount is 39.1µg g<sup>-1</sup>. In recent studies, the hazelnut skin is an important component of the food industry due to its significant phenolic compounds, high antioxidant activity and low cost and also rich in coloring agents. The most important problem in the emulsion type meat products is the emulsion stability and hydrolitic and oxidative degradation of the product. Emulsion stability is the indication of the amount of water and oil remaining in the emulsion without leaving the emulsion. Since the emulsions cannot remain stable, they are either heat treated to stabilize the emulsion or additives such as chemicals or protein concentrates are used to maintain stability. In addition, in order to prevent the coalescence of the oil droplets in the emulsions, the viscosity of the aqueous phase is increased and the shelf life is extended. In order to ensure the stability of emulsion products, the need for new fillers has increased and new substances are needed to replace soy. One of the ingredients that can be used for this purpose is both the phenolic substances and the fiber-rich hazelnut skin. In the study, the average composition of the hazelnut skin was determined as moisture (5.22 %), protein (6.96 %), fat (16.47 %), carbohydrate (69.38 %) and ash content (1.97 %). In the production of frankfurter sausages, it was added that the hazelnut skin to the composition in different proportions (0.5%, 1 and 2). It provided significant reductions in the amount of free fatty acids from the degradation criteria of the oils which had a significant effect on the quality of the stored product (60 days). At the end of the storage period, a decrease in free fat acid value of up to 50 % was achieved in the hazelnut skin products. As a result, meat products can be enriched with hazelnut skin to increase dietary fiber content and prevent hydrolitic and oxidative degradation.

Keywords: Hazelnut skin, emulsion, frankfurter Sausage, free fat acid, oxidative degradation



# Importance of Allergens for the Food Safety Management in the Meat Products Facilities

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Abstract: Food safety is defined as taking necessary measures to ensure reliable food production in the stages of the provision of raw materials, production, processing, storage, transportation, distribution, and consumption. This chain includes the traceability system of all stages from farm to fork. The hazards that the food can be exposed to are examined in four groups as physical, chemical, biological, and allergen which is recently new. Allergen hazards is listed as glutencontaining cereals, crustaceans and their products, egg and egg products, fish and fish products, peanut and peanut products, soybeans and soybean products, milk and dairy products (including lactose), nuts (almonds, hazelnuts, cashew, walnut, pistachio, and their products), celery and celery products, mustard and mustard products, sesame seed and sesame products, sulphur dioxide and sulphides, lupine and lupine products, molluscs and their products. Allergen hazards required on the labels of these products should be made and the result should be taken against allergy. Red meat or poultry meat is used as a raw material in meat products and these products do not contain allergen risk. The side ingredients, spice mix (ginger, coriander, nutmeg, black pepper, white pepper, etc.), soy protein, potato starch, powder garlic, salt, sodium nitrite (E250), artificial sheath, and similar substances in the production of emulsified meat products are identified as allergen hazards. One of the most important ingredients is soy protein and it must be stored and processed, separately. Because of the medium risk as a control measure, it is appropriate to take precautions with operational pre-requisite programs. As a preventive activity and verification activity, there should be no control of the supplier analyses and the process of transferring the soy protein from the time of admission to storage. Transferring closed containers designed for allergen transport can prevent the transmission caused by many reasons such as transfer packaging rupture in the facility. If the control measure in the examined step of the flowchart, such as other types of hazard of allergen hazards, does not occur, no further measures can be taken in the next step. In the production of meat products, there is no possibility of eliminating allergy in the process stages such as filling, heat treatment, and packaging.

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Keywords: Food hazards, control precaution, risk assessment, contamination, traceability



# **Determination of Radon Emanation Power and Mass Exhalation Rate of Sepiolite Minerals**

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Abstract: Radon (222Rn) is a naturally occurring radioactive noble gas (half-life 3.8 days) which is decay products of radium (226Ra) in the uranium (238U) series. The radon gas decays into short-lived radon progeny such as polonium (218Po), lead (214Pb), bismuth (214Bi) and polonium (214Po). Radon progeny are radioactive metal atoms which get caught in our respiratory tracts during inhalation. According to the World Health Organization (WHO), depending on where you live, 3%-14% of all incidences of lung cancer are caused by radon. Therefore, knowledge of the 222Rn emanation coefficient and 222Rn mass exhalation rate of building materials is very important to evaluate the individual contribution of each material to the total indoor radon exposure. Sepiolite have been widely used as additive raw material in ceramics and cement industry, paint, agriculture, fertilizer etc. In this study, the radon emanation power and mass exhalation rate of thirty sepiolite samples collected from open three sepiolite quarries (Beylikova, Polatlı and Sivrihisar) in Central Anatolia region of Turkey were determined by using a gamma-ray spectrometry with a high purity germanium detector (HPGe) detector. The average emanation coefficient and exhalation rate of radon of sepiolite samples were determined as 0.2 and 16.9 μBq/kg s, respectively.

Keywords: Radon Emanation Power, Mass Exhalation Rate, Sepiolite Minerals, Central Anatolia Region.

#### **1. INTRODUCTION**

Sepiolite structurally formed by two tetrahedral silica sheets and a central sheet of magnesium oxide-hydroxide is a member of 2:1 phyllosilicates family. The radiometric characterization and chemical compositions of sepiolite samples investigated to determine from available sepiolite deposits collected in Turkey /Ankara-Polath.Sepiolite is a naturally occurring clay mineral of sedimentary origin. It is a non-swelling, lightweight, porous clay with a large specific surface area. Chemically, sepiolite is a hydrousmagnesiumsilicate.(Si12O30Mg8(OH)4(H2O)4•8H2O).Sepiolite has wide range of industrial applications such as, animal feed bondants, paper, decolorization, pesticide and herbicide carries, raw materials in pharmaceutical, cleaning, detergent, paint, cosmetics, agriculture, fertilizer, livestock, rubber industries and production of ceramics, fiber, cat and pet litters and constuction sector. Sepiolite, formerly known as Meerschaum (sea froth), is a non-swelling, lightweight, porous clay with a large specific surface area. Unlike other clays, the individual particles of sepiolite have a needle-like morphology. The high surface area and porosity, as well as the unusual particle shape of this clay account for its outstanding sorption capacity and colloidal properties that make it a valuable material for a wide range of applications. Sepiolite is a very

uncommon clay because of both its peculiar characteristics and scarce occurrence. There are very few commercial deposits in the World. Most countries sepiolite production in the world (Turkey, United State of America, China and Spain).

#### 2. MATERIALS AND METHODS

All sepiolite samples collected were crushed and milled to fine powder then sieved through 1 mm mesh to remove stone and other impurities. The samples were dried to remove moisture. Each sample was packed in standard plastic dish and after appropriate closing of the cover and weighted. The hermetically sealed samples were stored for 4 weeks before counting to allow R226 to reach secular equilibrium. Each sepiolite sample being collected from deposits determined average gamma activity rate values by using gas proportional counters. By using G-M tube at 100 cm high, from 3 different regions (fifty samples) were collected average gamma activity rates 32.36 ,27.22and 21.85 µSv/h respectively .These results are low and below the world average(2.50mSv,UNSCEAR;United Nations Scientific Committee on the Effects of Atomic Radiation).

#### **Natural Sources of Background Radiation**

NORM refers to Naturally Occurring Radioactive Material which includes all radioactive elements, that presents in earth crust since it was created. Around 340 nuclides have been found in nature of which 70 are radioactive and found mainly among the heavy elements. Natural radiation can be classified into major.

#### U238 Decay Chain

It contains ten elements, starting with 238Uwith half-life of (4.468x109y) and ending with stable 206Pb.It includes the following elements: astatine, bismuth, polonium, protactinium, radium, radon, thallium, and thorium. The total energy released from Uranium-238 to Lead-206, including the energy lost by neutrinos, is 51.7 MeV.

#### 232Th Decay Chain

It consists of twelve elements, starting with 232Th with half-life of (1.42x1010y) and ending with the stable208Pb. The series include the following elements: actinium, bismuth, lead, polonium, radium, and radon. The total energy released as a result of Thorium-232 decaying to Lead-208, including the energy lost by neutrinos, is 42.6 MeV.

#### **HPGe Detectors**

HPGe detectors are made by highly purifying the element germanium and growing it into a crystal. This crystal goes through many processing steps ending in the attachment of positive and negative contacts which turn it into an electronic diode. A typical gamma-ray spectrometer is composed of an HPGe detector attached to a high voltage power supply, a mainamplifier, and a Multi-Channel Analyzer unit (MCA). The main function of a gamma-ray spectrometer is to absorb the energy of the incident photon and convert it into an electronic signal which is proportional to the photon energy deposited in the detector. The resultant electronic signals are used to measure the energies and the intensities of the gamma rays produced by a radioactive source, and that allows determining the types of radioisotopes present in the source and their concentrations.

## 3. RESULTS AND DISCUSSION

#### Result

## **3.1.Radiometric measurements**

A total of 10 sepiolite samples were collected randomly from Beylikova (Eskişehir), Polatlı (Ankara) and Sivrihisar (Eskişehir) sepiolite open quarries located in Central Anatolia of Turkey. The outdoor gamma dose rate of each quarry was measured three times at the point where the samples were collected. The outdoor gamma dose rate of each point was given as the average of these three measurements. The samples were coded according to location of the sampling point.

#### 3.2. Radon emanation coefficients (REC) and mass exhalation rate (MER)

The radon emanation coefficient is defined as the fraction of the total number of radon atoms generated from radium decay that arc released into the pore spaces of rock or soil. A soil's parent rock mineralogy and chemistry control the amount and distribution of radionuclides in the soil. Climate is a primary control on soil formation and with parent composition, determines the soil's emanating power and radon transport characteristics

#### Emanated radon concentration=(radium concentration × emanation coefficient).

Data on the radium content of soils is of primary importance in estimating geologic radon potential of an area.

222Rn and its decay products (218Po, 214Po, 214Pb and 214Bi) are the most important sources of human exposure to natural sources of ionising radiation. Therefore, knowledge of the 222Rn emanation coefficient and 222Rn mass exhalation rate of building materials is essential to assess the individual contribution of each material to the total indoor radon exposure. The emanation coefficient (EC) is ratio of the amount of 222Rn that enters pore spaces over the total amount of produced radon. The EC is a dimensionless parameter and represented as either a fraction or a percentage. In Table 1 shows the radiological average and world average values of the central Anatolia of Turkey .The measurement of the EC was carried out by using the gamma-ray spectrometry. This measurement method is non-destructive and indirect method. The outdoor absorbed gamma dose rates in air at a height of 1 m above the ground in sepiolite quarries were measured using dose rate meter (NEB.211L) that was produced at Çekmece Nuclear Research Centre in Istanbul. It has Geiger Muller detector and provides dose rate measurement range of 5  $\mu$ Rh-1 to 150 mRh-1. The activity concentrations of 226Ra, 232Th and 40K in the sepiolite samples were measured using a gamma-ray spectrometer with a high-resolution coaxial p-type vertical HPGe detector (GEM50P4-83).

|               | ····· ( |       |       |       |
|---------------|---------|-------|-------|-------|
|               | U238    | Th232 | K40   | Cs137 |
| 1             | 45.13   | 7.58  | 67.76 | 7.89  |
| 2             | 35.38   | 4.52  | 14.51 | 2.39  |
| 3             | 33.29   | 10.11 | 40.63 | 9.11  |
| 4             | 38.72   | 14.22 | 56.37 | 9.42  |
| 5             | 34.79   | 4.23  | 10.03 | 1.51  |
| 6             | 43.24   | 11.7  | 55.81 | 10.71 |
| 7             | 38.17   | 5.91  | 12.87 | 2.79  |
| 8             | 34.95   | 5.69  | 9.72  | 0.29  |
| 9             | 45.23   | 11.72 | 50.01 | 7.71  |
| 10            | 41.85   | 8.34  | 25.21 | 6.45  |
| Average       | 39.1    | 8.4   | 34.3  | 5.8   |
| World average | 35      | 30    | 400   | -     |

Table1. Activity concentration measurmant (beq/kg) of located in Central Anatolia of Turkey.

#### Discussion

The assessment of the background gamma radiation dose from natural sources is of particular importance because natural radiation is the largest contributor to the external dose of the humankind. These doses vary depending on the concentration of the natural radionuclides <sup>238</sup>U, <sup>232</sup>Th and <sup>40</sup>K, present in soil sands and rocks.

#### 4. CONCLUSION

In terms of radiometric characterization analysis, gamma activity rate of Ankara Polatlı zone are between (12 -40)  $\mu$ R/h so the radioactive gamma release rate are below the world average (2.5 mSv/h).Human beings are exposed to natural background radiation every day from the ground, air, water, building materials, the universe, and even elements in their own bodies. It was observed that most of natural radioactive elements present in soils derive from the members of the radioactive decay series of 238U and 232Th, along with 40K. In addition to the natural sources, artificial radionuclides like 137Cs can also be present resulting from fallout from atmospheric nuclear weapon testing and the nuclear power plant accident. Radioactive gamma release rate are below the world average. Knowledge of the activity concentrations of the radionuclides in sepiolite, emanation coefficient of sepiolite and radon mass exhalation rate of sepiolite thereby radiometric characterization of sepiolite pits or deposits is important to evaluate such risk.

#### REFERENCES

Abbaslou, H., et al., The compatibility of bentonite/sepiolite plastic concrete cut-off wall material.,

Constr. and Build. Mater., 124 (2016), 10, pp. 1165-1173.

Balci, S., Thermal decomposition of sepiolite and variations in pore structure with and without acid

pre-treatment, J. Chem. Tech. Biotechnol., 66 (1996), 1, pp. 72-78.

Murray, H.H., Zhou, H., Palygorskite and Sepiolite (Holmes). Industrial Minerals and Rocks: Commodities,

Markets, and Uses. Edited by Kogel, J. E., Trivedi, N.C., Barker, J. M., Krukowski, S. T. Published by society of Mining, Metallurgy, and Exploration, Inc. Littleton, Colorado, USA 2006.

Olivato, J.B., et al., Elaboration, morphology and properties of starch/polyester nano-biocomposites based

on sepiolit.

Turhan, Ş., et al., Natural radiation exposure and radon exhalation rate of building materials used

in Turkey, Nucl. Technol. Radiat. Prot., 33 (2018), 2, pp. 159-266.

United Nations Scientific Committee on the Effects of atomic Radiations (UNSCEAR, 2008),

Sources and effects of ionizing radiation. Volume I: Sources, Report to the General Assembly Scientific Annexes A and B. United Nations Publication, New York, USA, 2010

Yeniyol, M., Geology, mineralogy and genesis of Yenidoğan (Sivrihisar) sepiolite deposit, Mineral Res. Expl. Bull., 114 (1992), 1, pp. 51-64



# Heat Change Study of Proquazone and Proquazone Type Calcilytics as A Tow Novel Benzimidazole Derivatives

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**Abstract:** The improvement of two novel benzimidazole derivatives has been done, so the thermodynamic properties (heat capacity, entropy and enthalpy) have been calculated for temperature change, and discussed by using density functional theory calculations. as to calculate the energetic behaviour, and moment of the title compound in gas section B3LYP/6-311g(d,p) levels; Benzimidazole: is an aromatic heterocyclic compound which can be found from the synthesis of benzimidazole primarily based polyheterocycles; this compound was very important to the pharmacists for previous few decades because of its special role in a pharmacophore in healthcare chemistry and medical specialty.

Basically, benzimidazole may be a cyclic compound consisting of the fusion of benzol with iminazole that ultimately provides a privileged structure. Density functional theory calculations (B3LYP/6-311G(d,p)) were carried out to further study on the thermodynamic properties for the molecule. The thermodynamical parameters like heat capacity, entropy and enthalpy are found increasing comparing with the increase of the temperature (from 200 K to 1000 K) as well as the density functional theory is a very well established and efficient tool to predict various statistical thermodynamic properties of molecules which are important for the understanding the chemical processes.

Keywords: benzimidazole, heat capacity, thermodynamics, Entropy, Enthalpy.

## **1. INTRODUCTION**

Benzimidazole is an aromatic heterocyclic compound, the synthesis of benzimidazole primarily based polyheterocycles; took the attention of pharmacists from previous few decades because of its functions as a pharmacophore in healthcare chemistry and medical specialty. Basically, benzimidazole may be a cyclic compound consisting of the fusion of benzol with iminazole that ultimately provides a privileged structure [1].

This compound possesses several medicine's properties until currently, the foremost outstanding benzimidazole moiety is N- ribosyl dimethyl benzimidazole which is can be found in nature, and it is the axial matter for atomic number 27 in cobalamin [1].



Benzimidazole possess has a lot of biological activities, like antimicrobial, anti-fungal, antihistaminic, medication, antiviral, anti-oxidant, anti-cancer, anti-ulcerative etc., that's why benzimidazole derivatives can be considered as a vital moiety of the molecules of pharmaceutical interest.

There is biological connectedness of the many heterocyclic building blocks because of the structural similarity to purine nucleobase, and as benzimidazole derivatives conjointly, by selection inhibits the epithelium cell growth then suppresses the method of ontogenesis in vitro likewise as in vivo [2,3].

In addition to this activity, substituted benzimidazoles plays the role of potent inhibitors of parietal cell (H+/K+ ATPase) proton pump. Moreover, a benzimidazole when substituted with proline bisamide, it is helpful in curing insomnia and promotes sleep; it acts as a potent Orexin inhibitor, which was found from high throughput screening. Orexin plays a wide range of functions in CNS which includes regulation of normal sleep-wake cycle and feeding behaviours. Patients of narcolepsy have seen with a dis-functioning of neurons responsible for orexin production. Benzimidazoles when substituted with aminothiol carbamoyl hydrazine carbonyl, 1,3,4 thiadiazole, and 4H-1,2,4-triazole at 1st position via a methyl chain and the 2nd position is substituted with phenyl, p-chlorophenyl, pmethoxyphenyl, and pyridinyl rings, then the derivatives of benzimidazole moiety shows potent antioxidant properties [1].

The planar chromophoric ability of benzo-annulated-benzimidazole, such as benzimidazo [1,2a] quinolines, or benzimidazo [1,2-a] quinazolines, can be used to go between adjacent base pairs of DNA molecule in the intercalation process, and they can be used as fluorescent probes in assays(homogeneous) of biological molecules.

Some reports studied that ability of benzimidazoles substituted with hydrogen at 1st position were found active against E.histolytica. Proquazone agent (Fig1), is a non-steroidals anti-inflammatory NSAID (Nonsteroidal anti-inflammatory drugs), which are unlike most other NSAIDs.

This agent does not have a free acid group in its structure. It is advocated for use in rheumatoid arthritis, ankylosing spondylitis, osteoarthritis, musculoskeletal disorders, acute inflammatory conditions, and sever pain states such as dysmenorrhoea, postoperative pain, and headache.





#### **Thermodynamic Parameters**

The sum of translation, rotational, vibrational and electronic energies gives the total energy of the molecule. The calculated minimum energy (SCF energy) corresponds to optimized structure of the molecule. Zero-point Vibrational energy is the lowest possible energy of the quantum mechanical system. The amount of heat required to raise the temperature of substance by one degree is the heat capacity. The quantitative measure of randomness in the system is entropy [1]. In here, using B3LYP/6-311G(d,p) basis set the standard statistical thermodynamic parameters such as

enthalpy  $(H_m^0)$ , heat capacity  $(C_{p,m}^0)$  and entropy  $(S_m^0)$ , for the title molecule were obtained from the theoretical harmonic frequencies in the range of temperature 200-1000 K and tabulated in (Table1, Table2)[11,12]. The zero-point correction is found as 0.488281 (Hartree/Particle). For an accurate prediction in determining the thermodynamic functions, we used a scale factor for frequencies (0.96) [5,13].

| Table 1. Thermod | ynamic properti | es at different temp | eratures of 1 molecule. |
|------------------|-----------------|----------------------|-------------------------|

| T (K) | $H^0_{m  ( m kcal}$ mol-1) | $C^0_{p,m}$ (cal mol-<br>1 K-1) | $S_m^0$ (cal mol-1<br>K-1) |
|-------|----------------------------|---------------------------------|----------------------------|
| 200   | 7.847                      | 66.860                          | 135.353                    |
| 250   | 11.646                     | 81.187                          | 152.247                    |
| 300   | 16.169                     | 95.764                          | 168.700                    |
| 350   | 21.417                     | 110.097                         | 184.851                    |
| 400   | 27.365                     | 123.703                         | 200.716                    |
| 450   | 33.969                     | 136.294                         | 216.258                    |
| 500   | 41.175                     | 147.769                         | 231.431                    |
| 550   | 48.926                     | 158.151                         | 246.200                    |
| 600   | 57.172                     | 167.527                         | 260.543                    |
| 650   | 65.863                     | 176.002                         | 274.451                    |
| 700   | 74.957                     | 183.683                         | 287.928                    |
| 750   | 84.418                     | 190.666                         | 300.979                    |
| 800   | 94.212                     | 197.033                         | 313.619                    |
| 850   | 104.311                    | 202.854                         | 325.862                    |
| 900   | 114.688                    | 208.188                         | 337.724                    |
| 950   | 125.321                    | 213.086                         | 349.220                    |
| 1000  | 136.189                    | 217.590                         | 360.368                    |

**Table 2.** Thermodynamic properties at different temperatures of (2) molecules.

| T<br>(K) | $H^0_m$ (kcal mol-1) | $C^0_{p,m}$ (cal mol-1 K-1) | $S_m^0$ (cal mol-1<br>K-1) |
|----------|----------------------|-----------------------------|----------------------------|
| 200      | 10.720               | 89.723                      | 178.702                    |
| 250      | 15.795               | 109.398                     | 201.270                    |
| 300      | 21.862               | 129.271                     | 223.338                    |
| 350      | 28.911               | 148.532                     | 245.030                    |
| 400      | 36.893               | 166.560                     | 266.322                    |
| 450      | 45.739               | 183.041                     | 287.141                    |
| 500      | 55.369               | 197.907                     | 307.419                    |
| 550      | 65.703               | 211.239                     | 327.108                    |
| 600      | 76.669               | 223.188                     | 346.183                    |
| 650      | 88.200               | 233.917                     | 364.638                    |
| 700      | 100.242              | 243.586                     | 382.480                    |
| 750      | 112.742              | 252.331                     | 399.726                    |
| 800      | 125.660              | 260.270                     | 416.397                    |
| 850      | 138.956              | 267.501                     | 432.517                    |
| 900      | 152.599              | 274.106                     | 448.110                    |

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| 950  | 166.556 | 280.155 | 463.202 |
|------|---------|---------|---------|
| 1000 | 180.804 | 285.705 | 477.817 |

In accordance with the equipartition theorem, the translational energy, rotational energy and molecular vibrational intensities increase with temperature [6,7]. As a result the thermodynamic functions increase with increasing temperature ranging from 200 to 1000 K as shown in (Table1, and Table2). The correlation equations between enthalpy, heat capacity, entropy, and temperatures were fitted by linear and quadratic formulas and the corresponding fitting factors (R2) for these thermodynamic properties are 0.99969, 0.99952, 0.99999 respectively. By using these equations (1)-(3) [8.9], we can predict the attributes at any point of temperature without further computational procedures. It is inferred from the results that the thermodynamic parameters linearly depend on the temperature value [10]. The correlations between the thermodynamic properties and temperatures T values are shown in Fig. 2. The corresponding fitting equations are as follows:

(2)

(3)

H = -10.45765 + 0.07349T - 1.19271x10 - 4T2; (R2 = 0.99969)(1)

$$C = -3.0709 + 0.51119T - 2.24854x10 - 4T2$$
; (R2 = 0.99952)

S = 80.95514 + 0.50902T - 1.12167x10 - 4T2; (R2 = 0.99999)



Figure 2. Correlation graph of heat capacity, entropy and enthalpy with respect to temperature.

From the obtained results, thermodynamic properties are useful for the further studies of the title compound. For instance, the approximate direction of chemical reactions and the relation of thermodynamic energies with thermodynamic functions can be calculated from the above inferences[5].

#### 4. CONCLUSION

Density functional theory B3LYP/6-311G (d,p) calculations were carried out to further study on the thermodynamic properties for the molecule. The thermodynamically parameters like heat capacity, entropy and enthalpy are found increasing with the increase of the temperature (from 200 K to 1000K).

#### REFERENCES

Addison, S. R. (2001). Heat Capacity, Specific Heat, and Enthalpy Heat Capacities at Constant Volume and Pres-.

Chodera, J. D., & Mobley, D. L. (2013). Entropy-Enthalpy Compensation: Role and Ramifications in Biomolecular Ligand Recognition and Design. Annual Review of Biophysics, 42(1), 121–142. https://doi.org/10.1146/annurev-biophys-083012-130318

Einstein, A. (n.d.). no t t o N be C re ER pu T bl is he d o N be C re ER pu T bl is d, 154-184.

- Ford, K. W. (2016). Entropy and the second law of thermodynamics. Basic Physics, 371–396. https://doi.org/10.1142/9789813208025\_0014
- Jacob, K. T., & Rajitha, G. (2009). Discussion of enthalpy, entropy and free energy of formation of GaN. Journal of Crystal Growth, 311(14), 3806–3810. https://doi.org/10.1016/j.jcrysgro.2009.05.016
- Jang, H., Haq, M. R., Ju, J., Kim, Y., Kim, S. M., & Lim, J. (2017). Fabrication of all glass bifurcation microfluidic chip for blood plasma separation. Micromachines, 8(3). https://doi.org/10.3390/mi8030067
- Leff, H. S. (2012). Removing the Mystery of Entropy and Thermodynamics Part V. The Physics Teacher, 50(5), 274–276. https://doi.org/10.1119/1.3703541
- Pan, A., Biswas, T., Rakshit, A. K., & Moulik, S. P. (2015). Enthalpy-Entropy Compensation (EEC) Effect: A Revisit. Journal of Physical Chemistry B, 119(52), 15876–15884. https://doi.org/10.1021/acs.jpcb.5b09925
- Raja, M., Muhamed, R. R., Muthu, S., & Suresh, M. (2017). Synthesis, spectroscopic (FT-IR, FT-Raman, NMR, UV–Visible), first order hyperpolarizability, NBO and molecular docking study of (E)-1-(4-bromobenzylidene)semicarbazide. Journal of Molecular Structure, 1128(September), 481–492. https://doi.org/10.1016/j.molstruc.2016.09.017
- Rard, J. A., Plyasunov, A. V, Grenthe, I., Germain, L. S., & Îles, B. (1999). TDB-4 TEMPERATURE CORRECTIONS TO THERMODYNAMIC DATA AND Ignasi Puigdomènech. October, (October).
- Sarojinidevi, K., Subramani, P., Jeeva, M., Sundaraganesan, N., SusaiBoobalan, M., & VenkatesaPrabhu, G. (2019). Synthesis, molecular structure, quantum chemical analysis, spectroscopic and molecular docking studies of N-(Morpholinomethyl) succinimide using DFT method. Journal of Molecular Structure, 1175(August), 609–623. https://doi.org/10.1016/j.molstruc.2018.07.101
- Sharma, K., Melavanki, R., Patil, S. S., Kusanur, R., Patil, N. R., & Shelar, V. M. (2019). Spectroscopic behavior, FMO, NLO and NBO analysis of two novel aryl boronic acid derivatives: Experimental and theoretical insights. Journal of Molecular Structure, 1181, 474–487. https://doi.org/10.1016/j.molstruc.2018.12.086
- Starikov. (2013). Entropy-Enthalpy Compensation and its Significance in Particular for Nanoscale Events. Journal of Applied Solution Chemistry and Modeling, 2(2), 1–6. https://doi.org/10.6000/1929-5030.2013.02.02.7



# Design of The Tubular Heat Exchanger of a Cogeneration System Burning Hen Manure

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**Abstract:** One of the main problems in the world is the elimination of poultry manure that came from cage house. Poultry manure causes illnesses, bad smell and leads to formation of larval. To avoid these unwanted situations, it is necessary to disposal the poultry manure. When the environmental effect is considered, it is important that chicken manure can be eliminated in small, medium, and large-scale poultry house.

Combustion of laying hen manure is the most difficult process in literature and field application. There is no combined heat and power system that includes drying process of manure with waste heat, combustion with fluidized bed reactor and electric generation in the world. A special fluidized bed reactor design is required for the combustion process of the laying hen manure which include high alkaline and ash contents.

This study is about the oil heat exchanger design for heating the thermal oil from the hot gas resulting from the combustion of the poultry manure in a fluidized bed reactor. HTRI heat exchanger design software was used for this purpose. The heat transfer surface area and the number of the tubes in the heat exchanger were determined. The output temperatures obtained from the software and test facility are found to be similar.

Keywords: Heat exchanger, HTRI, heat transfer coefficient, LMTD, EMTD Poultry, Manure, Cogeneration.

# **1. INTRODUCTION**

According to Turkish Statistical Institute data, in 2016, the number of industrial scale poultry is 328 million. An average of 30.000 tons of 50% to 75% moisture content manure to be disposed of per day is taken out of these facilities. Although there are different disposal methods, the prohibition of the majority of the excreted excreta is unfortunately disturbed by the eyes and distant places because of the high investment costs, operating costs and high energy demand.

**Table 1.** Number of birds according to species

| Years | Laying     | Meat        | Turkey     | Goose      | Duck       |
|-------|------------|-------------|------------|------------|------------|
|       | Hens       | Chicken     | (quantity) | (quantity) | (quantity) |
|       | (quantity) | (quantity)  |            |            |            |
| 1991  | 50.826.656 | 88.379.548  | 3.132.676  | 1.599.831  | 1.112.015  |
| 1992  | 52.224.952 | 100.305.100 | 3.332.794  | 1.752.495  | 1.154.743  |
| 1993  | 58.179.047 | 120.080.935 | 3.340.241  | 1.687.596  | 1.171.961  |
| 1994  | 57.842.034 | 125.842.269 | 3.441.995  | 1.719.833  | 1.186.891  |
| 1995  | 57.324.654 | 71.689.773  | 3.291.000  | 1.745.163  | 1.199.925  |
| 1996  | 53.883.070 | 99.073.900  | 3.063.540  | 1.641.915  | 1.093.860  |
| 1997  | 61.401.783 | 104.870.702 | 5.327.501  | 1.794.610  | 1.828.792  |
| 1998  | 69.722.271 | 167.275.380 | 3.805.345  | 1.771.327  | 1.339.468  |
| 1999  | 71.885.207 | 167.862.730 | 3.762.516  | 1.670.916  | 1.294.824  |
| 2000  | 64.709.040 | 193.459.280 | 3.681.558  | 1.496.604  | 1.104.176  |
| 2001  | 55.675.750 | 161.899.442 | 3.254.018  | 1.397.560  | 913.748    |
| 2002  | 57.139.257 | 188.637.066 | 3.092.408  | 1.400.136  | 832.091    |
| 2003  | 60.399.520 | 217.133.076 | 3.994.093  | 1.336.775  | 810.910    |
| 2004  | 58.774.172 | 238.101.895 | 3.902.346  | 1.250.634  | 770.436    |
| 2005  | 60.275.674 | 257.221.440 | 3.697.103  | 1.066.581  | 656.409    |

| 2006 | 58.698.485  | 286.121.360 | 3.226.941 | 830.081   | 525.250 |
|------|-------------|-------------|-----------|-----------|---------|
| 2007 | 64.286.383  | 205.082.159 | 2.675.407 | 1.022.711 | 481.829 |
| 2008 | 63.364.818  | 180.915.558 | 3.230.318 | 1.062.887 | 470.158 |
| 2009 | 66.500.461  | 163.468.942 | 2.755.349 | 944.731   | 412.723 |
| 2010 | 70.933.660  | 163.984.725 | 2.942.170 | 715.555   | 396.851 |
| 2011 | 78.956.861  | 158.916.608 | 2.563.330 | 679.516   | 382.223 |
| 2012 | 84.677.290  | 169.034.283 | 2.760.859 | 676.179   | 356.730 |
| 2013 | 88.720.709  | 177.432.745 | 2.925.473 | 755.286   | 367.821 |
| 2014 | 93.751.470  | 199.976.150 | 2.990.304 | 911.990   | 399.820 |
| 2015 | 98.597.340  | 213.658.294 | 2.827.731 | 850.694   | 398.387 |
| 2016 | 108.689.236 | 220.322.081 | 3.182.751 | 933.353   | 413.841 |

If manure is not evaluated, they can be harmful just like industrial wastes. If the chicken is not processed and stored in an appropriate manner, it will cause loss of nutritional value, contaminate the soil surface and groundwater, and mediate spread of diseases. In order to avoid these unwanted situations, it is necessary to properly process the collect from the poultry houses. The manure taken from the poultry farm is about 75-80% moisture content. In this humidity manure cannot be stored, cannot be given directly to the soil and cannot be processed. Moisture content is reduced to about 20 - 25% by the processing of the manure.

The chicken manure characteristic and daily amount of manure could be different to each farm. The feed rations used, the weather conditions at the location, the breed of chickens, the age of the birds are the main reasons for this variability. Laying hen manure generally has a mixed and heterogeneous structure for thermal cycles. The most important properties of biomass which is chicken manure are moisture content, elemental composition, ash content, calorific value, fixed carbon fractions and volatile matter content. For example, the amount of energy to be obtained in the process of burning chicken manure is determined by the lower heat value (LHV) and the technical work is carried out accordingly. The lower calorific value is directly related to the moisture content in the manure. The higher the humidity in it, the lower the value of the thermal value decreases with humidity. The characteristic of manure is shown Table 2 (Font-Palma, 2012)

| Average values as dry basis (% weight) |           |           |  |  |
|--|-----------|-----------|--|--|
|  | Wet basis | Dry basis |  |  |
| Fixed carbon %                         | 1,32      | 3,04      |  |  |
| Volatile matter %                      | 16,62     | 43,19     |  |  |
| Moisture %                             | 75,44     | 27,63     |  |  |
| Ash %                                  | 6,62      | 26,14     |  |  |
| Carbon %                               | 41,96     | 35,2      |  |  |
| Hydrohen %                             | 4,62      | 2,07      |  |  |
| Oxigen %                               | 20,53     | 22,42     |  |  |
| Nitrogen %                             | 5,32      | 3,35      |  |  |
| Sulphur %                              | 0,62      | 0,84      |  |  |
| High heating value<br>(MJ/kg)          |           |           |  |  |
| Wet                                    | 14,96     |           |  |  |
| Dry                                    |           | 11,40     |  |  |
| Lower heating value<br>(MJ/kg)         |           |           |  |  |
| Wet                                    | 13,91     |           |  |  |
| Dry                                    |           | 10,53     |  |  |

Table 2. Laying hen manure elemental analysis and calorific values

Thermal energy obtained by the combustion of the chicken manure is converted into electricity by the Organic Rankine Cycle (ORC) which is closed loop. Waste heat from the ORC will be used in the manure drying system. This cogeneration plant, which is not practiced in the world due to the combustion process of laying hen manure, consists of 3 parts in total. The installation of the pilot plant will be carried out in the Manisa region. Güres Group supported all research and development activities.

- A- Manure drying system
- B- Manure combustion system
- C- Electricity generation by ORC



Figure 1. Cogeneration system schematic diagram

Detail of the Figure 1 is;

1- Fuel Feeding System: Chicken manure with 25-35% moisture content is fed homogeneously to the fluidized bed gasifier.

2- Chicken manure is gasified and burned with a minimum of 95% efficiency in a 750 ° C fluidized bed with partial air.

3- A secondary air supply is made to achieve complete combustion, resulting in a combustion gas that remains at 850-950  $^{\circ}$  C for a minimum of 2.5 seconds.

4- The ash-rich gas first passes through the radiation economizer and transfers heat to the thermal oil. Large particles are first caught under the radiation economizer.

5- The flue gas leaves at 330  $^{\circ}$ C, transferring it to the oil in the cassette type exchanger. Automatic cleaners are available on every cassette. Thus, the system works efficiently for a long time without stopping. In addition, the heat transfer metal surface temperature is kept below 350  $^{\circ}$ C and the corrosion mechanism caused by alkaline and chlorine in the manure is removed.

6- The fly ash in the gas from the oil economizer is kept in the cyclone and the cleaned combustion gas is sent to the air economizer.

7- The air economizer preheats the primary and secondary fresh air to reduce waste gas losses to a minimum. The heated fresh air provides gasification in the fluidized bed and stabilizes the combustion reactions at the combustion side.

8- The flue gas is passed through a wet scrubber, reducing the amount of dust below 50 mg / Nm3.

9- The ash resulting from the high ash content of the fuel is collected from many different points of the system, continuously removed from the system and collected in the ash silos. The ash containing high potassium and phosphorus content is regarded as an organomineral fertilizer raw material.

10- Organic Rankin Cycle system is used in 19% of the thermal oil energy for electricity generation and 81% for closed cycle hot water in the manure drying and / or poultry heating.

11- The manure drying system uses waste heat energy to dry wet manure a day to ensure the necessary fuel for the cogeneration system.

One of the important issues after installation of the system is the transfer of the thermal energy to be used in the thermal oil to be used for this by using the manual and utilities appropriate heat exchanger design process should be performed.

### 2. MATERIALS AND METHODS

#### Shell and Tube Heat Exchanger Design Procedures

Heat transfer or the size of heat exchanger

$$Q = U_o A_o \Delta T_m \tag{1}$$

The overall heat transfer coefficient  $U_0$  based on the O.D. of tubes can be estimated from the estimated values of individual heat transfer coefficients, the wall and fouling resistance and the overall surface efficiency using equation

$$\frac{1}{U_o} = \frac{A_o}{A_i} \left[ \frac{1}{\eta_i h_i} + \frac{R_{f_i}}{\eta_i} \right] + A_o R_w + \frac{R_{f_o}}{\eta_o} + \frac{1}{\eta_o h_o}$$
(2)

For the single tube pass, purely countercurrent heat exchanger, F= 1.00. For preliminary design shell with any even number of tube side passes, F may be estimated as 0.9

Heat load can be estimated from the heat balance as:

$$Q = (mC_p)_c (T_{c2} - T_{c1}) = (mC_p)_h (T_{h2} - T_{h1})$$
(3)

If one stream changes phase:

$$Q = mh_{fg} \tag{4}$$

LMTD (Log Mean Temperature Difference Method) calculation:

If three temperatures are known, the fourth one can be found from the heat balance,

$$\Delta T_{lm} = \frac{(T_{h1} - T_{c2}) - (T_{h2} - T_{c1})}{\ln \frac{(T_{h1} - T_{c2})}{(T_{h2} - T_{c1})}}$$
(5)

Heat transfer area can be calculated from equation (1). Number of tubes of diameter  $(d_o)$ , shell diameter  $(D_s)$  to accommodate the number of tubes  $(N_t)$ , with given tube length (L) can be estimated,

$$A_o = \pi_e N_t L \tag{6}$$

One can find the shell diameter  $(D_s)$ , which would contain the right number of tubes  $(N_t)$ , of diameter  $(d_t)$ .

The total number of tubes can be predicted in fair approximation as function of the shell diameter by taking the shell circle and dividing it by the projected area of the tube layout pertaining to a single tube  $A_1$ .

$$N_t = (CTP) \frac{\pi D_s^2}{4A_i} \tag{7}$$

Where CTP is the tube count calculation constant that accounts for the incomplete coverage of the shell diameter by the tubes.

Based on fixed tube sheet the following values are suggested:

| One tube pass:       | CTP = 0.93 |
|----------------------|------------|
| Two tube pass:       | CTP = 0.90 |
| Three tube pass:     | CTP = 0.85 |
| $A_1 = (CL) (P_T)^2$ |            |

Where CL is the tube layout constant:

CL=1.0 for  $90^{\rm o}$  and  $45^{\rm o}$ 

CL=0.87 for  $30^{\rm o}$  and  $60^{\rm o}$ 

Equation (7) can be written as:

$$N_{t} = 0.875 \left(\frac{CTP}{CL}\right) \frac{D_{s}^{2}}{(P_{r})^{2} d_{o}^{2}}$$
(9)

Where  $P_R$  is the Tube Pitch Ratio ( $P_R = P_T/d_o$ ).

The shell diameter in terms of main construction diameter can be obtained as from equations (6) and (9),

(8)

$$Ds = 0.637 \sqrt{\frac{CL}{CTP}} \left(\frac{A_o (P_r)^2 d_o}{L}\right)^{1/2}$$
(10)

#### **Tube Side Pressure Drop**

The tube side pressure drop can be calculated by knowing the number of tube passes  $(N_p)$  and length (L) oh heat exchanger,

The pressure drop for the tube side fluid is given by equation

$$\Delta P_t = 4f \, \frac{LN_p}{d_i} \, \rho \, \frac{u_m^2}{2} \tag{11}$$

$$\Delta P_{t} = 4f \, \frac{LN_{p}}{d_{i}} \frac{G_{i}^{2}}{2\rho} \tag{12}$$

The change of direction in the passes introduction in the passes introduction an additional pressure drop due to sudden expansions and contractions that the tube fluid experiences during a return that is accounted for allowing four velocity head per pass

$$\Delta P_t = 4N_p \frac{\rho u_m^2}{2} \tag{13}$$

The total pressure drop of the side becomes:

$$\Delta P_t = \left(4f \frac{LN_p}{d_i} + 4N_p\right) \frac{\rho u_m^2}{2} \tag{14}$$

#### Shell Side Pressure Drop

Kern suggested following correlations for the shell side heat transfer coefficient;

$$\frac{h_o D_e}{k} = 0.36 \left(\frac{D_e G_s}{\mu}\right)^{0.55} \left(\frac{c_p \mu}{k}\right)^{\frac{1}{3}} \left(\frac{\mu_b}{\mu_w}\right)^{0.14}$$
(15)

For,  $400 < \text{Re}_s = \frac{\sigma_s D_s}{\mu} \le 1 \times 10^6$ 

The equivalent diameter (De) can be given by

For square pitch;

$$D_{e} = \frac{4}{\pi d_{o}} \left( P_{T}^{2} - \frac{\pi d_{o}^{2}}{4} \right)$$
(16)

For triangular pitch;

$$D_{e} = \frac{4}{\pi d_{o}/2} \left( \frac{P_{T}^{2} \sqrt{3}}{4} - \frac{\pi d_{o}^{2}}{8} \right)$$
(17)

Bundle cross flow area  $A_s$ , at the centre of the shell;

$$A_s = \frac{D_s CB}{P_T} \tag{18}$$

Where, C = clearance between adjacent tubes

B = baffle spacing

 $P_T = pitch of tubes$ 

$$D_s =$$
 equivalent diameter

Shell side mass flow rate G<sub>s</sub>;

$$G_s = \frac{m}{A_s} \tag{19}$$

Where, m = mass flow rate

This method is based on Kern theory. The shell side pressure drop depends on the number of tubes, the number of times the fluid passes the tube bundle between the baffles and the length of each crossing.

The pressure drop on the shell side is calculated by the following expression:

$$\Delta P_s = f \frac{G_s^2 (N_b + 1) D_s}{2\rho D_s \phi_s} \tag{20}$$

Where,  $\phi_s = (\mu_b / \mu_s)^{0.14}$ 

 $N_b = Number of baffles$ 

 $(N_b + 1) =$  Number of times fluid passes to the tube bundle

Friction factor (*f*) calculated from:

$$f = \exp(o.576 - 0.19 \ln \text{Re}_s)$$
 (21)

Where,

$$400 < \operatorname{Re}_{s} = \frac{\sigma_{s} D_{s}}{\mu} \le 1 \times 10^{6}$$
<sup>(22)</sup>

The correlation has been tested based on data obtained on actual exchangers. The friction coefficient also takes entrance and exit losses into account.

# 3. RESULTS AND DISCUSSION

## Heat Exchanger Design of Cogeneration System

The establishment of the facility where the research and development activities are completed within the Güres Group is completed and the data obtained from this will be compared with the data obtained in the HTRI. The HTRI program also allows us to graphically retrieve the data.

The thermal oil circulates at about 263.9 ° C from the ORC and is distributed to three different heat exchangers. The first heat exchanger in the system which is a shell and tube heat exchanger will be evaluated here.

The following data are given:

Gas inlet temperature =  $846.4 \circ C$ 

Gas outlet temperature = 719 ° C

Oil inlet temperature =  $263.9 \circ C$ 

Gas mass flow rate=18.94 kg/s,

Oil mass flow rate=36.6 kg/s

Based on the calculations given in the above equations and using the HTRI software, the heat transfer surface area is calculated as  $96.6 \text{ m}^2$ . In the software, 13x14 tube bundle with rectangular arrangement was chosen. The configuration in the HTRI software is shown in Figure 2. The tubular heat exchanger design conditions are also provided to the HTRI software and they are tabulated in Table 3. Table 4 presents the output obtained from the HTRI software such as the layouts of the tubes (transverse and longitudinal pitches), heat transfer coefficients of the tube side, shell side and overall heat transfer coefficients.

The tubular heat exchanger which is a part of the full system was constructed and used in the ORC system within the Güres Group. An AUTOCAD picture of the construction of the tubular heat exchanger is illustrated in Figure 3. Table 5 shows the oil outlet temperature obtained from the software and obtained from the real facility.



Figure 2. and 3. Heat exchanger appearance in HTRI, Figure 3: Production picture designed in Autocad.

| Table 3. | Design | conditions | given to    | the | HTRI | software |
|----------|--------|------------|-------------|-----|------|----------|
|          |        |            | G · · · · · |     |      |          |

| Process Conditions            |             | Out    | side      | Tube    | side         |
|-------------------------------|-------------|--------|-----------|---------|--------------|
| Fluid name                    | gas         |        |           | yag     |              |
| Fluid condition               |             |        | Sens. Gas |         | Sens. Liquid |
| Total flow rate               | (kg/s)      |        | 18,940    |         | 36,600       |
| Weight fraction vapor, In/Out |             | 1,000  | 1,000     | 0,000   | 0,000        |
| Temperature, In/Out           | (Deg C)     | 846,40 | 720,00    | 264,00  | 293,56       |
| Skin temperature, Min/Max     | (Deg C)     | 274,49 | 305,50    | 271,31  | 301,67       |
| Pressure, Inlet/Outlet        | (kPa)       | 1,000  | 0,000     | 0,000   | 0,000        |
| Pressure drop, Total/Allow    | (kPa) (kPa) | 31,548 | 0,000     | 374,911 | 0,000        |
| Midpoint velocity             | (m/s)       |        | 20386,0   |         | 3,67         |
| - In/Out                      | (m/s)       |        |           | 3,62    | 3,71         |
| Heat transfer safety factor   | ()          |        | 1         |         | 1            |
| Fouling                       | (m2-K/W)    |        | 0,000000  |         | 0,000000     |

# Table 4. Outputs obtained from the HTRI software

| Exchanger Performance     |             |         |             |                    |            |             |    |
|---------------------------|-------------|---------|-------------|--------------------|------------|-------------|----|
| Outside film coef         | (W/m2-K)    |         | 42,73       | Actual U           | (W/m2-K)   | 41,778      |    |
| Tubeside film coef        | (W/m2-K)    |         | 3360,34     | Required U         | (W/m2-K)   | 55,622      |    |
| Clean coef                | (W/m2-K)    |         | 41,778      | Area               | (m2)       | 96,603      |    |
| Hot regime                |             |         | Sens. Gas   | Overdesign         | (%)        | -24,89      |    |
| Cold regime               |             | Se      | ens. Liquid | Tul                | be Geometr | У           |    |
| EMTD                      | (Deg C)     |         | 502,9       | Tube type          |            | Plain       |    |
| Duty                      | (MegaWatts) |         | 2,703       | Tube OD            | (mm)       | 42,300      |    |
|                           | Unit Geo    | metry   |             | Tube ID            | (mm)       | 34,300      |    |
| Bays in parallel per unit |             |         | 1           | Length             | (m)        | 4,280       |    |
| Bundles parallel per bay  |             |         | 1           | Area ratio(out/in) | ()         | 1,23324     |    |
| Extended area             | (m2)        |         | 96,603      | Layout             |            | Inline      |    |
| Bare area                 | (m2)        |         | 96,603      | Trans pitch        | (mm)       | 154,000     |    |
| Bundle width              | (m)         |         | 2,030       | Long pitch         | (mm)       | 100,000     |    |
| Nozzle                    |             | Inlet   | Outlet      | Number of passes   | ()         | 14          |    |
| Number                    | ()          | 1       | 1           | Number of rows     | ()         | 14          |    |
| Diameter                  | (mm)        | 154,051 | 154,051     | Tubecount          | ()         | 182         |    |
| Velocity                  | (m/s)       | 2,34    | 2,39        | Tubecount Odd/Eve  | n ()       | 13 /        | 13 |
| R-V-SQ                    | (kg/m-s2)   | 4586,39 | 4699,52     | Tube material      | Ca         | arbon steel |    |

# Table 5. Oil outlet temperature

|                                    | Result from<br>experimental<br>set up | HTRI<br>Result |
|------------------------------------|---------------------------------------|----------------|
| <b>Oil outlet temperature</b> (°C) | 295,8                                 | 293,69         |

## 4. CONCLUSION

In this study, tubular heat exchanger of a cogeneration system which uses hen manure was designed with HRTI software. Heat transfer surface area was obtained and number of tubes and their arrangements were determined. Number of iterations and their comparison can be analyzed easily. Part of the systems such as heat exchangers in the full system can be optimized with the software in a short time which contributes to the better thermal performance of the overall system.

### REFERENCES

- 1. Arturo R L, Miguel T V & Pedro Q D. (2011) "The Design Of Heat Exchanger", science research. Vol 3 pp 911-920
- 2. Kakkan, S (1999). "Heat Exchangers Selection, Rating and Thermal Design". pp 263-274
- 3. Leong kc & Toh kc (1998), "shell and tube heat exchanger design software for educational applications", int.j.engng.ed. vol14 pp 217-234
- 4. Shah, RK (2003). "Fundamental of heat exchanger design" Rochster Institute of Technology. pp 381
- 5. Su Thet Mon Than, (2008) "Heat Exchanger Design", world academy of science engineering and technology. pp 604-611
- 6. Design Validation of Shell and Tube Heat Exchanger by HTRI Nirmal S. Parmar
- 7. https://biruni.tuik.gov.tr/hayvancilikapp/hayvancilik.zul, Accessed on January 18, 2018
- 8. Shao, Yuanyuan and Wang, Jinsheng and Preto, Fernando and Zhu, Jesse and Xu, Chunbao Charles, 2012, "Ash Deposition in Biomass Combustion or Co-Firing for Power/Heat Generation", Energies, 5, 12, 5171--5189
- Obernberger, I.; Dahl, J.; Brunner, T. "Formation, Composition and Particle Size Distribution of Fly Ashes from Biomass Combustion Plants." In Proceedings of the 4th Biomass Conference of the Americas, Oxford, UK; Elsevier Science Ltd.: Oakland, CA, USA, 1999; pp. 1377–1385.
- 10. Font Palma, C. "Characterisation, kinetics and modelling of gasification of poultry manure and litter: An overview", Energy Conversion and Management 53 (2012) 92–98



# Effect of Rotation On the Melting of Pcm in A Horizontal Annulus

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**Abstract:** In this study, effects of the rotating surface on the heat transfer and the melting time in the melting process of the phase change materials (pcm) was examined numerically. The rotational speeds of 0 rad/s, 0.5 rad/s and 1 rad/s were imposed on the inner cylinder of a horizontal annulus. The inner cylinder is hot and the outside cylinder is cold. In the annulus part melting of PCM was analyzed numerically. It was observed that the heat transfer rate and melting were affected by the variation of rotational speed of the inner surface.

Keywords: phase change material, melting, heat transfer, rotation.

## **1. INTRODUCTION**

Energy has become a fundamental need for people. Energy production and adverse effects on energy efficiency in the use of and dependency on foreign resources and environmental conservation issues have been put forward. This awareness is a very important issue in our country, which has limited energy resources. In thermal energy storage applications, PCMs are the most important reasons for preferring high volumetric storage and isothermal behaviors [1-4].

Phase change materials are widely used today in solar energy storage, heat pumps, heating and air conditioning in buildings, heat distribution systems. As storage of energy eliminates the time or rate imbalance between energy production and consumption, it is possible to use existing energy resources more effectively. Energy storage plays an important role in energy saving as well as balancing the difference between production and consumption. While ensuring the protection of primary energy resources, it also prevents the waste of energy and makes the systems work more economically. By providing a regular energy flow, it increases the efficiency of the energy production systems and ensures its reliability.

In this study, melting of PCM in a horizontal annulus under the effects of a rotating surface was examined numerically.

# 2. MATERIALS AND METHODS

# 2.1. Numerical Analysis

Continuity, momentum and energy equations for two-dimensional time-dependent laminar flow are expressed as follows;

**Continuity Equation:** 

$$\Delta \vec{V} = \mathbf{0} \tag{1}$$

Momentum Equation:

$$\frac{\partial y}{\partial t} + \vec{V} * \nabla \vec{V} = \frac{1}{\rho} * \left\{ -\nabla P + \mu * \nabla^2 \vec{V} + \rho * \vec{g} * \beta * (T - T_{ref}) \right\} + \vec{S}$$
(2)

**Energy Equation:** 

$$\frac{\partial h_{sen}}{\partial t} + \frac{\partial h_{lat}}{\partial t} + \nabla * \left( \vec{V} * h_{sen} \right) = \nabla \left( \frac{k}{\rho * c_p} * \nabla h_{sen} \right)$$
(3)

The enthalpy of the material is the sum of the sensible heat enthalpy,  $h_{sen}$ , and latent heat enthalpy,  $h_{lat}$ ;

$$h_{sum} = h_{sen} + h_{lat} \tag{4}$$

$$h_{sen} = h_{ref} + \int_{T_{ref}}^{T} C_p * dT$$
<sup>(5)</sup>

The constant  $C_p$  in the multiplication state is separated from the integral. The latent heat content can be written in terms of the latent heat of the material, L:

$$h_{lat} = \sum_{i=1}^{n} \lambda_{i} * L$$

Here, it can be changed between 0 (for solid state) and L(for liquid state). The total symbol implies that the total latent heat is calculated by a sum of the latent heat of each cell included in the area examined at each time step. The liquid ratio is described as follows;

$$\lambda = \begin{cases} \frac{h_{lat}}{L} = 0, & if \quad x < 0\\ \frac{h_{lat}}{L} = 1, & if \quad x \ge 0\\ \frac{h_{lat}}{L} = \frac{T - T_{solid}}{T_{liquid} - T_{solid}}, & if \quad T_{solid} < T < T_{liquid} \end{cases}$$
(7)

S is the resource term defined in the equation below. This parameter is added to the momentum equation to add the effects of phase change in convective heat transfer.

$$\vec{S} = \frac{(1-\lambda)^2}{\lambda^3} A_{mush} * \vec{V}$$
(8)

The coefficient  $A_{mush}$  in this equation, is the mite region constant. This constant usually takes a value between 10<sup>4</sup> and 107.

The local Nusselt number at the hot wall is an ideal indicator showing how convection influences overall conduction through the cylinder:

$$Nu = \frac{h * L_{ref}}{\lambda} \tag{9}$$

where  $L_{ref}$  is the reference length,  $\lambda$  is the thermal conductivity and h is the heat transfer coefficient:

$$h = \frac{q}{T_w - T_{ref}} \tag{10}$$

q is the convective heat flux,  $T_w$  is the wall temperature, and  $T_{ref}$  is the reference temperature.

### **Simulation Model**

The simulation model is used to examine the rotational speed effects on the melting and heat transfer. On the outer surface of the cylinder, the pcm is solid. On the inner surface of the cylinder it is liquid. The properties of the phase-changing material used are given in Table 2;

Table 1. The main differences between different heat storage methods

| Description                      | Parameter | Value                                  |
|----------------------------------|-----------|--|
| Density                          | $ ho_0$   | 7500 kg/m <sup>3</sup>                 |
| Heat Capasity                    | $C_p$     | 200 J/kg.K                             |
| Thermal Conductivity             | k         | 60 W/m.K                               |
| Coefficient of thermal expansion | $a_p$     | 2,67.10 <sup>-4</sup> K <sup>-1</sup>  |
| Kinematic viscosity              | ν         | 8,0.10 <sup>-7</sup> m <sup>2</sup> /s |
| Fusion Temperature               | $T_f$     | 505 K°                                 |

(6)

| Latent heat of fusion | $\Delta H$ | 60 kj/kg |
|-----------------------|------------|----------|
|                       |            |          |

In the above table, the melting temperature of the phase change material is 505 K. At the outer cylinder surface, temperature was at 508 K temperature while in the inner cylinde it is 503 K. A schematic of the model is given in Figure 1.



Figure 1. Geometry and boundary conditions at starting time

The inner part of the annulus is rotating with agular rotational speeds of 0, 0.5 and 1 rad /s.

# 3. RESULTS AND DISCUSSION

Changes in the structure of the phase change material by heat transfer was expected. Since the temperature applied to the liquid zone is above the melting temperature of FDM, the solid state structure stores the latent heat by changing the phase. By adding the rotation rates to the liquid region where the temperature is applied, it is aimed to accelerate the phase change and the following data are obtained.

In the case that the rotational speed is 0 rad/s, the phase change and temperature differences in the structure of the material are shown in Figure 2 and Figure 3.

The curved line in the above figures shows the interface between the solid and the liquid.



a.



**Figure 2.** Time-dependent variations of temperature distribution in the flow region and solid region when the rotation speed is 0 rad/s (a.100 s, b.500 s, c.1000 s, d.2000 s)



**Figure 3.** Time-dependent streamline distribution of phase change material when the rotation speed is 0 rad/s (a.100 s, b.500 s, c.1000 s, d.2000 s)

At the same temperature values, when the rotating speed of 0.5 rad / s is given to the inner hot cylinder, the contours in Figure 4 and Figure 5 are obtained;







**Figure 4.** Time-dependent isotherm distribution of phase change material when the rotation speed is 0,5 rad/s (a.100 s, b.500 s, c.1000 s, d.1500 s, e.2000 s)





**Figure 5.** Time-dependent streamline distribution of phase change material when the rotation speed is 0,5 rad/s (a.100 s, b.500 s, c.1000 s, d.1500 s, e.2000 s)

Finally, at the same temperature values, the rotational speed of the inner cylinder is 1 rad / s to give Figure 6 and Figure 7;



**Figure 7.** Time-dependent isotherm distribution of phase change material when the rotation speed is 1 rad/s (a.100 s, b.500 s, c.1000 s, d.1500 s, e.2000 s)



**Figure 8.** Time-dependent streamline distribution of phase change material when the rotation speed is 1 rad/s (a.100 s, b.500 s, c.1000 s, d.1500 s, e.2000 s)

All of the above analyzes were made for phase change material with a melting temperature of 505 K. With the effect of the rotation force to phase change material, the temperature contours and velocity vectors in the liquid area were obtained The sloped line between the solid and liquid surfaces shows the mushy zone. By applying a temperature above the melting temperature of the phase change material to the inner cylinder, the heat transfer to the phase change material causes melting the solid state phase change material. In the beginning, all regions have heat transfer by conduction. Then it starts to show the effect of natural convection by melting the phase change material. Initially the melting rate is very fast but it starts to slow down over time. The phase change material receives heat from the heat transfer fluid and stores it at the start of the melting.

It is seen that ther are significant changes in the flow field and isotehrm countours with the influence of rotataion.

Table 2 shows the comparison of the Nusselt Numbers obtained after 3 different rotational speeds (0 rad/s, 0.5 rad/s and 1 rad/s);

Table 2. Comparison of Nusselt Numbers

| Rotation Speed (rad/s) | Time (s) | Nusselt Number (W/m <sup>2</sup> .K) |
|------------------------|----------|--------------------------------------|
| 0                      | 2000     | 5.5738                               |
| 0.5                    | 2000     | 5.5914                               |
| 1                      | 2000     | 5.5248                               |

## 4. CONCLUSION

In this study, the numerical study of the melting process for the Phase Change Material was carried out without applying the rotational force and applying the rotational force. Numerical studies were two-dimensional, time-dependent and were implemented using the COMSOL commercial software package. The analyses results show that melting rate is faster at the uppermost section due to buoyant effect and effects of rotation on the fluid flow and heat transfer rate are significant.

#### REFERENCES

1.Cano, D., Funéz, C., Rodriguez, L., Valverde, J.L. and Sanchez-Silva, L. (2016)., "Experimental investigation of a thermal storage system using phase change materials", Applied Thermal Engineering, 107: 264-270.

2.Tay, N.H.S., Bruno, F. and Belusko, M. (2013). "Comparison of pinned and finned tubes in a phase change thermal energy storage system using CFD", Applied Energy, 104: 79-86.

3.Huang, MJ., Eames, PC., Hewitt, NJ., (2006). "The application of a validated numerical model to predict the energy conservation potential of using phase change materials in the fabric of a Building", Solar energy Materials&Solar Cells 90, s.1951-1960.

4.Neeper, D.A., (2000). "Thermal dynamics of wallboard with latent heat storage", Solar Energy 68 (5) s. 393-403.



# Measurement of Natural Background Outdoor Gamma Dose Rate in Karabük, Turkey

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Abstract: The external exposure of human beings indoors and outdoors to ionizing radiation occurs mainly from natural background radiation that emitted by primordial radionuclides in the earth's crust, as well as cosmogenic radionuclides, that are produced by the interactions of cosmic ray particles incident on the earth's atmosphere with the nuclei of atmospheric constituents. In this study, outdoor absorbed gamma dose rate (OAGDR) was measured at a height of 1 m in 40 different points in industrial Karabük city center using dose rate meter. The outdoor annual effective radiation dose (OAERD) and excessive life time cancer risks (LTCR) due to external exposure were estimated for adults. The OAGDR values varied from 71 to 244 nG/h with an average of 152 nGy/h. The values of OAERD and LTCR estimated for adults were found in the range of 88 to 299  $\mu$ Sv/y with an average of 186  $\mu$ Sv/y and 3.5 x 10-4 to 1.2 x 10-3 with an average of 1000  $\mu$ Sv/y.

Keywords: Natural radioactivity, Gamma dose rate, Annual effective dose, Lifetime cancer risk.

## **1. INTRODUCTION**

The outdoor gamma dose rate resulting from terrestrial (or primordial) and cosmogenic radionuclides can be an important source of external radiation. Therefore, the measurement of gamma dose rate in outdoor air is very important to assess the external radiation exposure and the risk to the public individual for both natural and artificial radiation (UNSCEAR, 1988, 2000).

There are two types of radiation sources as natural and artificial. The largest contribution to natural radiation come from cosmic and terrestrial radionuclides which emit ionizing radiation Such as alpha-, beta- and gamma-ray (Rafique et al., 2014). The exposure to cosmic rays depends on altitude and solar activity while terrestrial radiation comes from primordial nuclides present in the earth's crust whose distribution depends on geological characteristics of locations (Hazrati et al., 2010).

Since certain levels of outdoor gamma dose rates pose a potential threat to human health, it is necessary to determine and monitor the level of outdoor gamma dose rate especially in densely populated areas. (Yeşilkanat et al., 2015). This is significantly helpful for both determining public dose rates to examine the effect of radiation on health as well as it can be used as a reference for establishing regulatory control actions and standards for radiation protection (Quindos et al., 1994; Tzortzis et al., 2003).

In this study, outdoor absorbed gamma dose rates at a height of 1 m in 40 points in Karabük city center were measured by the dose rate measuring device. The outdoor annual effective radiation dose (OAERD) and excessive life time cancer risks (LTCR) due to external exposure were estimated for adults.

# 2. MATERIALS AND METHODS

When the beam of a gamma ray photon arrives at any material, it is removed separately in a single event. This phenomenon can be a real absorption process, in which case the photon disappears or the photon is emitted from the beam (Knoll 2000).

The ability of the radiation or any particle to generate an electrical signal depends on the production of charged secondary particles. The gamma ray must interact with a detector to appear. The charged particles produce a signal inside the detector crystal by ionizing and stimulating atoms in the material in which the detector is made (Gilmore, 2008; Karabıdak, 2010).

Gamma dose rate measurement was performed by the dose rate measuring device (NEB.211L) produced in the Çekmece Nuclear Research and Training Center. The measuring range of the device with the Geiger-Müller detector is 5  $\mu$ R/h to 150 mR/h. The measurements were repeated 5 times at the same point. The mean value of each point was given as the average of the measured values in  $\mu$ R/h at 5 points.

Gamma dose rate results from earth and cosmic radiation measured at  $\mu$ R/h at different points were multiplied by 8.8 times to convert to nGy/h.

## 3. RESULTS AND DISCUSSION

The gamma dose rate results measured for points were given in Table 1 and comparison of the gamma dose rate with each other is shown in Graph 1.

| Sample     | Outdoor   | Sample | Outdoor   | Sample     | Outdoor   | Sample | Outdoor   |
|------------|-----------|--------|-----------|------------|-----------|--------|-----------|
| code       | gamma     | code   | gamma     | code       | gamma     | code   | gamma     |
|            | dose rate |        | dose rate |            | dose rate |        | dose rate |
|            | (nGy/h)   |        | (nGy/h)   |            | (nGy/h)   |        | (nGy/h)   |
| <b>S1</b>  | 182.7     | S11    | 243.6     | S21        | 127.02    | S31    | 127.02    |
| S2         | 147.9     | S12    | 200.1     | S22        | 169.65    | S32    | 169.65    |
| <b>S</b> 3 | 105.27    | S13    | 71.34     | S23        | 137.46    | S33    | 137.46    |
| <b>S4</b>  | 122.67    | S14    | 174.87    | S24        | 121.8     | S34    | 163.56    |
| <b>S</b> 5 | 108.75    | S15    | 182.7     | S25        | 208.8     | S35    | 209.67    |
| <b>S6</b>  | 182.7     | S16    | 174.0     | S26        | 187.92    | S36    | 174.87    |
| <b>S7</b>  | 121.8     | S17    | 156.6     | S27        | 82.65     | S37    | 122.67    |
| <b>S8</b>  | 156.6     | S18    | 166.17    | S28        | 138.33    | S38    | 114.84    |
| <b>S9</b>  | 217.5     | S19    | 200.97    | S29        | 181.83    | S39    | 81.78     |
| S10        | 194.88    | S20    | 107.88    | <b>S30</b> | 105.27    | S40    | 147.03    |

Table 1. Measured outdoor gamma dose rates.

Graph 1. Comparison of gamma dose rate values



In outdoor, the annual effective radiation dose (OAERD) received by the individuals exposed to the gamma radiation is estimated by accepting the effective dose coefficient 0.7 Sv/Gy from the air-absorbed gamma-ray dose. OAERD was estimated using the formula given below, considering that 20% of the time of individuals was spent in outdoor.

$$OAERD = OAGDR. 0.7.8766.0.2. 10^{-3}$$
(1)

where the OAGDR is the gamma dose rate given in Table 1. The results of OAERD caused by radionuclides and cosmic radiation in the earth's crust are shown in Graph 2.

Graph 2. Comparison of OAERD values



Lifetime cancer risk (LTCR) is considered to be a value that indicates the maximum number of cases of cancer in individuals exposed to a certain level of ionizing radiation. The lifetime risk of cancer was calculated by the relation given below (Attc1, 2016):

LTCR = OAERD.ML.RF

where OAERD is the annual effective radiation dose, ML is the mean life expectancy (accepted as 70 years) and RF value is a risk factor of is 0.05/Sv (ICRP, 1990). Comparisons of estimated LTCR values were shown in Graph 3.

Graph 3. Comparison of LTCR values



Comparison of measured gamma dose rate values with the values in the literature is given in Table 2. As can be seen from the Table 2, the gamma dose rate measured in Karabük province is larger than the others except Artvin province.

**Table 2.** Measured gamma dose rate, across in different cities of Turkey and comparison with gamma dose rates measured in different countries.

(2)

| Country  | Gamma<br>dose rate<br>absorbed | Country               | Gamma<br>dose rate<br>absorbed |
|----------|--------------------------------|-----------------------|--------------------------------|
|          | in air                         |                       | in air                         |
| Malaysia | 92                             | Turkey<br>(Çanakkale) | 55                             |
| China    | 62                             | Turkey<br>(Artvin)    | 175                            |
| Poland   | 45                             | Turkey<br>(Adana)     | 65                             |
| Bulgaria | 70                             | Turkey<br>(Yalova)    | 80                             |
| Romania  | 59                             | Turkey<br>(Çankırı)   | 84                             |
| Greece   | 56                             | Turkey<br>(Karabük)   | 152                            |
| Albania  | 71                             |                       |                                |

## 4. CONCLUSION

The gamma dose rate values measured in the forty different points of Karabük and originating from the terrestrial and cosmogenic radionuclides varied between 71 nGy/h and 244 nGy/h with an average of 152 nGy/h. Annual effective dose of radiation estimated varied from 88  $\mu$ Sv/y to 299  $\mu$ Sv/y with an average of 186  $\mu$ Sv/y. The average annual effective radiation dose value is approximately five times lower than the recommended maximum value of 1000 Sv/y. The lifetime risk of cancer varied between 3.5 x 10-4 to 1.2 x 10-3 with an average of 7.4 x 10-4.

This study can be used as a basis for future research and the data obtained in this study may be useful for natural radioactivity mapping. Furthermore, the results can be used as reference data to monitor the possible contamination of possible radioactivity in the future.

### REFERENCES

- Atıcı, E. (2016). Kapadokya Bölgesinde Yapı Taşı Olarak Üretilen Tüflerdeki Doğal Radyoaktivitenin Gama Spektrometrik Yöntemle Ölçülmesi. Yayınlanmış Yüksek Lisans Tezi, Nevşehir Hacı Bektaş Veli Üniversitesi Fen Bilimleri Enstitüsü. Nevşehir.
- Gilmore, G.R. (2008). Practical Gamma-Ray Spectrometry. New York: John Wiley and Sons.
- Hazrati, S., Sadeghi, H., Amani, M., Alizadeh, B., Fakhimi, H., Rahimzadeh, S., (2010). Assessment of gamma dose rate in indoor environments in selected districts of Ardabil Province, Northwestern Iran. Int. J. Occup. Hyg. 2, 42-45.
- Karabıdak, S. M. (2010). X ve Gama Işını Dedektörlerinde Ölü Zaman Düzeltilmesi İçin Modeller. Doktora Tezi, Karadeniz Teknik Üniversitesi Fen Bilimleri Enstitüsü. Trabzon.
- Knoll, G.F. (2000). Radiation Detection and Measurement. New York: John Wiley and Sons.
- Quindos, L., P. Fernandez, J. Soto, C. Rodenas and J. Gomez (1994). "Natural radioactivity in Spanish soils." Health physics 66(2): 194-200.
- Rafique, M., Rahman, S.U., Basharat, M., Aziz, W., Ahmad, I., Lone, K.A., Ahmad, K., (2014). Evaluation of excess life time cancer risk from gamma dose rates in Jhelum valley. J. Radiat. Res. Appl. Sci. 7, 29-35.
- Tchorz-Trzeciakiewicz, D. E., & Solecki, A. T. (2018). Variations of radon concentration in the atmosphere. Gamma dose rate. Atmospheric Environment, 174, 54–65.
- Tzortzis, M., H. Tsertos, S. Christofides and G. Christodoulides (2003). "Gamma-ray measurements of naturally occurring radioactive samples from Cyprus characteristic geological rocks." Radiation Measurements 37(3): 221-229.
- UNSCEAR, (1988). Sources Effects and Risks of Ionizing Radiation Annex a: Exposures from Natural Sources of Radiation. United Nations Scientific Committee on the Effects of Atomic Radiation United Nations, New York.
- UNSCEAR, (2000). Sources Effects and Risks of Ionizing Radiation: Annex B: Exposures from Natural Radiation Sources. United Nations Scientific Committee on the Effects of Atomic Radiation United Nations, New York.
- Velasco, H. (2019). Modifications in the gamma dose rate in air due to downward and lateral mobility of 137Cs in the soil. Journal of Environmental Radioactivity, 198, 159–164.

Yeşilkanat, C. M., Kobya, Y., Taşkin, H., & Çevik, U. (2015). Dose rate estimates and spatial interpolation maps of outdoor gamma dose rate with geostatistical methods; A case study from Artvin, Turkey. Journal of Environmental Radioactivity, 150, 132– 144.



# Theoretical Studies on The Corrosion Inhibition of Stainless Steel by Organic Inhibitors in Phosphate Industries in 15% H2SO4 (2- aminomethyl phenol) & 2-(aminomethyl) benzenethiol

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**Abstract:** Corrosion is a destructive attack of the metal through its interaction with the environment, the best way to combat it is to prevent it. Metals and their alloys are exposed to acids in industries, and the acids play a very important part in the corrosion of metals, those acids could be chemical, petrochemical reagents and Industrial acids.

This is a theoretical study, conducted on two compounds, (2- aminomethyl phenol) & 2-(aminomethyl benzenethiol, to determine which is the best for use as an inhibitor to reduce the corrosion process of the stainless steel used for the manufacture of phosphate in the presence of sulfuric acid 15%.

In our theoretical study, the calculations have been carried out at the B3LYP level of theory using Gaussian-09 series of program package, which were based on 6-311G (d,p) basis set. Through the results obtained, the presence of a number of electrons in the process of interaction with the surface of the metals to reduce the reduction of erosion.

In combination, aminoethyl phenol and aminoethyl benzenethiol can be inhibitors but only aminoethyl benzenethiol

The best, because of the abundant number of electrons, which helps to form a layer insulated on the surface of metals.

Keywords: DFT, 2- aminomethyl phenol, 2- aminomethyl benzenethiol, Homo, Lumo.

# **1. INTRODUCTION**

Corrosion is a destructive attack of the metal through its interaction with the environment. It is responsible for many losses primarily in the industrial field. Clearly, the best way to combat it is to prevent it. Among the different ways to avoid or prevent the destruction or deterioration of the metal surface, corrosion inhibitor is one of the best-known methods of corrosion protection and one of the most useful in the industry[1]. This method is to continue standing because of low cost and method of practice. The use of corrosion inhibitors is the most economical and practical means of controlling metal corrosion in corrosive media. Interest in the use of inorganic compounds as friendly as corrosion inhibitors has been extended to the use of inorganic compounds to prevent metal corrosion. This phenomenon requires constant research on better corrosion inhibitors, due to the vast differences in media encountered in industry that remain a focal point in corrosion control as inhibitors slow down corrosion on metals. Corrosion is one of the major concerns in the durability of materials and structures; as a result, studies are constantly being conducted to develop an effective means to combat corrosion. Corrosion can be seen as a global phenomenon[2]. The serious consequences of corrosion have become a global problem. In addition to our daily encounters with this type of degradation, corrosion causes plant downtime, waste of valuable resources, product loss or contamination, reduced efficiency, expensive maintenance or costly additional designs; it also threatens safety and hinders technological progress. Corrosion interferes with human safety, disrupts industrial processes and poses a danger to the environment. Corrosion awareness and adaptation to appropriate and timely control measures holds the key in reducing corrosion failure. Definitions: Corrosion is the degradation or destruction of metals and alloys in an environment by chemical or electrochemical means. In simple terms, corrosion processes involve interactions with environmental minerals.[3]

stainless-steel corrosion consists of two types; General corrosion (or uniform corrosion) which the uniform loss of metal over an entire surface. Stainless steel with a pH value of less than 1 are more prone to general corrosion, Galvanic

corrosion (or bimetallic corrosion) which defined an electrochemical process where one metal corrodes preferentially compared to another in the presence of electrolyte. An example of this is when stainless steel comes into contact with rain, causing it to corrode.

There are two types of corrosion inhibitors, Organic inhibitors These materials are characterized by high molecular weight structures, incorporating nitrogen or phosphorous groups. They are usually highly polar molecules and Inorganic inhibitors, Salts of some metals and amphoteric elements act as corrosion inhibitors.[4] Quite often these materials have tenacious film forming or passivation effects. In some instances, they react with the metal surface.

This study, conducted on two compounds, (2- aminomethyl phenol) & 2-(aminomethyl benzenethiol, to determine which is the best for use as an inhibitor to reduce the corrosion process of the stainless steel used for the manufacture of phosphate in the presence of sulfuric acid 15%. In our theoretical study, the calculations have been carried out at the B3LYP level of theory using Gaussian-09 series of program package, which were based on 6-311G (d,p) basis set.

# 2. MATERIALS AND METHODS

## **Theoretical Study**

(computational chemistry methods used gauss 05) All different calculations will be performed with of complete geometry optimization by using the standard Gaussian 5.0 software package.

These calculations have been carried out at the B3LYP level of theory using Gaussian-0 9series of program package [5]. The calculations were based on 6-311G (d,p) basis set. The method which has been widely implemented to study the relationship between inhibitory activation efficiency of the molecules and their electronic properties [6]. In order to set up correlation between experimental activity and structural and electronic characteristics of the investigated inhibitors, the geometry of the molecules was optimized by the density functional theory (DFT)], with the Beck's three parameter exchange functional [5] along with the Lee– Yang–Parr correlation functional (B3LYP). The optimized structure of the investigated inhibitors.

The quantitative structure activity relationship (QSAR) is a mathematical demonstration of biological activity in terms of structural descriptors of a series of homologue molecules [7,8,9]. The objective of QSAR is to explore new molecules with required properties using chemical intuition and experience changed into a mathematically quantified and computerized form. Formerly a correlation is recognized, the structure of any number of compounds with preferred properties can be predicted. Therefore, QSAR methodology saves resources and accelerate the process of development of new molecules and drugs [8]. Success of QSAR in the development of new drug molecules and prediction of toxicity of molecules is highly significant [7,8,9]. Quantum chemical descriptors have been extensively used in QSAR studies in biochemistry. Recently the uses of quantum chemical descriptors. In particular, net atomic charges, HOMO–LUMO energies, frontier orbital electron densities and super diagonalizabilities have been used to correlate with various biological activities [10].



In this theoretical study All calculation was performed with Gaussian 05 program package, Density functional theory (DFT) method at the B3LYP functional with:

6-311G (d,p), methods basis sets .

## To determine the quantum chemical :- .

- 1) dipole -moment (D)
- 2) EHOMO, ELUMO, HOMO-LUMO energy gap
- 3) absolute electronegativity ( $\chi$ ),
- 4) absolute hardness ( $\eta$ ),
- 5) softness ( $\sigma$ ),
- 6) proton affinity (PA),
- 7) electrophilicity ( $\omega$ )
- 8) nucleophilicity ( $\epsilon$ )
- 9) Atomic coordinates of optimized molecule.
- 10) Optimized parameters: atomic distances and angles.
- 11) Mulliken atomic charges.

Quantum chemical calculations were carried out using the standard Gaussian 05 software package .

The molecular optimization of the studied tetrazoles was achieved using the functional hydride B3LYP density functional theory (DFT) formalism, having an electron basis set : B3LYP/6-311G (d,p)

basis sets will be performed on two compounds for all atoms .

The quantum chemical parameters were calculated for molecules in the neutral as well as protonated form for comparison. It is well known that the phenomenon of electrochemical corrosion occurs in the liquid phase. Quantum chemical parameters such as the ELUMO (energy of the highest Occu-pied molecular orbital), EHOMO (energy of the lowest unoccupied molecular orbital).

 $\Delta E = EHOMO - ELUMO$  (energy band gap).

# Theoretical Study for the Two types of an Organic Inhibitors:

2-(amino methyl) phenol

2- (aminomethyl)benzenethiol



Figure 1.

# 3. RESULTS AND DISCUSSION

## Results

The use of quantum chemical calculations is very important in studying the correlation between molecular structure and corrosion inhibition efficiency. The Quantum chemical methods and molecular modeling techniques are widely considered to characterize the molecules in terms of reactivity shape and binding properties. Quantum chemical parameters like ionization energy, electron affinity, highest occupied molecular orbital energy ( $E_{HOMO}$ ) and lowest unoccupied molecular orbital energy ( $E_{LUMO}$ ), the energy gap ( $\Delta E$ ), dipole moment(DM), Electronegativity( $\chi$ ), chemical potential ( $\mu$ ), Global electrophilicity index( $\omega$ ), global molecular nucleophilicity ( $\epsilon$ ), Mullikan Atomic(TNC), Nucleofugality, Electrofugality, polarizability, Hyperpolarizability Absolute hardness ( $\eta$ ) and global softness (S). The highest occupied molecular orbital (EHOMO) and the lowest unoccupied molecular orbital (ELUMO) are very useful to elucidate the chemical reactivity of a molecule. are very useful and provide important hints to predict theoretically the inhibition efficiencies of molecules. (Table 1).

| Mol. no  | номо    | LUMO    | eV      | номо    | LUMO    | E_G    | Hardness |
|----------|---------|---------|---------|---------|---------|--------|----------|
| 1-AP-gp  | -0.2206 | -0.0032 | 27.2116 | -6.0037 | -0.0865 | 5.9172 | 2.9586   |
| 1-AP-p   | -0.3667 | -0.1783 | 27.2116 | -9.9774 | -4.8529 | 5.1245 | 2.5622   |
| 1-AP-aq  | -0.2274 | -0.0102 | 27.2116 | -6.1882 | -0.2773 | 5.9109 | 2.9555   |
| 1-AP-aqp | -0.2526 | -0.0387 | 27.2116 | -6.8734 | -1.0517 | 5.8216 | 2.9108   |
| 2-BZ-gp  | -0.2218 | -0.0099 | 27.2116 | -6.0342 | -0.2683 | 5.7659 | 2.8829   |
| -2-ВZ-р  | -0.3576 | -0.1821 | 27.2116 | -9.7317 | -4.9552 | 4.7765 | 2.3882   |
| 2-BZ-aq  | -0.2278 | -0.0157 | 27.2116 | -6.1985 | -0.4261 | 5.7724 | 2.8862   |
| 2-BZ-aqp | -0.2569 | -0.0481 | 27.2116 | -6.9909 | -1.3089 | 5.6821 | 2.8410   |

| Mol. no  | Softness | Electronegativity | Chemical potential | Electrophilicity<br>index | Nucleofugality | Electrfugality |
|----------|----------|-------------------|--------------------|---------------------------|----------------|----------------|
| 1-AP-gp  | 0.1690   | -10.6040          | 10.6040            | 166.3371                  | 629.4273       | 86.4671        |
| 1-AP-p   | 0.1951   | -8.6171           | 8.6171             | 95.1289                   | 316.2192       | 46.9675        |
| 1-AP-aq  | 0.1692   | -10.5117          | 10.5117            | 163.2827                  | 618.2539       | 84.3737        |
| 1-AP-aqp | 0.1718   | -10.1691          | 10.1691            | 150.5054                  | 563.4959       | 76.6751        |
| 2-BZ-gp  | 0.1734   | -10.5887          | 10.5887            | 161.6185                  | 592.7929       | 85.5929        |
| -2-BZ-p  | 0.2094   | -8.7400           | 8.7400             | 91.2145                   | 277.3668       | 48.1759        |
| 2-BZ-aq  | 0.1732   | -10.5065          | 10.5065            | 159.2998                  | 586.0721       | 83.8001        |
| 2-BZ-aqp | 0.1760   | -10.1103          | 10.1103            | 145.2033                  | 528.4473       | 75.0639        |



Fig.2 Optimized geometries, the HOMO, the LUMO and the total electron density of each compound for the neutral molecules.



#### Discussion

The higher the EHOMO value, the stronger the electron donating capability of the inhibitor will be and, therefore, the better the observed inhibition efficiency will be. The LUMO implies the capability of the molecules to accept electrons from the metallic surface. The lower the value of ELUMO, the more it will be prone towards accepting electrons., it can be concluded that the capability of donating electrons follows the order: ELUMO signifies the propensity of molecules to accept electrons. The lower value of the ELUMO is the stronger is the ability of molecules to accept electrons.

The gap energy between the frontier orbitals ( $\Delta E = ELUMO - EHOMO$ ), is usually of great importance in describing the static molecular reactivity. Indeed, large values of the energy gap.

### 4. CONCLUSION

The highest values of EHOMO indicate better tendency towards the donation of electron to the other molecule the lowest value of LUMO energy indicates better tendency accepting the electrons to the metal surface.

A molecule with a small frontier orbital gap is generally associated with a high chemical reactivity, low kinetic stability.

The lower the values of the (DE = EHOMO-ELUMO) energy gap the better will be the inhibition efficiency since the energy required to remove an electron from the last occupied orbital will be minimum.

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#### REFERENCES

- [1]. Corrosion Science, Vol. 38, No. 11, pp. 2019-2029, 1996.
- [2]. Corrosion Science, Vol. 43, pp. 1615-1626, 2000.
- [3] . Foad El Sherbini, E. E. Mater Chem Phys 1999, 60, 286.
- [4]. Singh RK\* and Kumar R, Corrosion Protection of Stainless Steel by Organic H2SO4, Singh and Kumar, J Powder Metall Min 2014,3:2
   Inhibitors in Phosphate Industries in 15%
- [5]. Frisch M. J., Trucks G. W. and Schlegel H. B. et al. (2003). Gaussian 03 Rev. Gaussian, Inc.: Pittsburgh PA.
- [6].Young D. C. (2001). A practical guide for applying techniques to realworld problems in Computational Chemistry (New York: JohnWiley & Sons Inc.) 630.
- [7]. H., Katzenellenbogn J. A., Garg R. and Hansch C. (1999). Comparative QSAR analysis of estrogen receptor ligands. Chem. Rev 99(3):723-744.
- [8]. Gupta S. P. (1991). QSAR (quantitative structure-activity relationship) studies on local anesthetics. Chem. Rev. 91(6):1109-1119.
- [9]. Gupta S. P., Singh P. and Bindal M. C. (1983). QSAR studies on hallucinogens. Chem. Rev. 83: 633-649.
- [10]. Karelson M., Lobanov V. S. and Katritzky A. R. (1996). Quantum-Chemical Descriptors in QSAR/QSPR Studies. Chem. Rev. 96(3):1027-1044.



# Evaluation of Internal Exposure due to Radon Concentration in Soil Samples from Karabük Province

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Abstract: Internal exposure arises from the intake of natural radionuclides by inhalation and ingestion. Inhalation exposure is mainly caused by alpha and beta radiation from the radioactive radon gas (a daughter product of <sup>226</sup>Ra in the <sup>238</sup>U series), which emanates from rocks and soils and tends to concentrate in enclosed spaces like houses, and its short-lived decay products (<sup>218</sup>Po, <sup>214</sup>Pb, <sup>214</sup>Bi, and <sup>214</sup>Po). In this study, the radon concentrations in the outdoor air resulting from the topsoil samples collected from 39 different locations in the Karabük industrial city were determined based on the activity concentration of <sup>226</sup>Ra contained in the soil samples. For this purpose, the activity concentration of <sup>226</sup>Ra and radon emanation coefficients of the soil samples were measured by gamma-ray spectrometry with the measurement of porosity and true density of soil samples. The radon concentration in outdoor air varied from 31 to 140 Bq/m<sup>3</sup> with an average of 71 Bq/m<sup>3</sup>. Annual effective dose due to inhalation of radon estimated for adults varied from 291 to 1330  $\mu$ Sv/y with an average of 974  $\mu$ Sv/y. The average value of annual effective dose is lower than the recommended limit of 1000  $\mu$ Sv/y.

Keywords: Radon, internal exposure, soil samples, Karabük

## **1. INTRODUCTION**

<sup>238</sup>U and <sup>232</sup>Th are present in earth's crust in varying amounts. These two elements are the parent elements of the "Uranium" and "Thorium" radioactive decay series. The radioactive gas radon is a decay product of radium, which is a radioactive product of uranium. Both uranium and radium are present in most soils and rocks and therefore radon is ubiquitous in air. Radon has a half-life of approx. 3.8 days. Radon is the greatest source of public exposure to radiation. Nearly half of the radiation dose received by the general population originates from radon. Radon gas enters the indoors from different sources, such as soil or rock adjacent to the house, earth-based building materials, water supplies, and natural gas. Radon and its daughters can cause a significant health hazard when present in elevated concentrations in the indoor air. They enter the human body through inhalation. The alpha particles emitted from the inhaled <sup>222</sup>Rn has high damaging potential to the lung tissue and are considered to be a causative agent for lung cancer in human beings (WHO, 1988; Ferlay et al., 2007; Tomášek, 1999; UNSCEAR, 2000). The scope of this paper is (a) to examine for the first time the radon internal exposure properties of soil samples from Karabük Province, (b) to estimate the annual effective dose due to inhalation of radon, (c) to compare this dose with the recommended limit value.

# 2. MATERIAL AND METHODS

For radioactivity analyses of soils sampling sites in uncultivated locations, away from buildings and trees, were chosen from the Karabük region. Thirty nine surface soil samples were collected randomly from locations (Figure 1).



Figure 1. Sampling sites of Karabük region

After clearing the ground surface of stones, pebbles, vegetation... 1 kg of the sample from the first 15 cm of soil was placed in labeled polyethylene bags and then transferred to Laboratory. The samples were ground, homogenized and sieved to about 100 mesh by a crushing machine. The samples were then dried at 110 <sup>o</sup>C for 24 h to ensure that moisture was completely removed. About 160 g of each sample was sealed in gas-tight, radon impermeable, plastic cylindrical polyethylene containers. To reach secular equilibrium, each sample was sealed for 1 month. Measurement of the radioactivity (<sup>238</sup>U) in the samples was conducted with a coaxial HPGe detector of 50% relative efficiency and a resolution of 1.9 keV at the 1332 keV gamma of <sup>60</sup>Co (ORTEC, GEM50P4-83 model). The quality assurance of the measurements was carried out by periodical efficiency and energy calibrations and repeating sample measurements. The sample counting time was 50 000 s so as to obtain gamma-spectra with good statistics.

## 3. RESULTS AND DISCUSSION

The activity concentrations and the statistical uncertainty of  $^{226}$ Ra in the samples are presented in Table 1. The activity concentrations of  $^{226}$ Ra ranged from  $13 \pm 2$  to  $59 \pm 9$  Bqkg<sup>-1</sup>. The lowest  $^{226}$ Ra activity concentration was measured in sample number 7 and the highest  $^{226}$ Ra activity concentration was measured in sample number 39. The United Nations Scientific Committee on the Effects of Atomic Radiations (UNSCEAR) reported that the average activity concentration of  $^{226}$ Ra in the Earth's crust was 32 Bqkg<sup>-1</sup> (UNSCEAR, 2008). Table 1 compares the activity concentrations of  $^{226}$ Ra in the samples with the average of the Earth's crust. This shows that the activity concentration of  $^{226}$ Ra measured in the soil samples is nearly the same with the average of the Earth's crust.

The radon concentration in soil gas (Cs) in the absence of radon transport is given as follows (UNSCEAR, 2000):

$$C_s(\operatorname{Bq} \mathrm{m}^{-3}) = \frac{A_{Ra} \cdot f \cdot \rho \cdot (1-p)}{p}$$
(1)

where  $A_{Ra}$  is the activity concentration of <sup>226</sup>Ra measured for the soil samples, f is the emanation factor (0.3), r is the true density of the soil, and p is the total porosity.

| Table 1. | <sup>226</sup> Ra | activity | concentrations, | radon | concentrations | and | annual | effective | dose | due t | o inhalation | of r | adon | in |
|----------|-------------------|----------|-----------------|-------|----------------|-----|--------|-----------|------|-------|--------------|------|------|----|
| Karabük  | soil sa           | mples    |                 |       |                |     |        |           |      |       |              |      |      |    |

|             | Activity concentration         | Radon concentration           | AERD        |
|-------------|--------------------------------|-------------------------------|-------------|
| Sample      | ( <b>Bq kg</b> <sup>-1</sup> ) | ( <b>Bq m</b> <sup>-3</sup> ) | (mSv/y)     |
| code        | <sup>226</sup> Ra              | Air                           | EInhalation |
| S1          | 30±4                           | 71                            | 674         |
| S2          | 21±3                           | 51                            | 479         |
| S3          | 31±4                           | 74                            | 698         |
| S4          | 24±4                           | 58                            | 549         |
| S5          | 15±2                           | 36                            | 337         |
| S6          | 22±3                           | 53                            | 498         |
| <b>S</b> 7  | 13±2                           | 31                            | 291         |
| S8          | 15±2                           | 36                            | 338         |
| S9          | 25±4                           | 61                            | 575         |
| S10         | 15±2                           | 35                            | 335         |
| S11         | 47±7                           | 112                           | 1056        |
| S12         | 41±6                           | 97                            | 916         |
| S13         | 38±6                           | 90                            | 855         |
| S14         | 32±5                           | 76                            | 722         |
| S15         | 27±4                           | 65                            | 613         |
| S16         | 35±5                           | 83                            | 789         |
| S17         | 34±5                           | 82                            | 772         |
| S18         | 33±5                           | 79                            | 749         |
| S19         | 27±4                           | 64                            | 609         |
| S20         | 26±4                           | 62                            | 585         |
| S21         | <u>39±6</u>                    | 94                            | 886         |
| S22         | 34±5                           | 82                            | 776         |
| S23         | <u>30±4</u>                    | 72                            | 679         |
| S24         | 28±4                           | 67                            | 632         |
| S25         | 42±6                           | 101                           | 956         |
| S26         | 44±6                           | 105                           | 997         |
| \$27        | 29±4                           | 68                            | 647         |
| \$28        | <u>33±5</u>                    | 79                            | 744         |
| \$29        | 30±4                           | 72                            | 679         |
| \$30        | 18±3                           | 43                            | 406         |
| <u>\$31</u> | 17±2                           | 41                            | 386         |
| <u>\$32</u> | 30±4                           | 72                            | 683         |
| \$33        | 19±3                           | 45                            | 424         |
| <u>\$34</u> | 31±5                           | 126                           | 698         |
| \$35        | 57±8                           | 136                           | 1287        |
| S36         |                                | 60                            | 5/3         |
| <u>S3/</u>  | 20±4                           | 62                            | 592         |
| 538         | 20±3                           | 48                            | 454         |
| 839         | 39±9                           | 140                           | 1550        |
| Average     | 30                             | /1                            | 0/4         |
| SD          |                                | 25                            | 241         |
| Min         | 13                             | 31                            | 1220        |
| Max         | 59                             | 140                           | 1330        |

The average value of  $\rho$  was estimated to be 2340 kg m<sup>-3</sup> and the average value of p was estimated to be 0.45. The concentration of the radon in air (Ca) was estimated by the following equation (Elnimr et al., 2017):

$$C_a \left( \text{Bq m}^{-3} \right) = C_s \sqrt{\frac{d_s}{D_a}} \tag{2}$$

where Cs is the concentration of <sup>222</sup>Rn in the soil given in Equation (1), ds is the diffusion rate constant of <sup>222</sup>Rn in the soil ( $0.5 \pm 10-4 \text{ m}^2 \text{ s}^{-1}$ ), and Da is the diffusion rate constant of <sup>222</sup>Rn in the air ( $5 \text{ m}^2 \text{ s}^{-1}$ ). The activity concentrations of the radon in air are given in the third column of Table 1. The values of Ca ranged from and 31 to 140 Bq m<sup>-3</sup> with an average of 71 Bq m<sup>-3</sup>. The average concentration of radon in air is lower than the reference level of 100 Bq m<sup>-3</sup> recommended by the World Health Organization (2009).

The annual effective dose rate ( $E_{INH}$ ) coming from inhalation of radon gas was estimated taking into account the equilibrium factor (0.6 for outdoors), the conversion factor for radon (9 nSv h<sup>-1</sup>/Bq m<sup>-3</sup>), and the outdoor occupancy (0.2), implying that 20% of time is spent outdoors (UNSCEAR, 2008).

$$E_{\rm INH} \,(\mu {\rm Svy^{-1}}) = C_{\rm a} \ge 0.6 \ge 9 \ge 8766 \ge 0.2 \ge 10^{-3} \tag{3}$$

where Ca is the concentration of radon in air given in Equation (2) (Altıkulac et al., 2016; Karataslı et al., 2016). The values of  $E_{INH}$  estimated for the soil samples are presented in the last column of Table 1. The average value of  $E_{INH}$  was found to be 674 mSv y<sup>-1</sup> (range: 291 to 1330 mSv y<sup>-1</sup>).

#### 4. CONCLUSION

The activity concentrations of the natural radionuclide in the soil samples collected from the Karabük region in Turkey were measured. The average activity concentration of <sup>226</sup>Ra determined in the soil samples is nearly same with the world average, The annual effective dose rate from inhalation of radon does not exceed the recommended values. The results show that there is no significant radiological hazard for the human population living in the area.

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#### REFERENCES

- Altıkulac A, Turhan S & Gümüş H (2016). Activity concentration of terrestrial and anthropogenic radionuclides (<sup>226</sup>Ra, <sup>222</sup>Rn, <sup>232</sup>Th, <sup>40</sup>K, and <sup>137</sup>Cs) in soil samples. Environmental Earth Sciences, 75(41): 1–8.
- Elnimr M A, Turhan S, Khalid M M, Ali Madee Y G, Gala H, Kurnaz A, & Hançerlioğulları A (2017). Radiological impact assessment of nuclear weapon depots in Valley Rwagh, Libya. Environmental Forensics, 18(3): 207–213.
- Ferlay J, Autier P, Boniol M, Heanue M, Colombet M & Boyle P (2007). Estimates of the cancer incidence and mortality in Europe in 2006. Ann. Oncol. 18: 581–592.
- Karataslı M, Turhan S, Varinlioglu A & Yegingil Z (2016). Natural and fallout radioactivity levels and radiation hazard evaluation in soil samples. Environmental Earth Sciences, 75(5): 1–9.
- Tomášek L (1999). Risks from Radon Inhalation. In: Baumstark-Khan C., Kozubek S., Horneck G. (eds) Fundamentals for the Assessment of Risks from Environmental Radiation. NATO Science Series (Series 2. Environmental Security), vol 55. Springer, Dordrecht.
- UNSCEAR (2000). United Nations Scientific Committee on the Effects of AtomicRadiations Sources, Effects and Risks of Ionizing Radiation, paragraph 99, United Nations, New York.
- UNSCEAR (2008). United Nations Scientific Committee on the Effects of Atomic Radiations Sources and Effects of Ionising Radiation, United Nations Scientific Committee on the Effects of Atomic Radiation, United Nations Publication, New York, USA.
- WHO (1988). World Health Organization, International Agency for Research on Cancer, IARC Monographs on Evaluation of Carcinogenic Risks to humans, monograph No 43, Man-made Mineral Fibers and Radon.
- WHO (2009). World Health Organization. Handbook on Indoor Radon. A Public Health Perspective. Geneva, Switzerland: World Health Organization.



# Assessment of Excessive Lifetime Cancer Risk due to Natural Radioactivity (Terrestrial and Cosmic) in the Sepiolite Quarries

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Abstract: Human beings have been exposed to ionizing radiation emitted from natural radioactive sources. In this study, the outdoor absorbed gamma dose rates in air at a height of 1 m above the ground in sepiolite quarries were measured using dose rate meter. Annual effective radiation dose (AERD) and excessive life time cancer risks (ELCR) due to external exposure were estimated for sepiolite quarry workers. The average AERD and ELCR value estimated for workers in Beylikova, Polatlı and Sivrihisar quarries were found as 120  $\mu$ Sv/y and 4.8 × 10<sup>-4</sup>, 132  $\mu$ Sv/y and 5.3 × 10<sup>-4</sup> and 130  $\mu$ Sv/y and 5.2 × 10<sup>-4</sup>, respectively. The average AERD value was approximately 8 times lower than the recommended limit of 1000  $\mu$ Sv/y.

Keywords: Natural radioactivity, annual effective radiation dose, lifetime cancer risk, sepiolite

## **1. INTRODUCTION**

People are inevitably exposed to ionizing radiation everywhere as long as they live. The ionizing radiation source can be divided into two categories: natural and artificial radioactive sources. Approximately 80% of annual effective radiation dose exposed to an adult person (3 mSv) comes from natural radioactive sources and 20% from medical applications (UNSCEAR 2008). Natural radioactive sources consist of terrestrial or primordial originating from the earth's crust radionuclides which are in the radioactive series of uranium (U-238), thorium (Th-232) and actinium (U-235), and radioactive potassium (K-40) and cosmogenic radionuclides (H-3, Be-7, C-14, etc.) (UNSCEAR 2008). While the activity concentrations of terrestrial radionuclides vary from region to region, cosmic radiations vary depending on the altitude (UNSCEAR 2008).

Recently, it has been of great interest to evaluate the ionizing radiation level and its impact on the environment (Karataşlı et al. 2016; Altıkulaç et al. 2016; Aborisade et al. 2017; Gören et al. 2017; Łokas et al. 2017; Turhan et al. 2018). This is due to the negative effects of ionizing radiation on biological tissues. When high energetic ionizing radiation interacts with biological tissues, it is then released to cause ionization. Charged particles and free radicals can thus damage cell structure change and deoxyribonucleic acid (DNA).

In this study, the annual effective radiation dose and excessive lifetime cancer risk caused by the natural radioactivity were estimated on the basis of the measured outdoor gamma dose rates for workers in three sepiolite quarries located in Ankara and Eskişehir provinces in Central Anatolia region in Turkey.

## 2. MATERIALS AND METHODS

Sepiolite, which is a magnesium hydrosilicate ( $Si_{12}O_{30}Mg_8(OH)_4(H_2O)_4\cdot 8H_2O$ ), is a naturally occurring clay mineral of sedimentary origin. Due to its absorption (or adsorption), rheological and catalytic properties depending on physicochemical properties such as porosity, surface area, fibrous structure, crystal morphology and composition, sepiolite have been widely used as additive raw material in pharmaceutical, cleaning-detergent, paper, paint, cosmetic agriculture, fertilizer, livestock, ceramics and cement industry. In this study, the annual effective radiation dose and excessive lifetime cancer risk were estimated on the basis of the outdoor gamma dose rates measured in air at a height of 1 m above the ground at the 30 different points (S) in sepiolite quarries using dose rate meter for adults working in open sepiolite quarries of Polath (Ankara), Beylikova (Eskişehir) and Sirvihisar (Eskişehir) located in Central Anatolia region of Turkey.

The annual effective radiation dose due to external exposure was estimated from outdoor external gamma radiation dose rate taking into account the conversion factor for adults (0.7 Sv/Gy) and the outdoor occupancy (0.2) implying that 20% of time is spent outdoors. The annual effective dose rate (AERD) was estimated using the following formula (UNSCEAR 2008):

AERD (
$$\mu$$
Sv/y) = OGDR  $\cdot 0.7 \cdot 8766 \cdot 0.2 \cdot 10^{-3}$  (1)

where OGDR is the measured outdoor gamma absorbed gamma dose rate taken from the study by Hançerlioğulları et al. (2019).

The possibility of cancer risk caused by the external exposure to natural radiation in the studied sepiolite quarries for mine workers can be assessed using the excess lifetime cancer risk. The excess lifetime cancer risk (ELCR) was estimated based on the AERD given in Eq.1 using the equation (Karataşlı et al. 2016):

## $ELCR = AERD \cdot DL \cdot RF$

where DL is the average human life duration of 70 years and RF is risk factor given to be 0.057/Sv for stochastic effects produced low background radiation (Qureshi et al. 2016).

(2)

# 3. RESULTS AND DISCUSSION

#### **Results**

The annual effective radiation dose and excessive lifetime cancer risk values are given in Table 1. It can be seen from Table 1 that the AERD values estimated for workers in Beylikova, Polatlı and Sivrihisar sepiolite quarries varied from 57 to 202  $\mu$ Sv/y, 39 to 243  $\mu$ Sv/y and 49 to 262  $\mu$ Sv/y, respectively. Also, the ELCR values estimated for workers in Beylikova, Polatlı and Sivrihisar sepiolite quarries varied from 2.3 × 10<sup>-4</sup> to 8.1 × 10<sup>-4</sup>, 1.6 × 10<sup>-4</sup> to 9.7 × 10<sup>-4</sup> and 2.0 × 10<sup>-4</sup> to 1.0 × 10<sup>-3</sup>, respectively.

| Sample code | Quarry     | AERD (µSv/y) | ELCR                  |
|-------------|------------|--------------|-----------------------|
| S1          | Beylikova  | 182          | 7.3 x10 <sup>-4</sup> |
| S2          |            | 202          | 8.1 x10 <sup>-4</sup> |
| <b>S</b> 3  |            | 137          | 5.5 x10 <sup>-4</sup> |
| <b>S4</b>   |            | 170          | 6.8 x10 <sup>-4</sup> |
| S5          |            | 88           | 3.5 x10 <sup>-4</sup> |
| <b>S6</b>   |            | 117          | 4.7 x10 <sup>-4</sup> |
| S7          |            | 78           | 3.1 x10 <sup>-4</sup> |
| <b>S8</b>   |            | 94           | 3.8 x10 <sup>-4</sup> |
| S9          |            | 80           | 3.2 x10 <sup>-4</sup> |
| S10         |            | 57           | 2.3 x10 <sup>-4</sup> |
| S11         | Polatlı    | 200          | 8.0 x10 <sup>-4</sup> |
| S12         |            | 184          | 7.3 x10 <sup>-4</sup> |
| S13         |            | 172          | 6.9 x10 <sup>-4</sup> |
| S14         |            | 133          | 5.3 x10 <sup>-4</sup> |
| S15         |            | 70           | 2.8 x10 <sup>-4</sup> |
| S16         |            | 243          | 9.7 x10 <sup>-4</sup> |
| S17         |            | 127          | 5.1 x10 <sup>-4</sup> |
| S18         |            | 82           | 3.3 x10 <sup>-4</sup> |
| S19         |            | 70           | 2.8 x10 <sup>-4</sup> |
| S20         |            | 39           | 1.6 x10 <sup>-4</sup> |
| S21         | Sivrihisar | 221          | 8.8 x10 <sup>-4</sup> |
| S22         |            | 188          | 7.5 x10 <sup>-4</sup> |
| S23         |            | 96           | 3.8 x10 <sup>-4</sup> |

Table 1. The AERD and ELCR values

| Table 1 continued |            |     |                       |
|-------------------|------------|-----|-----------------------|
|                   | <b>S24</b> | 125 | 5.0 x10 <sup>-4</sup> |
|                   | S25        | 94  | 3.8 x10 <sup>-4</sup> |
|                   | S26        | 74  | 2.9 x10 <sup>-4</sup> |
|                   | S27        | 49  | 2.0 x10 <sup>-4</sup> |
|                   | S28        | 119 | 4.7 x10 <sup>-4</sup> |
|                   | S29        | 74  | 2.9 x10 <sup>-4</sup> |
|                   | <b>S30</b> | 262 | 1.0 x10 <sup>-3</sup> |

## Discussion

The average value of AERD due to terrestrial and cosmic radiation was found as 120  $\mu$ Sv/y, 132  $\mu$ Sv/y and 130  $\mu$ Sv/y for workers in Beylikova, Polath and Sivrihisar sepiolite quarries, respectively. The AERD average values are significantly lower than the average value of 456  $\mu$ Sv/y (total of external terrestrial and cosmic exposure) (UNSCEAR 2008). The average value of ELCR was found as  $4.8 \times 10^{-4}$ ,  $5.3 \times 10^{-4}$  and  $5.2 \times 10^{-4}$  for workers in Beylikova, Polath and Sivrihisar sepiolite quarries, respectively. The ELCR average values are lower than the world's average of  $1.45 \times 10^{-3}$  (Qureshi et al. 2016).

### 4. CONCLUSION

The results obtained from the study reveals that sepiolite quarries do not pose any health hazards from the radiological point of view.

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#### REFERENCES

- Aborisade M A, Gbadebo A M, Adedeji O H, Okeyode I C & and Ajayi O A (2017). Excess lifetime Cancer risk and Radiation Pollution hazard indices in rocks and soil of some selected mining sites in Nasarawa State, Nigeria. Aegean Journal of Environmental Sciences (AEJES), 3: 1-18.
- Altıkulaç A, Turhan Ş & Gümüş H (2016). Activity concentration of terrestrial and anthropogenic radionuclides (<sup>226</sup>Ra, <sup>222</sup>Rn, 232Th, <sup>40</sup>K, and <sup>137</sup>Cs) in soil samples. Environmental Earth Sciences, 75-41: 1-8.
- Hançerlioğulları A, Ali Madee Y G, Kurnaz A & Turhan Ş (2019). Radiometric properties of sepiolite minerals from quarries in Central Anatolia of Turkey. Nuclear Technology and Radiation Protection, 34(2): 149-156.
- Gören E, Turhan Ş, Kurnaz A, Garad AMK, Duran C, Uğur FA & Yeğingil Z (2017). Environmental evaluation of natural radioactivity in soil near a lignite-burning power plant in Turkey. Applied Radiation and Isotopes, 129:13-18.
- Karataşlı M, Turhan Ş, Varinlioğlu A & Yeğingil Z (2016). Natural and fallout radioactivity levels and radiation hazard evaluation in soil samples. Environmental Earth Sciences, 75-424: 1-9.
- Łokas E, Zwoliński Z, Rachlewicz G, Gąsiorek M, Wilkosz G & Samolej K (2017). Distribution of anthropogenic and naturally occurring radionuclides in soils and lakes of Central Spitsbergen (Arctic). Journal of Radioanalytical and Nuclear Chemistry, 311: 707-717.
- Qureshi A A, Tariq S, Din K U, Manzoor S, Calligaris C & Waheed A (2014). Evaluation of excessive lifetime cancer risk due to natural radioactivity in the rivers sediments of Northern Pakistan. Journal of Radiation Research and Applied Sciences 7(4): 438-447.
- Turhan Ş, Gören E, Uğur F A, Karataşlı M & Yeğingil Z (2018). Study of natural and artificial radioactivity in environmental soil samples from Eastern Anatolia Region of Turkey. Radiochimica Acta, 106: 161-168.
- UNSCEAR (2008). Sources and effects of ionizing radiation. United Nations Scientific Committee on the Effects of Atomic Radiation, United Nations Publication, (2010), New York, USA.



# The Effect of Austempering Temperature on Wear Behavior in Ggg40 Cast Irons

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**Abstract:** In this study, the effects of austempering temperature heat treatment on microstructure, hardness and abrasion wear of austempered ductile cast irons GGG40 (DCI) were investigated. For this purpose, it was determined that austenitizing temperature was 900 °C and austenitizing time was 90 minutes for GGG40 (DCI) samples. By applying austempering temperature as 320 °C and 380 °C in two individual parameters, it was kept in salt bath for 120 minutes in order to see the effect of upper and lower bainite/ausferrite morphology.

In order to determine structural changes occurring in ductile cast irons GGG40 (DCI) after heat treatment, Optical Microscopy, SEM, EDS, and X-ray analyses were carried out after they were cleaned and polished by using traditional polishing method. Microhardness measurements were also taken from the surface of materials by using Vickers hardness scale (HV). Wear tests were carried out in wear test device displaying vertical pivoted motion with 320 and 800 mesh abrasive by applying 10, 20, 30 N load and using 8 m and 15 m road distance. Values of abrasion loss in materials were measured while examining wear surfaces of the samples with SEM microscope. Wearing surface morphology of the samples was examined by SEM-EDS and X-ray analyses.

As a result of this study; when the austempering temperature was increased to 380 °C, roughness of ferrite and high carbon austenite was observed, and residual austenite was determined to decrease. Since the austempering temperature is low, it has been observed that pinned ferrites nucleate around the spheroidal graphite in the structure. At 380 °C, the amount of ferrite decreases in samples that are austempered, which is thought to be due to the austempering temperature being high. At the lower cooling point, the amount of ferrite increases and the volume proportion decreases because of the decrease in ausferrite formation and the increase in ferrite nucleation. As the austempering temperature increases, this ratio is reversed. Hardness values increased as austempering temperature increased.

Keywords: GGG40, Austempering Temperature, Microstructure, Abrasive Wear, Microhardness.

# **1. INTRODUCTION**

Ductile cast irons (DCIs) are defined as a material group having mechanical properties of steel and the production properties of cast iron. DCIs have about double strength of gray cast iron and they can be casted very easier than steel. Moreover, the great cost advantage in its production has made its usage area more and more widespread compared to both steel and other cast irons. Nowadays, ductile cast irons are successfully used especially in the automotive industry. Depending on the development of ductile cast iron technology, ways of further improving the mechanical properties of DCI have been sought. However, in the studies, understanding that it is impossible for graphite to bring morphology other than spherical has forced the researchers to study for developing the mechanical properties through matrix modification [1].

Depending on these studies, "Austempered Ductile Cast Iron" (ADI) has been developed by applying austempering heat treatment to ductile cast irons to give a bainitic microstructure to the cast iron. Based on the austempering process, it is stated that significant increases can be achieved in mechanical properties of ductile cast irons such as strength, ductility, toughness, and wear resistance. The excellent properties of the ADI are related to the ferrite ( $\alpha$ ) and the high-carbon austenite ( $\gamma$ yk) which form the microstructure. Austempered steel has a structure containing ferrite and iron carbide. Therefore, while the product formed by austenitizing DCI is called ausferrite, the product obtained in steel is called bainite [2-7]. DCI also contains ductile graphites in its microstructure. High Si content in DCI prevents carbide precipitation during the austempering reaction and maintains the  $\gamma$ yk content of the stable high-carbon austenite [8]. Generally, sufficient hardenability of this material can be ensured by preventing the formation of perlite during cooling to the
austempering temperature by adding alloy elements like Ni, Mo and Cu in low quantities into ADI. In the conventional austempering process (it is named as single-step in this study), casting materials are left in the temperature range of 850-950 °C for a sufficient time (usually 1-2 hours) until a fully austenite ( $\gamma$ ) matrix is obtained. They are then rapidly cooled down to 250-400 °C temperature range, kept at the selected temperature for 1-4 hours and then cooled down in the air at room temperature. Figure 1. shows schematically the conventional single step austempering process.



**Figure 1.** Schematic View of the Conventional (Single Step) Austempering Process [7] The ADI heat treatment cycle is performed in a two-step process.

In Step I, austenite decomposes into ferrite ( $\alpha$ ) and high-carbon austenite ( $\gamma$ yk).

 $\gamma = \alpha + \gamma y k$  (1)

If casting material is kept at austempering temperature for a long time, Step II reaction begins and high-carbon austenite ( $\gamma yk$ ) decomposes into ferrite and carbide:

 $\gamma y k = \alpha + \epsilon \text{ (carbide)} (2)$ 

Akray observed as a result of austenitization and austempering heat treatment applied to the GGG40 quality DCI samples that the microstructure was composed of bainitic ferrite and residual austenite in different morphology and rates depending on the austempering temperature and the wear resistance increased 4.5 times compared to the casting form with austempering performed under optimum conditions [1]. Kus applied austempering heat treatment at different temperatures to improve the wear resistance of DCI with GGG40 ferritic structure. It was observed that the austempering heat treatment increased the wear resistance, tensile and yield resistance of ductile cast iron. The microstructure was significantly dependent on the austempering temperature and as the austempering temperature increased, the wear resistance decreased [9]. Çetin observed as a result of his studies that the hardness of DCIs austenitized at 900 °C and having a ferritic and perlitic structure increased depending on the increased austenite temperature and time and decreased austempering temperature (250, 320, 370 and 400 °C) and the abrasion losses decreased depending on this increase in the hardness [10]. In their study, Zhou et al., obtained a DCI bainite + martensitic structure by applying a controlled heat treatment in ferritic DCI having the chemical composition of 3.2-3.8% C, 2.5-3.0% Si, 2.5-3.0% Mn, 0.06% S, and 0.06% P [11]. In the study conducted by Ovali et al., on the fatigue behaviors of austempering temperature in GGG40 ferritic DCIs it was observed that the fatigue strengths of austempered samples at low (315 °C) and high (375 °C) austempering temperatures in different inter-critical austenitic temperatures increased with the fatigue limits of the samples with increased ausferrite volume rate [12]. In the studies conducted by Okay et al. on DCIs it was determined that the hardness of the samples increased with the austempering heat treatment and the wear resistance increased in direct proportion to the hardness of the sample in metal-metal abrasion tests [13]. In his studies, Oktay investigated the effects of austempering and induction hardening heat treatments in GGG70 class alloyed DCIs and observed that the fatigue strength of the samples increased in direct proportion to the increase in the surface hardness [14]. In their studies, Voigt et al., observed that Ductile Cast Irons with Dual Phased Matrix Structure (DPMDCI) containing martensite or ausferrite along with ferrite in the microstructure had higher total elongation compared to traditionally heat treated DCIs [15]. In their studies, Karatas et al., investigated the effect of heat treatment temperatures on the microstructure in the production of acicular

ductile GGG60 cast iron. It was concluded as a result of the studies that optimum austentizing temperature should be about 900 °C and the temperature range of 350-400 °C needs to be selected to obtain the most efficient bainitic formation without martensite formation in the formation of bainitic structure during the cooling in the samples with the highest hardness value measured as 426 HB [16]. In the studies of Hasirci it was observed that the formation of cementite occurring depending on the increase in austempering time resulted in a decrease in impact strength and elongation; whereas, it led to an increase in yield strength and also in hardness [17]. In their study, Sugiyama et al., partially austenitized ductile cast iron with 60% ferrite + 40% perlite in its microstructure at various critical temperatures (800 °C, 815 °C, 830 °C, and 840 °C) and then austemperized them. At these temperatures, ausferrite in the volume rates of 20%, 40%, 80% and 90% was obtained after partial austenitizing [18]. Kilicli observed that the microhardness of the ausferrite varied in DCIs depending on the austempering time. In his experimental studies, it was observed that the hardness decreased up to 120 min austempering time and increased after 120 min [19]. In their studies, Rashidi et al., determined that yield and tensile strength increased when the partial austenitizing time was increased to 400 seconds between critical temperatures [20]. In the studies conducted by Wen et al. using perlitic ductile cast iron, the samples were austempered for 510 seconds in salt baths at different temperatures (300 °C, 325 °C, 350 °C, 375 °C) after austenitizion at 900 °C for 60 min and cooled down to the room temperature by quenching in order to produce DPMDCI. They then obtained ausferrite +tempered martensite in the microstructure by tempering at 200 °C for 120 min [21].

In this study, the effect of austempering heat treatment performed at 320 and 380 °C for 120 min on the hardness and abrasive wear behavior in GGG40 DCI samples after austenitizing heat treatment at 900 °C for 90 min was investigated.

### 2. MATERIAL AND METHOD

#### **Preparation of Experimental Samples**

Before the process, DCI experimental samples casted commercially with 30 mm diameter and 120 mm length were machined in a CNC lathe and brought to 10mm diameter and 100 mm length to make them appropriate for the experiment (Figure 2.). While Table 1 shows the chemical properties of the used GGG40 DCI sample, Table 2 shows its physical properties and Table 3 shows its mechanical properties.

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## Figure 2. Macro Photograph Of GGG40

The samples to be used in the experiments were examined in 3 groups. These groups were;

- 1. Ductile Cast Iron GGG40 in cast state (unheat-treated),
- 2. Pieces austenitized at 900 °C and austempered for 120 min at 320 °C,
- 3. Pieces austenitized at 900 °C and austempered for 120 min at 380 °C.

**Table 1.** Chemical Composition of GGG40 Ductile Cast Iron

| %                         | С        | Si      | Mn      | S       | Р      |
|---------------------------|----------|---------|---------|---------|--------|
| GGG40 Standard            | 3.4-3.85 | 2.3-3.1 | 0.1-0.3 | 0.2 max | 0.1max |
| GGG40 Experimental Sample | 3.550    | 2.457   | 0.159   | 0.014   | 0.028  |

### Table 2. Physical Properties of GGG40 Ductile Cast Iron

| Specific weight               | 7.2g/m3                                 |
|-------------------------------|---|
| Typical microstructure        | At least 80% Ferrite                    |
| Electrical conductivity       | 20-100 °C -36,5 watt / m °C             |
| Thermal expansion coefficient | 20-100 °C -11,10 - 6 / m <sup>0</sup> C |

### Table 3. Mechanical Properties of GGG40 Ductile Cast Iron

| Tensile strength   | 400 N/mm <sup>2</sup> |
|--------------------|-----------------------|
| Yield strength     | 250 N/mm <sup>2</sup> |
| Hardness           | 160-190 HB            |
| Elasticity modulus | 165 N/mm <sup>2</sup> |
| Fatigue strength   | 220 N/mm <sup>2</sup> |

## **Heat Treatments**

It is composed of two steps as austempering heat treatment and austempering after austenitization.

### Austenitizing

For the austempering heat treatment, PID controlled electric resistance furnace without atmosphere control working with  $\pm 1$  sensitivity, 220 V and 25 A having 5kW power was used. In heat treatments, K type (Cr-CrAl) thermocouple was used. The samples were placed in a sample pan and thermocouple wires were placed in contact with the sample surface. The samples were placed in the furnace where temperature measurements were conducted throughout the experiment. The experiment was conducted at 900 °C.

### Austempering

In order to provide isothermal retention in austempering heat treatment, a salt bath with 50% KNO3 + % NaNO3 admixture was used. As a salt bath, a PID controlled salt bath working at 220V and 25A with  $\pm 1$  sensitivity and the power of 5kW was used. Figure 3 shows the schematic view of the salt bath.



Figure 3. Salt Bath in Which Austempering Heat Treatment Was Performed [11]

## **Metallographic Examinations**

Ductile cast irons subjected to heat treatment and surface hardening process and the untreated ductile cast iron samples were cut respectively in 10x10mm dimensions in order to examine the microstructures and the metallographic examinations were conducted. Conventional cleaning and polishing method was used and after sanding with 80, 120, 240, 500, 800 and 1000 mesh sandpaper, respectively, they were made ready for etching by polishing with 3 µm diamond paste. For the etching process, 2% Nital was used and etching was performed for 2 seconds. In order to view the microstructures of the samples with completed preparation processes, each sample was examined by using a Nikon MA2000 optical microscope and JEOL brand JSM-7001F scanning electron microscope.

## **Microhardness Tests**

In order to examine the microhardness of the experimental samples, they were austenitized together with the DCI samples in the casting state and then 6 hardness values were obtained from the regions of the austempered samples subjected to metallographic treatment with equal intervals. Hardness tests were carried out in EMCOTEST DuraScan brand microhardness device by applying 50 gr load at 10 sec. loading speed at 1.3 mm intervals from the center of the sample.

#### Wear Tests

The wear test was carried out by the friction of the experimental sample on a rotating disc. The wear test was realized in a Siemens wear device with AC motor having a power of 3kW (Figure 4.). The wear device has a hardened tray having a vertical rotational movement. The load having an impact on the sample and the rotational speed of the disc can be easily adjusted in the wear test. The experiments were carried out at room temperature at a fixed shear rate of 1m/s under 10, 20, 30 N load and 8 and 15 m shear distance. Two measurements were made for each sample. 320 and 800 Mesh SiC abrasives were used in the wear test. Each sample was weighed with an electronic scale with 0.0001 gr sensitivity before the wear test and then they were cleaned with alcohol and weighed again after subjecting to abrasion process. Abrasion losses were recorded.



Figure 4. Schematic View of the Wear Test Device [5].

## **Experimental Results and Discussions**

## **Results of microstructure**

Figure 5 shows Ferrite+Perlite matrix microstructures of the DCI samples used in this test. As seen in the following figures, it was observed that ferrite was formed around graphite in the internal structure of the material. In addition to these ferrite rings, perlite formation was also observed in the other sections and it was composed of a typical "calf eye" structure.



Figure 5. Microstructure of DCI (GGG40) Cast Samples (a) and (b) Pearlitic DCI

DCI samples whose microstructures were ferrite+perlite were subjected to austenitization process at 900 °C for 90 min and austempering process at 320 °C for 120 min and the effects of austempering process on hardness and microstructures of these samples were investigated. These samples were examined under an optical microscope by separating into two groups due to their austempering temperatures. The samples in the microstructure images given in Figure 6 as a result of the austempering heat treatment were austempered for 120 min at 320 °C. It was observed that the matrix in these samples was composed of ferrite, high-carbon austenite (HCA) and residual austenite (RA). With the effect of 120-min austempering in the samples, ausferrite needles were clearly seen in the microstructure [10]. The samples whose microstructure images because of the austempering heat treatment are given in Figure 7 were austempered at 380 °C for 120 min. In these samples, coarseness of ferrite and high carbon austenite in the microstructure and a decrease in the residual austenite were observed depending on the increased austempering heat. Because of the low austempering

temperature, pinned ferrites around spheroidal graphites in the structure nucleated which shows that the austenite in the structure was cooled more and the pinned like structures formed as a result of this lower cooling size [10].

It was observed that ferrite and high carbon austenite needles became coarser and the upper ausferrite structure formed in high austempering temperature. The amount of ferrite forming in this structure decreased which was thought to be caused by high austempering temperature. Due to the decreased ausferrite formation and increased ferrite nucleation at the lower cooling point, the amount of ferrite increased and the volume rate decreased. As the austempering temperature increased, this ratio was reversed [10].



Figure 6. The Samples Austenitized at 900 °C and Austempered at 320 °C for 120 Min.



Figure 7. The Samples Austenitized at 900 °C and Austempered at 380 °C for 120 Min

## **Results of Microhardness**

Figure 8 shows the average hardness values of the DCI samples austempered and DCI in casting state in this test. According to the results given in the graph, total average hardness value of DCI samples in the casting state was determined as 195.73 HV, total average hardness value of the ADI 120 minute 320 °C austempering samples was determined as 457,47 HV and total average hardness value of the ADI 120 minute 380 °C austempering samples was determined as 473,69 HV. According to these obtained data, the hardness of the ADI 320 °C austempering sample increased by 233,72% compared to the DCI in the casting state.

| М      | MİCROHARDNESS (HV) |                          |                          |  |  |
|--------|--------------------|--------------------------|--------------------------|--|--|
| 600,00 |                    | 457,47                   | 473,69                   |  |  |
| 400,00 | 195,73             |                          |                          |  |  |
| 200,00 |                    |                          |                          |  |  |
| 0,00 · |                    |                          |                          |  |  |
|        | DCI                | ADCI<br>120 MİN<br>320 C | ADCI<br>120 MİN<br>380 C |  |  |

Figure 8. Tensile Strength of DCI and ADCI

## **Investigation of the Wear Surfaces**

SEM images of the samples subjected to the wear test are present in the figures (Figure 9 and Figure 10). As is seen in SEM images, materials in the casting state underwent excessive deformation. Wear test results of DCI in casting state and ADI are given in the below table (Table 4). Mass loss of DCI material in casting state was determined with the friction coefficient between 0.021-0.029 g under 10N load,

|                              | r in r    |           |                      |           |           |           |
|------------------------------|-----------|-----------|----------------------|-----------|-----------|-----------|
|                              | Comp. No1 | Comp. No2 | Comp. No3            | Comp. No4 | Comp. No5 | Comp. No6 |
| Samples                      | 10N 320   | 10 N 800  | 20N 320              | 20N 800   | 30N 320   | 30N 800   |
|                              | Abrasive  | Abrasive  | Abrasive             | Abrasive  | Abrasive  | Abrasive  |
| Casting group                |           |           |                      |           |           |           |
| first                        | 20.497    | 20.047    | 20.051               | 20.458    | 19.917    | -         |
| measurement                  |           |           |                      |           |           |           |
| Casting group                | 20.473    | 20.026    | 20.03                | 20.431    | 19 888    | _         |
| 1 <sup>st</sup> sanding      | 20.475    | 20.020    | 20.03                | 20.451    | 17.000    |           |
| Casting group                | 20 444    | 20.002    | 20.003               | 20 403    | 19 857    | _         |
| 2 <sup>nd</sup> sanding      | 20.111    | 20.002    | 20.005               | 20.405    | 17.057    | _         |
| ADI 120 min                  |           |           |                      |           |           |           |
| 320 °C                       |           |           |                      |           |           |           |
| austempering                 | 20.211    | 20.356    | 20.14                | 20.065    | 20.366    | 20.824    |
| first                        |           |           |                      |           |           |           |
| measurement                  |           |           |                      |           |           |           |
| ADI 120 min                  |           |           |                      |           |           |           |
| 320 °C                       | 20,194    | 20.33     | 20.113               | 20.042    | 20.354    | 20.808    |
| austempering                 | -01171    | -0.00     |                      | 2010 12   | 201001    | 20.000    |
| 1 <sup>st</sup> sanding      |           |           |                      |           |           |           |
| ADI 120 min                  |           |           |                      |           |           |           |
| 320 °C                       | 20.181    | 20.316    | 20.1                 | 20.028    | 20.343    | 20.79     |
| austempering 2 <sup>nd</sup> |           |           |                      |           |           |           |
| sanding                      |           |           |                      |           |           |           |
| ADI 120 min                  |           |           |                      |           |           |           |
| 380 °C                       |           |           | <b>a</b> a <b>a-</b> |           |           |           |
| austempering                 | 21.484    | 21.461    | 20.37                | 20.467    | 20.361    | 20.822    |
| first                        |           |           |                      |           |           |           |
| measurement                  |           |           |                      |           |           |           |
| ADI 120 min                  |           |           |                      |           |           |           |
| 380 °C                       | 21.477    | 21.454    | 20.368               | 20.452    | 20.353    | 20.81     |
| austempering 1 <sup>st</sup> |           |           |                      |           |           |           |
| ADI 120 min                  |           |           |                      |           |           |           |
| ADI 120 MIN<br>380.ºC        |           |           |                      |           |           |           |
| 300°C                        | 21.47     | 21.443    | 20.357               | 20.444    | 20.343    | 20.79     |
| austempering                 |           |           |                      |           |           |           |
| 2 Sanung                     | 1         | 1         | 1                    | 1         |           | 1         |

Table 4. Abrasion Losses of the Samples Under Loads of 10, 20 And 30 N



Figure 9. Wear SEM images of DCI samples



Figure 10 Wear SEM images of ADI samples

## **EDS Analysis**

Figure 11 and Figure 12 show respectively EDS analysis graphs taken  $10\mu$ m from the wear surfaces of ADI samples austempered at  $320^{\circ}$ C for 120 min and ADI samples austempered at  $380^{\circ}$ C for 120 min as a result of the wear tests. As is understood from the EDS analysis results, it was determined whether the material used in the tests was DCI from the % of the alloy elements. As a result of EDS analysis; 21.15 % C, 0.26% Mg, 1.59 % Si and 77% Fe elements were determined in the microstructure of ADI samples austempered at 320°C for 120 min and 28.30% C, 0.22% Mg, 2.44% Si and 69.04% Fe elements were determined in ADI samples austempered at 320°C for 120 min.



| Element | % Weight | %        |
|---------|----------|----------|
|         | _        | Atomical |
| С       | 21.15    | 59.90    |
| Mg      | 0.26     | 0.34     |
| Si      | 1.59     | 1.76     |
| Fe      | 77       | 43       |
| Totals  | 100      | 100      |

Figure 11. EDS Analysis of The Wear Surface of The ADCI 320 °C-120 min.

| Element | %      | %        |
|---------|--------|----------|
|         | Weight | Atomical |
| С       | 28.30  | 63.88    |



| Mg     | 0.22  | 0.24  |
|--------|-------|-------|
| Si     | 2.44  | 2.36  |
| Fe     | 69.04 | 33.52 |
| Totals | 100   | 100   |

Figure 12. EDS Analysis of The Wear Surface of The ADCI 380 °C-120 min.

#### 4. CONCLUSION

1. Matrix in the samples austempered at 320 °C was observed to be composed of ferrite, high-carbon austenite (HCA) and residual austenite (RA).

2. When the austempering temperature was increased to 380 °C, coarsening of ferrite and high carbon austenite was observed and the residual austenite decreased. Since the austempering temperature of 320 °C was low, pinned ferrites nucleated around the spheroidal graphites in the structure.

3. Ferrite amount in the samples austempered at 380 °C decreased and this was thought to be caused by high austempering temperature. Due to the decreased ausferrite formation and increased ferrite nucleation at lower cooling point, ferrite amount increased and the volume ratio decreased. As the austempering temperature increases, this ratio was reversed.

4. When the hardness results obtained from the experimental samples were evaluated, hardness values increased depending on the increased austempering time. The hardness of the ADI 320 °C austempering sample increased by 233.72% compared to the DCI in casting state. The hardness of the ADI 380 °C austempering sample increased by 242.01% compared to the DCI in casting state.

5. Abrasion losses of the experimental samples were observed to decrease with increasing austempering temperature. It was observed that the abrasion loss reduced with increasing austempering temperature under all loads applied.

6. Under the dry sliding conditions, wear of the experimental samples in casting state was observed as surface adhesion and plastic deformation.

7. The mass losses of DCI material in the casting state were determined with the friction coefficient between 0.021-0.029 g under 10N, between 0.021-0.028 g under 20N, and between 0.029-0.031 g under 30N load.

8. ADI sample was subjected to austempering wear test for 120 min at 320 °C and the mass losses were determined with the friction coefficient between 0.013-0.016 g under 10N, between 0.013-0.027g under 20N, and between 0.011-0.012 g under 30N load.

9. ADI sample was subjected to austempering wear test for 120 min at 380 °C and the mass losses were determined with the friction coefficient between 0.007-0.011 g under 10N, between 0.002-0.015g under 20N, and between 0.08-0.010 g under 30N load.

10. As a result of EDS analysis taken 10 $\mu$ m away from the wear surfaces of the ADI 120 min 320  $^{0}$ C austempering samples as a result of wear tests, it was found that the material used was DCI and the microstructure contained 21.15% C, 0.26% Mg, 1.59% Si, and 77% Fe elements.

11. As a result of EDS analysis taken 10 $\mu$ m away from the wear surfaces of the ADI 120 min 380  $^{0}$ C austempering samples as a result of wear tests, it was found that the material used was DCI and the microstructure contained 28.30% C, 0.22% Mg, 2.44% Si and 69.04% Fe elements.

12. Consequently, it was found that the wear resistance of DCI samples increased depending on the increase in austempering temperature in austempering heat treatment after austenitization process.

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#### REFERENCES

- [1] Akray, I., "The Effect of Coatings on Wear Characteristics of Spherodial Graphite Cast Iron", Master's Thesis, Istanbul Technical University, İnstitute of Science, 2007.
- [2] Cetin, M., Gul, F., "Effect of cooling in austempering on abrasive wear behavior of Austempered spheroidal graphite cast iron", Journal of the Faculty of Engineering and Architecture of Gazi University, Vol: 21(2), 2006, 359-366,
- [3] Putatunda, S. K., Gadicherla, K., "Influence of Austenitizing Temperature on Fracture Toughness of a Low Manganese Austempered Ductile Iron (ADI) with Ferritic as Cast Structure", Materials Science and Engineering, 1999, A 268, 15–31.
- [4] Rao, P., Putatunda, S.K., "Investigations on the Fracture Toughness of Austempered Ductile Irons Austenitized at Different Temperatures, Materials Science and Engineering", 2003, A 349, 136-149.
- [5] Rao, P., Putatunda, S.K., "Investigations on the Fracture Toughness of Austempered Ductile Iron Alloyed with Chromium, Materials Science and Engineering", 2003, A 346, 254-265.
- [6] Jianghuai Yang, J., Putatunda, S.K., "Improvement in Strength and Toughness of Austempered Ductile Cast Iron by a Novel Two-Step Austempering Process", Materials and Design 25, 2004., 219–230.
- [7] Zimba, J., Simbi, D.J., Navara, E., "Austempered Ductile Iron: An Alternative Material for Earth Moving Component", Cement & Concrete Composites, 2003, 25, 643-649.
- [8] Putatunda, S.K., "Development of Austempered Ductile Cast Iron (ADI) with Simultaneous High Yield Strength and Fracture Toughness by a Novel Two-Step Austempering Process, Materials Science and Engineering", 2001, A 315, 70-80.
- [9] Kus, H., "The Wear Behaviour of Austempered Ductile Iron", Master's Thesis, Selçuk University, Graduate School of Natural and Applied Sciences, 2007.
- [10] Cetin, M., "The Improvement of Wear Behaviour of Austempered Ductile Irons", Phd Thesis, Gazi University, Institute of Science and Technology, 2005.
- [11] Zhou, R. Jiang, Y., Lu, D., Zhou, R., Li., Z., "Development and characterization of a wear resistant bainite/martenzite ductile iron by combination of alloying and a controlled cooling heat treatment", Wear, 2001, 250:529-534.
- [12] Ovalı, I. Kilicli, V. Erdogan, M., "The Effect of Austempering Temperature from intercritical austenising temperatures on Microstructure and Fatigue Strength in Austempered Ductile Cast Iron", International Iron & Steel Symposium, 02-04 April 2012, Karabuk, Turkey
- [13] Okay, F., Sert, H., Habalı, K., Gul, F., "Effect of TİN Coating by Physical Vapour Deposition On the Wear Behaviour of Austempered Ductile Iron", Journal of the Faculty of Engineering and Architecture of Gazi University, 2010, Vol: 25(4), 787-794.
- [14] Oktay, E., "The Effect of Induction Surface Hardening Process on Mechanical Properties of The Austempered Ductile Iron", Master's Thesis, Gazi University, Institute of Science, 2011.
- [15] Voigt, R.C., Eldoky, L.M., Chiou, H.S., "Francture of ductile cast irons with dual matrix structure", A.F.S, 1989, 94:645-656.
- [16] Karatas, S., Teker, T., Yilmaz, S. O., "The Effect of Heat Treatment Temperature on The Microstructure of G.G.G-60 Acicular Sphero Cast Iron", International Iron & Steel Symposium, 02-04 April 2012, Karabuk, Türkiye.
- [17] Hasirci, H., "Effects of Alloying Elements (Cu and Ni) And Autempering Time On Microstructer and Mechanical Properties in Austempered Spheroidal Graphite Cast Irons", M. Sc. Thesis, Gazi University, Institute of Science and Technology, 2000.
- [18] Sugiyama, Y., Katsutoshi, A., Matsuoka, S., "Improvement of static strength and fatigue crack propagation resistance in ductile cast iron by austempering from ( $\alpha$ + $\gamma$ ) phase region", Journal of the society of Materials science, 1991, 40:675-681.
- [19] Kilicli, V., "Effect of Partially Austenisation and Austempering Heat Treatment on Microstructures and Mechanical Properties in Ductile Cast Iron", Master's Thesis, Gazi University, Institute of Science and Technology, 2004.
- [20] Rashidi, A.M., Moshrefi-Torbati, M., "Dual matrix structure (DMS) ductile cast iron: The effect of heat treating variables on the mechanical properties", International Journal of Cast Metals Research, 2001,13:293-297.
- [21] Wen, D.C., Lei, T.S., "The mechanical properties of a low alloyed austempered ductile iron in upper ausferrite region", I.S.I.J. International, 2000, 39:493-500.



## A Study on Heavy Metal Pollution of Soil Samples from Karabük Industrial Province

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**Abstract:** It is crucial problem that urban and agricultural soils become polluted with heavy metals due to mining, manufacturing, and the use of synthetic products such as pesticides, paints, batteries, industrial waste, and land application of industrial or domestic sludge etc. Heavy metals (Cr, Mn, Fe, Ni, Cu, Cd, As etc.) also occur naturally, but rarely at toxic levels but excess heavy metal accumulation in soils is toxic to humans and other animals. In the study, the concentration of heavy metal (Ti, V, Cr, Mn, Fe, Ni, Cu, Zn, Ga, As, Zr, Nb and Pb) in topsoil samples collected around industrial Karabük province was analyzed by using energy dispersive X-ray fluorescence (EDXRF) spectroscopy. The average concentrations of Ti, V, Cr, Mn, Fe, Ni, Cu, Zn, Ga, As, Zr, Nb and Pb detected in soil samples were found as 2929, 109, 136, 1379, 33406, 68, 28, 72, 11, 18, 115, 5 and 12  $\mu$ g/g, respectively. The heavy metals pollution in soils was assessed using geo-accumulation index (Igeo) and enrichment factors (EF). The average Igeo and EF value calculated for Ni and As was found as 1.1 and 2.2 and 5.3 and 18, respectively. The Igeo and EF results indicate that the study area is moderately to significant polluted with Ni and As.

Key Words: Heavy metal, soil, geo-accumulation index, enrichment factor, EDXRF

### **1. INTRODUCTION**

In addition to the numerous benefits of technological developments, industrialization and social life to humanity, heavy metal pollution, which is one of the effects that disrupts the unwanted and ecological balance, is increasing day by day. This formation is important environmental pollution and even very small concentrations are toxic (Karataş 2004).

Each source of contamination has its own damaging effects to plants, animals, and ultimately to human health, but those that add heavy metals to soil and water are of serious concern due to their persistence in the environment and carcinogenicity to human beings. They cannot be destroyed biologically but are only transformed from one oxidation state or organic complex to another (Garbisu and Alkorta 2001; Gisbert et al. 2003). Therefore, heavy metal pollution poses a great potential threat to the environment and human health.

Heavy metals are naturally occurring elements with large atomic weight and at least 5 times greater density of water density. The toxicity of heavy metals depends on many factors such as exposure dose, exposure, age, gender, hereditary characteristics. Arsenic, cadmium, chromium, lead and mercury are among the important metals in terms of public health according to their toxicity (Tchounwou et al. 2012).

In the study, the concentrations of heavy metals in topsoil samples collected around industrial Karabük province were determined by using energy dispersive X-ray fluorescence (EDXRF) spectroscopy. The heavy metal pollution in soil samples was evaluated by calculating the ecological parameters or indicators (ge-oaccumulation index and enrichment factor).

#### 2. MATERIAL AND METHODS

Karabük is located in the Western Black Sea region in the Northwest of Turkey. Karabük has an area of 4067 km2 and surrounded by the coordinates of 4518148 N-4603891 N and 424593 E-512511 E. The first iron and steel plant were built in 1937 in Karabük called "Karabük Iron and Steel Plant". It is one of the main steel producers in Turkey and has been in operation continuously since being constructed in 1937. Eighteen topsoil (up to a depth of 5 cm) samples were collected around Karabük province. The soil samples (approximately 0.5 kg) were brought to the sample preparation laboratory and allowed to dry in laboratory for several days. The samples were dried at 110 °C until a constant weight was reached.

Pebbles, plant residues and other particles were removed, and the samples were grounded, homogenized and codded. A total of 50 g of the soil samples was pressed to obtain a pellet for X-ray fluorescence measurements.

Heavy metal concentrations of the soil samples were determined by using EDXRF spectrometric method which is reliable, fast, accurate and repeatable. EDXRF spectrometer equipped with a thick binary Pd/Co alloy anode X-ray tube (50 W, 60 kV). The EDXRF spectrometer has an HAPG polarizer to improve the sensitivity to elements in the Na–Cl range and a bandpass filter to improve the performance for element detection in K–Mn range. The EDXRF spectrometer optimizes the excitation using polarization and secondary targets. It has an autosampler for up to 12 items and software modules. The target changer with up to eight polarization and secondary targets offers many different excitation conditions, ensuring the optimal determination of all elements from K to U.

#### 3. RESULTS AND DISCUSSION

Potential heavy metal pollution of the soil samples was evaluated estimating geo-accumulation index (Igeo) and enrichment factor (EF). The Igeo is one of the ecological indicators used to assess the degree of heavy metal contamination of the area under investigation. The Igeo was calculated by the following equation (Muller, 1969):

(1)

$$\mathbf{I}_{\text{geo}} = \log_2 \left( \frac{\mathbf{C}_n}{\mathbf{F} \mathbf{C} \times \mathbf{B}_n} \right)$$

where Cn is the metal concentration of the soil sample, Bn is the average geochemical reference or background value of the Earth's crust, and FC is the factor of natural fluctuations because of possible differences in the reference values and very small anthropogenic influences, which equals 1.5 (Özkul 2016). In calculating this index, Yaroshevsky's world composition was taken as reference values (Yaroshevsky 2006). The Igeo values consist of seven categories, as shown in the second row of Table 1.

The values of Igeo estimated for each metal for the evaluation of pollution of the area investigated in Karabük province are given in Table 2.

| Value                | Contamination category (soil quality)     |
|----------------------|---|
| $I_{geo} \leq 0$     | Practically uncontaminated                |
| $0 < I_{geo} < 1$    | Uncontaminated to moderately contaminated |
| $1 < I_{geo} < 2 $   | Moderately contaminated                   |
| $2 < I_{geo} < 3$    | Moderately to heavily contaminated        |
| $3 < I_{geo} < 4$    | Heavily contaminated                      |
| $4 < I_{geo} < 5$    | Heavily to extremely contaminated         |
| $5 < I_{\text{geo}}$ | Extremely contaminated                    |

Table 1. Pollution category according to ge-oaccumulation index values

| Table 2. | Values | of ge-oa | ccumulation | index | estimated | for the | investigated are | a |
|----------|--------|----------|-------------|-------|-----------|---------|------------------|---|
|          |        | ·        |             |       |           |         |                  |   |

| Metal | I <sub>geo</sub> |         |         |  |
|-------|------------------|---------|---------|--|
|       | Minimum          | Maximum | Average |  |
| Ti    | -3.20            | -0,62   | -1,32   |  |
| V     | -7,84            | -5,01   | -6,10   |  |
| Cr    | -1,96            | 1,10    | -0,15   |  |
| Mn    | -2,72            | 2,57    | -1,10   |  |
| Fe    | -3,17            | 0,11    | -1,23   |  |
| Ni    | -0,53            | 2,68    | 1,05    |  |
| Cu    | -3,39            | -0,72   | -1,79   |  |
| Zn    | -1,57            | 1,29    | -0,11   |  |

| Ga | -4,86 | -2,77 | -3,54 |
|----|-------|-------|-------|
| As | 0,85  | 5,64  | 2,20  |
| Zr | -2,94 | -0,13 | -1,12 |
| Nb | -8,99 | -5,64 | -6,88 |
| Pb | -6,46 | -2,69 | -4,72 |
|    | ,     | ,     | ,     |

The enrichment factor (EF) was used to distinguish the anthropogenic metal source from natural process metal concentrations arising from human activities and to assess the degree of metal contamination (Li et al. 2017). The EF was estimated using the following equation based on the standardization of a measured element against a reference element (Özkul 2016):

$$E_F = \frac{\binom{\binom{C_n}{}_{Ref}}{}_{sample}}{\binom{\binom{C_n}{}_{Ref}}{}_{Background}}$$
(2)

where Cn is the concentration of any element or metal and CRef is the concentration of a reference element or metal in the examined environment. In this study, the enrichment factor was calculated in reference to Sr and Yaroshevsky's earth crust composition (Yaroshevsky 2006). The  $E_F$  values consist of five classifications; the classifications according to the  $E_F$  results are given in the third row of Table 3. The  $E_F$  values estimated for each heavy metal are given in Table 4.

Table 3. Degree of contamination of enrichment factor values

| Value             | Contamination category (soil quality)  |
|-------------------|--|
| EF < 2            | Deficiency to minimal enrichment (DME) |
| $2 \leq EF < 5$   | Moderate enrichment (ME)               |
| $5 \leq EF < 20$  | Significant enrichment (SE)            |
| $20 \leq EF < 40$ | Very high enrichment (VHE)             |
| $EF \ge 40$       | Extremely enrichment (EE)              |

\_\_\_\_

Table 4. Enrichment factor values estimated for the investigated area

| Metal |         | $E_F$   |         |                         |
|-------|---------|---------|---------|-------------------------|
| -     | Minimum | Maximum | Average | Degree of<br>enrichment |
| Ti    | 0,2     | 3,5     | 1,0     | DME                     |
| V     | 0,01    | 0,1     | 0,03    |                         |
| Cr    | 0,3     | 7,0     | 2,2     | ME                      |
| Mn    | 0,1     | 7,3     | 1,5     | DME                     |
| Fe    | 0,2     | 2,7     | 1,0     | DME                     |
| Ni    | 0,6     | 20,3    | 5,3     | SE                      |
| Cu    | 0,1     | 3,5     | 0,7     | DME                     |
| Zn    | 0,4     | 7,4     | 2,1     | ME                      |
| Ga    | 0,1     | 0,7     | 0,2     |                         |
| As    | 2,8     | 172,4   | 18      | SE                      |
| Zr    | 0,2     | 5,1     | 1,3     | DME                     |
| Nb    | 0,0     | 0,1     | 0,03    | DME                     |
| Pb    | 0,09    | 0,3     | 0,09    | DME                     |

#### 4. CONCLUSION

In the study, the heavy metals (Ti, V, Cr, Mn, Fe, Ni, Cu, Zn, Ga, As, Zr, Nb and Pb) pollution (or contamination) of soil samples from industrial Karabük province was investigated by estimating ecological parameters. The average Igeo estimated for Ni and As was found as 1.1 and 2.2. The average EF value estimated for Ni and As was found as 5.3 and 18, respectively. The Igeo and EF results indicate that the study area is moderately to significant polluted with Ni and As.

#### REFERENCES

- Karataş M., (2004). Konya Ana Tahliye Kanalında Ağır Metallerin İncelenmesi Bitki ve Topraktaki Birikiminin Tespiti. Master's thesis, Selçuk Üniversitesi, Konya, Turkiye.
- Garbisu C, Alkorta I (2001) Phytoextraction: a cost effective plant-based technology for the removal of metals from the environment. Bioresour Technol 77(3):229–236
- Gisbert C, Ros R, de Haro A, Walker DJ, Pilar Bernal M, Serrano R, Avino JN (2003) A plant genetically modified that accumulates Pb is especially promising for phytoremediation. Biochem Biophys Res Commun 303(2):440–445.
- Tchounwou, P.B., Yedjou, C.G., Patiolla, A.K., & Sutton, D.J. (2012). Heavy metals toxicity and the environment. EXS, 101, 133-164.
- Garad A. M. K., (2019) Kangal Termik Santrali Çevresindeki Yüzey Toprağındaki Ağır Metal Kirliliğinin ve Asitleşmenin Ekolojik Açıdan Değerlendirilmesi. PhD Thesis, Kastamonu University (Unpublished), Kastamonu, Turkiye.
- Muller, G. (1969) Index of geoaccumulation in sediments of the Rhine River. Geological Journal, 2, 108–118.
- Özkul, C. (2016). Heavy metal contamination in soils around the Tunçbilek thermal power plant (Kütahya, Turkey). Environmental Monitoring and Assessment 188(284), 1-12.
- Huang, X., Hu, J., Qin, F., Quan, W., Cao, R., Fan, M., & Wu, X. (2017). Heavy metal pollution and ecological assessment around the
- Jinsha coal-fired power plant (China). International Journal of Environmental Research Public Health 14-1589, 1-12
- Yaroshevsky AA (2006) Abundances of chemical elements in the earth's crust. Geochem International 44(1):48-55.
- Li HH, Chen LJ, Yu L, Guo ZB, Shan CQ, Lin JQ, Gu YG, Yang ZB, Yang YX, Shao JR, Zhu XM, Cheng Z (2017) Pollution characteristics and risk assessment of human exposure to oral bioaccessibility of heavy metals via urban street dusts from different functional areas in Chengdu, China. Sci Total Environ 586:1076-1084.



## A New Method based on Partial Transmit Sequence Selected Mapping for Peak to Average Power Ratio (PAPR) Reduction in OFDM for Optic Communications

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**Abstract:** Frequency selective damping or increased resistance to narrow band interference is one of the reasons why OFDM is preferred. The disadvantage of the high PAPR multi-carrier systems is that the transmitter's high power amplifier can be severely limited in its power efficiency. A high PAPR ratio reduces system performance by disrupting the linearity of the transmitter and restricts the working area. Appropriate signal processing techniques are required to reduce high PAPR. A variety of PAPR reduction techniques are available for OFDM systems, traditional and intuitive. Traditional techniques for reducing PAPR in literature: size trimming, trimming and filtering, block coding, tone reservation, adding tone, active star cluster expansion, selective mapping are techniques such as the Partial transmit sequence. Partial transmit sequence technique is the most emphasized PAPR reduction technique by researchers. When the PTS technique is used, there is no loss of information in the mark and there is no problem in increasing the bit energy levels. Intuitive methods such as component optimization, genetic algorithm and harmoni search have shown rapid development in recent years to reduce complex calculations used in traditional methods. In this paper the selected mapping is combined with Partial transmit sequence and we got the good result for this implementing. For increase the data we used the selective mapping and for the transmission data we used the Partial transmit sequence.

Keywords: Orthogonal Frequency Multiplexing Division, Partial transmit sequence, fading, Fast Fourier Transform.



## Improving the Properties of High-Calcium Fly Ash-Based Geopolymers with Metakaolin Addition

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Abstract: Concrete is the most common used construction material. Cement, water and aggregates are the main constituents of concrete. Cement production process has some harmful effects for the environment due to the  $CO_2$  emission during calcination of limestone. Therefore, many of the recent studies are focused on eco-friendly building materials. Geopolymer is one of these green building materials. Afşin-Elbistan (AE) thermal power plant is the most energy and fly ash producing one in Turkey. Unlike other fly ashes, AE fly ash has high CaO and SO<sub>3</sub>, low SiO<sub>2</sub>+Al<sub>2</sub>O<sub>3</sub>+Fe<sub>2</sub>O<sub>3</sub> content. Therefore, it does not suit to the fly ash standards and cannot be utilized in the cement and concrete industry. In this study, AE fly ash was used as geopolymer raw material. In order to improve the properties of AE fly ash-based geopolymer, 0%, 15%, 30% and 45% metakaolin was used. Unit weight, ultrasonic pulse velocity, bending strength and compressive strength tests were performed on the samples at 7 and 28 days. Results indicated that metakaolin incorporation increased the engineering properties of the geopolymers. In this way, at least 55% of the geopolymer raw materials can be chosen from AE fly ash. The use of AE fly ash in geopolymer production can be a different utilization field, which cannot be utilized enough and remains as environment pollutant.

Keywords: Afşin-Elbistan fly ash, geopolymer, metakaolin, strength, improvement

## **1. INTRODUCTION**

Global warming and climate change is the most serious environmental problem for the earth in the last years. Greenhouse gases are the main cause of the global warming which are carbon dioxide, methane and nitrous oxide. It is known that, 5-7% of the total  $CO_2$  emission is produced by cement industry (McLellan et al., 2011). 1 ton of cement production generates approximately 1 ton of  $CO_2$  (Naskar and Chakraborty, 2016). Therefore, these data increases the importance of green concretes without Portland cement (Junaid et al., 2015). Geopolymer concrete does not contain any Portland cement. Therefore, it is considered one of the green concretes (Reed et al., 2014).

Geopolymers, which are also named as alkali-activated binders are first developed by Davidovits (Davidovits, 1982). It can be made by using aluminosilciate source mineral raw materials by mixing with an alkaline solution for activating such as sodium silicate or sodium hydroxide (Davidovits, 1991; Duxon et al., 2007). Geopolymer concrete has two major environmental benefits. First, greenhouse gases are being reduced by the usage a binder without Portland cement and second, most of the raw materials of geopolymers are industrial by-products, which are pollutant for the environment (Naupane, 2016). As aluminosilicate raw materials, blast furnace slag, fly ash, bottom ash, silt and metakaolin can be used (Ravikumar et al., 2010; Chindaprasirt et al., 2007; Chindaprasirt et al., 2009, Lampris et al., 2009). Metakaolin is the most effective mineral material as geopolymer alumiosilicate source (Davidovits, 1991; Duxon et al., 2007).

Afşin-Elbistan thermal power plant is the most energy generating thermal power plant in Turkey. Besides it is the most fly ash generating one. 15-20% of the total fly ashes in Turkey belong to this power plant. On the other hand, it cannot be utilized in the cement and concrete industry, which is the main consumer of the coal fly ashes, due to the low sum of  $SiO_2+Fe_2O_3+Al_2O_3$ , high CaO and SO<sub>3</sub> content. Therefore, there are researches on finding new utilization fields for this fly ash (Mahyar and Erdoğan, 2015; Şahin et al., 2015).

In this study, AE fly ash was used as geopolymer raw material. In order to improve the properties of AE fly ash-based geopolymer, 0%, 15%, 30% and 45% metakaolin was used. Unit weight, ultrasonic pulse velocity, bending strength and compressive strength tests were performed on the samples at 7 and 28 days.

## 2. MATERIAL AND METHODS

## Materials

Crushed sand which has a specific gravity of 2.72 was used as aggregates. The fly ash was obtained from Afşin-Elbistan Thermal power plant, which is located in Kahramanmaraş region. The metakaolin was commercially obtained. The properties of fly ash and metakaolin are given in Table 1.

| Oxide                          | Content (wt%) |            |  |  |
|--------------------------------|---------------|------------|--|--|
|                                | FA            | Metakaolin |  |  |
| SiO <sub>2</sub>               | 22.73         | 52.64      |  |  |
| Al <sub>2</sub> O <sub>3</sub> | 9.67          | 42.16      |  |  |
| Fe <sub>2</sub> O <sub>3</sub> | 4.63          | 1.29       |  |  |
| CaO                            | 42.94         | 0.04       |  |  |
| MgO                            | 1.99          | 0.31       |  |  |
| SO <sub>3</sub>                | 9.01          | -          |  |  |
| Na <sub>2</sub> O              | 0.28          | 0.07       |  |  |
| K <sub>2</sub> O               | 0.52          | 0.46       |  |  |
| Loss on ignition (%)           | 5.21          | 0.49       |  |  |
| Property                       |               |            |  |  |
| Specific Gravity               | 2.42          | 2.53       |  |  |
| Fineness (cm <sup>2</sup> /g)  | 2200          | 10685      |  |  |

**Table 1.** Properties of fly ash and metakaolin

As activator sodium hydroxide (SH) was used. The SH was obtained as commercially in flakes form. The concentration of the solution was kept constant at 14M. Materials used in the study are shown it Fig. 1.

#### 2.2. Methods

In the study, metakaolin was used as fly ash replacement material with the amounts of 0%, 15%, 30% and 45%. The activator and aggregate amount kept constant. The mixture design is given in Table 2. The fly ash, metakaolin and aggregates were dry mixed in a laboratory type mortar mixer for 60s. After dry mixing, the activator solution was added. Besides, in order keep the consistency in a constant value, some water was added. The consistency was kept constant between 16-18mm. The fresh mixture was mixed for 180s. After mixing period, the mixture cast into 40x40x160mm molds. The samples were oven cured for 24 hours at 85 °C. After curing, the de-molded samples were kept in  $20\pm2$  °C at 65% relative humidity for 7 and 28 days.





Table 2. Mixture design

| Sample code | Fly ash | Fine aggregate | NaOH |
|-------------|---------|----------------|------|
| R           | 0       | 1350           | 220  |
| MK15        | 60      | 1350           | 220  |
| MK30        | 120     | 1350           | 220  |
| MK45        | 180     | 1350           | 220  |

After these periods, the samples were subjected to unit weight, ultrasonic pulse velocity (UPV), bending and compressive strength tests.

## 3. RESULTS AND DISCUSSION

### 3.1. Unit Weights

The unit weight results are given in Fig. 2.



#### Figure 2. Unit weight results

The unit weights are changing between 1.98-2.18g/cm<sup>3</sup>. A cement-based mortar has a unit weight of approximately 2.4g/cm<sup>3</sup>. The unit weights of geopolymers are lower. There is no significant change between the samples, but it is possible to conclude that, the geopolymer samples are lightweight compared to cement-based mortars.

#### 3.2. Ultrasonic Pulse Velocity (UPV)

The UPV results are given in Fig. 3.



Figure 3. UPV results

The UPV results are changing between 2431-3038m/s. The lowest results were obtained from the reference samples which are produced completely with fly ash. The metakaolin addition increased the UPV results. The highest results were obtained from MK45 sample. It is known that there is a strong relationship between pore structure and mechanical properties. UPV gives an idea about the pore structure of the materials. Higher UPV results indicate more denser and compact microstructure. Therefore, it is possible to conclude that metakaolin addition lead to a denser structure in the geopolymer samples.

## 3.3. Bending Strength

The results of bending strength tests are given in Fig. 4.





The reference sample has a bending strength value of 2.7MPa and 2.9MPa for 7 and 28 days respectively. Metakaolin addition increased the bending strength of the samples. The highest values were obtained from MK45 sample both in 7 and 28 days, which are 4.46MPa and 4.76MPa, respectively. The metakaolin addition increased the bending strength 165.2% and 167%, for 7 and 28 days.

#### 3.4. Compressive Strength

The compressive strength results are given in Fig. 5.





The reference results are 6.89MPa and 7.13MPa for 7 and 28 days. It can be seen from the Fig. 5 that the there is a strong increasing trend by the metakaolin addition. The highest results were obtained from MK45 sample. The values are

24.85MPa and 26.46MPa for 7 and 28 days. The increments are 360.7% and 371.1%. Metakaolin addition increased the compressive strength of the fly ash based sample over 3 times. The usability of metakaolin as geopolymer raw material is already known. Therefore, it has a very positive effect on strength results. This is related to the chemical properties of metakaolin, which is a very rich alumino-silicate source. These results indicate the possible use of the non-standard Afşin-Elbistan fly ash as a geopolymer raw material when combined with metakaolin.

#### 4. CONCLUSIONS

In this study, the use of Afşin-Elbistan fly ash with metakaolin addition as supplementary material. According to the results of the experimental studies following conclusions can be drawn;

- The unit weights of the samples are between 1.98-2.18g/cm<sup>3</sup>. There is no significant change with the metakaolin addition. However, the unit weights of the samples are lower than conventional cementitious mortars.
- The UPV results were increased with the increase of metakaolin amount. These results indicate the denser and compacter microstructure of metakaolin containing groups.
- Bending strength values of samples were increased with the increase of metakaolin addition. The highest values were obtained from MK45. 45% of metakaolin addition was increased the bending strength to 165.2% and 167% compared to reference sample in 7 and 28 days, respectively.
- Metakaolin addition significantly increased the compressive strength of the sample. The highest values were obtained from MK45. The values are close to 25MPa. An increment of 360.7% and 371.1% for 7 and 28 days, respectively.
- The strength results slightly increased by age, but the increment is not significant in most cases.

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#### REFERENCES

- Chindaprasirt P, Chareerat T, Sirivivatnanon V (2007). Workability and strength of coarse high calcium fly ash geopolymer. Cem. Concr. Compos., 29(3): 224-229.
- Chindaprasirt P, Jaturapitakkul C, Chalee W, Rattanasak U (2009). Comparative study on the characteristics of fly ash and bottom ash geopolymers. Waste Manage., 29: 539-543.
- Davidovits J (1982). Mineral polymers and methods of making them. USA patent: 4349386.
- Davidovits J (1991). Geopolymer: inorganic polymeric new materials. J. Therm. Anal, 37: 1633-1656.
- Duxon P, Mallicoat S W, Lukey G C, Kriven W M, Van Deventer J S J (2007). The effect of alkali and Si/Al ratio on the development of mechanical properties of metakaolin-based geopolymers. Colloid. Surf. A, 292: 8-20.
- Junaid M T, Kayali O, Khennane J. (2015). A mix design procedure for low calcium alkali activated fly ash-based concrete. Construction and Building Materials, 79: 301-310.
- Lampris C, Lupo R, Cheeseman C R (2009). Geopolymerisation of silt generated from construction and demolition waste washing plants. Waste Manage., 29: 368-373.
- Mahyar M, Erdoğan S. T (2015). Phosphate-activated high-calcium fly ash acid-based cement. Cem. Concr. Compos., 63: 96-103.
- McLellan B C, Williams R P, Lay J, Van Riessen A, Corder C D (2011). Costs and carbon emissions for geopolymer pastes in comparison to ordinary Portland cement. J. Cleaner Prod., 19(9): 1080-1090.
- Naskar S, Chakraborty A K (2016). Effect of nano materials in geopolymer concrete. Perspectives in Science, 8: 273-275.
- Naupane K (2016). Fly ash and GGBFS based powder-activated geopolymer binders: a viable sustainable alternative of Portland cement in concrete industry. Mech. Mater., 103: 110-122.
- Ravikumar D, Peethamparan S, Neithalateh N (2010). Structure and strength of NaOH activated concretes containing fly ash or GGBFS as the sole binder. Cem. Concr. Compos., 32(6): 399-410.
- Reed M, Lokuge W, Karunasena W (2014). Fibre-reinforced geopolymer concrete with ambient curing for in situ applications. J. Mater. Sci, 49 (12): 4297-4304.
- Şahin M, Mahyar M, Erdoğan S. T (2015). Afşin Elbistan uçucu külü ve yüksek firin cürufu içeren çimentosuz bağlayıcılar hazırlanması. In: Proceedings of the 9th National Concrete Congress, 16-18 April, Antalya, pp. 181-190.



# Hyperconjugative and Physical Properties of Substituted 3-(4-(1,3,5-triazin-2-yl)-phenyl)-2aminopropanoic Acids as Novel Tryptophan Hydroxylase Inhibitors

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**Abstract:** In this study, DFT/B3LYP level of theory with 6-311G ++(2d,2p) basis sets and RHF level with 6-311G ++(2d, 2p) basis sets were used to calculate a set of molecular descriptors for two compounds substituted in 3-(4-(1,3,5) triazin-2-yl) phenyl),2-amino propionic acids, 2-amino-3-[4-[4-(1-naphthalen-2-ylmethylamino)-1,3,5-triazin-2-yl]phenyl]propanoic acid (compound 1)and 2-amino-3-[4-[4-(1-naphthalen-2-ylethylamino)-1,3,5-triazin-2-yl]phenyl] propanoic acid (compound 2).We aimed in this study to investigate the leading factors controlling the magnitude of hyperconjugative effects, such as HOMO–LUMO energy gap ( $\Delta E$ ) hardness ( $\eta$ ), softness (S), chemical potential ( $\mu$ ), electronegativity ( $\chi$ ), polarizability ( $\alpha$ ), molar volume (Vm), and hyperpolarizability ( $\beta$ )] using DFT/B3LYP level of theory with 6-311G++(2d,2p) basis set and compared the results of both title compounds with the results from using and Restricted Hartree-Fock (RHF) as another level with the same basis sets 6-311G++(2d,2p).

Keywords: TPH1inhiitors, Hyper conjugative, polarizability, hyperpolarizability, molar volume.

### **1. INTRODUCTION**

Serotonin (5-hydroxytryptamine, 5-HT) is an important material produced in the modulation of behavior and responsible on gastrointestinal (GI) function. Tryptophane hydroxylase type I (TPH1) is the rate-limiting enzyme of serotonin synthesis in the GI tract. TPH1 expressed highly in the gland of pineal and the outer circumference. It controls the synthesis of more than 90% of 5-HT synthesis in the periphery (Ouyang L et al 2012). Haihong Jin and co-workes (2009) discovered the substituted triazines as a novel class of inhibitors of tryptophan hydroxylase."This class of TPH inhibitors can selectively reduce serotonin levels in murine intestine after oral administration without affecting levels in the brain" (Haihong et al 2009). In this paper, we reported a piece of theoretical study of 2-amino-3-[4-{4-(1-naphthalen-2ylmethylamino)-1,3,5-triazin-2yl}]phenyl propanoic acid (copound 1) and 2-amino-3-[4-{4-(1-naphthalen-2ylethylamino)-1,3,5-triazin-2-yl}phenyl]propanoic acid (copound 2) (Haihong et al 2009) Figure 1 and 2 respectivily.



Figure 1. 2-amino-3-[4-[4-(1-naphthale- 2ylmethylamino)-1,3,5-triazin-2-yl]phenyl]propanoic acid { Compound 1}



Figure 2. 2-amino-3-[4-[4-(1-naphthale-2ylethylamino)-1,3,5-triazin-2-yl]phenyl]propanoic acid { Compound 2}

We focused on establishing and exploring the leading factors controlled the magnitude of hyperconjugative intramolecular interaction, such as HOMO–LUMO energy gap ( $\Delta E$ ) hardness ( $\eta$ ), softness (S), chemical potential( $\mu$ ), electronegativity( $\chi$ ), polarizability ( $\alpha$ ), molar volume (V<sub>m</sub>), and the first hyperpolarizability ( $\beta$ ) using the B3LYP [DFT] with 6-311G++(2d,2p) basis set. The results were compared with the results from using RHF as another level with the same basis set 6-311G++(2d, 2p).

#### 2. MATERIAL AND METHODS

The two title molecules of Fig 2.3 have been fully optimized at the B3LYP and RHF levels by using 6-31G++ (2d, 2p) basis sets. The HOMO-LUMO energy gap( $\Delta E$ ), hardness ( $\eta$ ), softness (S), chemical potential ( $\mu$ ), electronegativity ( $\chi$ ), molar volume (mv), the polarizability ( $\alpha$ ), and the first hyperpolarizability ( $\beta$ ) of the two title compounds have been computed at B3LYP and RHF levels.

The EHOMO and ELUMO are used to calculate the electronegativity ( $\chi$ ) and hardness ( $\eta$ ) by the following equations::

$$\chi = \frac{E_{(LUMO)} + E_{(HOMO)}}{2} \tag{1}$$

$$\eta = \frac{E_{(LUMO)}^2 - E_{(HOMO)}}{2}$$
(2)

The chemical potential( $\mu$ ) which known as the negative of electronegativity ( $\chi$ ), Softness (S), electrophilicity index ( $\omega$ ), can be calculated

$$\mu = -\frac{E_{(LUMO)} + E_{(HOMO)}}{2} \tag{3}$$

The softness (S), electrophilicity index ( $\omega$ ), nucleofugality  $\Delta E_n$ , and electrofugality  $\Delta E_e$  can be calculated from hardness ( $\eta$ ) and The chemical potential ( $\mu$ ) by following equations:

$$S = \frac{1}{2\eta} = \frac{1}{E_{(LUMO)} - E_{(HOMO)}}$$
(4)

Global electrophilicity index ( $\omega$ ) defined as

$$\omega = \frac{\mu^2}{2\eta} \tag{5}$$

$$\Delta E_{n} = \frac{(\mu + \eta)^{2}}{2\eta} \tag{6}$$

$$\Delta E_{e} = \frac{(\mu - \eta)^{2}}{2\eta}$$
 (Prasad O et al 2011) (7)

The polarizability ( $\alpha$ ) and anisotropic polarizability ( $\Delta \alpha$ ), and hyperpolarizability ( $\beta$ ) are calculated based on the following equations (Hatua K, Nandi PK 2013)

$$\alpha = \frac{\alpha_{xx} + \alpha_{yy} + \alpha_{zz}}{3} \tag{8}$$

Anisotropic polarizability ( $\Delta \alpha$ )

$$\Delta \alpha = \sqrt{\frac{(\alpha_{xx} - \alpha_{yy})^2 + (\alpha_{yy} - \alpha_{zz})^2 + (\alpha_{zz} - \alpha_{xx})^2 + \alpha_{xy}^2 + \alpha_{xz}^2 + \alpha_{yz}^2}{2}}$$
(9)

where  $\alpha_{xx}$ ,  $\alpha_{yy}$ ,  $\alpha_{zz}$ ,  $\alpha_{xy}$ ,  $\alpha_{xz}$  and  $\alpha_{yz}$  are polarizability tensor. The first hyperpolarizability ( $\beta$ )

$$\beta = \sqrt{\beta_x^2 + \beta_y^2 + \beta_z^2} \tag{10}$$

#### **3. RESULTS AND DISCUSSION**

Geometries of the two compounds were fully optimized by using Lee-Yang-Parr correlation functional B3LYP and RHF level with the 6-311G++(2d, 2p) basis set.

# Comparsion of the Nature of Hyperconjugative Interactions for the Title Compounds using the Two Different Levels B3LYP/6-311G++(2d, 2p) and RHF/6-311G++(2d, 2p)

The optimized molecular structure for the compounds 1,2 with the numbering scheme of the atoms are shown in Figure 4,5. The hyperconjugative interactions of the compounds 1, 2 using two different levels B3LYP/6-311G++(2d, 2p) and RHF/6-311G++(2d, 2p). are illustrated in Table 1.2 respectively.



Figure 4. Structure of the compound 1.



Figure 5. Structure of the compound 2.

Table 1. Hyper-conjugated interaction energies of compound

| Donor (i) | Туре | Acceptor (j) | Туре | (E)kcal/mol DFT C1 | (E) kcal/mol RHF C1 |
|-----------|------|--------------|------|--------------------|---------------------|
| N32- H33  | σ    | C36- C37     | π*   | 0.86               | 1.20                |
| N32- H33  | σ    | C38- C41     | π*   | 0.92               | 2.47                |
| C34- H35  | σ    | C38- C41     | π*   | 0.61               | 5.66                |
| C34- H53  | σ    | C38- C41     | π*   | 1.57               | 9.44                |
| C34- H53  | σ    | C46- C49     | π*   | 5.34               | 21.74               |
| C37- H40  | σ    | C38- C41     | π*   | 0.94               | 6.01                |
| LP1N29    |      | C25-N27      | π*   | 51.15              | 78.66               |
| LP1N32    |      | N26-C28      | π*   | 58.46              | 88.25               |

**Table 2.** Hyper-conjugated interaction energies of compound 2.

| Donor (i) | Туре | Acceptor (j) | Туре | (E)kcal/mol DFT C1 | (E)kcal/mol RHF C1 |
|-----------|------|--------------|------|--------------------|--------------------|
| N7- H8    | σ    | C1- N2       | π*   | 2.68               | 36.10              |
| N7- H8    | σ    | C4- N5       | π*   | 6.45               | 102.60             |
| C45-H49   | σ    | C1-N2        | π*   | 36.21              | 19.99              |
| C45-H49   | σ    | C4-N5        | π*   | 51.76              | 111.99             |
| C45-H49   | σ    | C43-C47      | π*   | 2.69               | 10.81              |
| C50-H54   | σ    | C4-N5        | π*   | 5.72               | 30.93              |
| C53-H56   | σ    | C43-C47      | π*   | 19.63              | 1579.06            |
| LP2O29    |      | C1-N2        | π*   | 0.81               | 145.61             |
| LP2O29    |      | C4-N5        | π*   | 6.24               | 693.67             |
| LP1N32    |      | C1- N2       | π*   | 128.13             | 264.31             |

As it is seen in Table 1, we investigated that the use of B3LYP/6-311G++(2d, 2p) results less intramolecular hyperconjugated interaction energies compared to the use of RHF/6-311G++(2d, 2p). For example, the intramolecular hyperconjugated interaction  $\sigma(N32-H3) \rightarrow \pi^*(C36-C37)$  was recorded 0.86 kcal mol<sup>-1</sup> by use of B3LYP/6-311G++(2d, 2p). This value increased to record 1.20 kcal mol<sup>-1</sup> by use RHF/6-311G++(2d, 2p). The intramolecular hyperconjugated interaction  $\sigma(C34-CH53) \rightarrow \pi^*(C46-C39)$  was recorded 5.34 kcal mol<sup>-1</sup> by use of B3LYP/6-311G++(2d, 2p) level. This value increased to record 21.49 kcal mol<sup>-1</sup> by use RHF/6-311G++(2d, 2p).level. The Lon pair intramolecular hyperconjugated interaction illustrated an enormous increase by use RHF/6-311G++(2d, 2p).level compared with B3LYP/6-311G++(2d,2p)

As instant, the intramolecular hyperconjugated interaction  $\sigma(\text{LP1N29}) \rightarrow \pi^*(\text{C25-N27})$  was recorded 51.15 kcal mol<sup>-1</sup> by use of B3LYP/6-311G++(2d, 2p). This value increased to record 78.66 kcal mol<sup>-1</sup> by use RHF/6-311G++(2d, 2p). The

intramolecular hyperconjugated interaction  $\sigma(LP1N32) \rightarrow \pi^*(N26-C28)$  was recorded 58.46 kcal mol<sup>-1</sup> by use of B3LYP/6-311G++(2d, 2p). This value increased to record 88.25 kcal mol<sup>-1</sup> by use RHF/6-311G++(2d, 2p). As similar as results of compound 1, The results of compound 2 Table 2 using B3LYP/6-311G++(2d, 2p) recorded less intramolecular hyper-conjugated interaction energies compared to the use of RHF/6-311G++(2d, 2p). For instant, the intramolecular hyperconjugated interaction  $\sigma(N32-H33) \rightarrow \pi^*(C36-C37)$  was recorded 0.86 kcal mol<sup>-1</sup> by use of B3LYP/6-311G++(2d, 2p). This value increased to 1.20 kcal mol<sup>-1</sup> with the RHF/6-311G++(2d, 2p) method calculation. The intramolecular hyperconjugated interaction  $\sigma(N7-H8) \rightarrow \pi^*(C1-N2)$  was recorded 2.68 kcal mol<sup>-1</sup> by use of B3LYP/6-311G++(2d, 2p) level. This value increased to 36.10 kcal mol<sup>-1</sup> with the RHF/6-311G++(2d, 2p) method calculation. The Lon pair intramolecular hyperconjugated interaction illustrated an enormous increase by use RHF/6-311G++(2d, 2p).level compared with B3LYP/6-311G++(2d, 2p). For example, the intramolecular hyperconjugated interaction  $\sigma(LP2O29) \rightarrow \pi^*(C4-N5)$  was recorded 6.24 kcal mol<sup>-1</sup> by use of B3LYP/6-311G++(2d, 2p). This value increased to 693.67 kcal mol<sup>-1</sup> with the RHF/6-311G++(2d, 2p) method calculation treatment B3LYP level versus RHF level significantly increases the magnitude of intramolecular hyperconjugated interaction energy.

# Comparsion of the Nature of Hyperconjugative Interactions between Compound 1 and Compound 2 using the B3LYP/6-311G++(2d, 2p) Level

The hyperconjugative interactions of the compounds 1,2 using B3LYP/6-311G++(2d, 2p) level are illustrated in Table 3

| Copound 1 intramolecular hyperconjugated interaction |      |              | Copound 2 intramolecular hyperconjugated interaction |                      |           |      |              |      |                      |
|--|------|--------------|--|----------------------|-----------|------|--------------|------|----------------------|
| Donor (i)  | Туре | Acceptor (j) | Туре   | (E)kcal/mol B3LYP C1 | Donor (i) | Туре | Acceptor (j) | Туре | (E)kcal/mol B3LYP C2 |
| C34- H35   | σ    | C38- C41     | π*   | 0.61                 | C34- H35  | σ    | C10- C12     | π*   | 0.72                 |
| C41- H45   | σ    | O2- C9       | π*   | 0.57                 | C41- H45  | σ    | C10- C12     | π*   | 3.41                 |
| LP1N32   |      | N26- C28     | π*   | 58.46                | LP1N32    |      | C1- N2       | π*   | 128.13               |

Table 3. The hyperconjugative interactions of the compounds 1,2 using B3LYP/6-311G++(2d, 2p) level.

The data of Tables 3 allow one to conclude that the replacement of a Hydrogen atom for compound 1 by a methyl group for compound 2 results an increase in the intramolecular hyper-conjugated interaction energy resulted from use of B3LYP with 311G++(2d, 2p) basis set for compound 2 compared to compound 1 results. For example, the intramolecular hyper-conjugated interaction energy from the donor  $\sigma$ (C34-H35) was recorded 0.61 kcal mol<sup>-1</sup> to  $\pi^*$ (C38-C41) for compound 1 and it was recorded 0.72 kcal mol<sup>-1</sup> to  $\pi^*$ (C10-C12) for compound 2. The Lone pair intramolecular hyperconjugated interaction energy from  $\sigma$ (LP1N32) was recorded 58.46 kcal mol<sup>-1</sup> to  $\pi^*$ (N26-C28) for compound 1 and it was recorded 182.13 kcal mol<sup>-1</sup> to  $\pi^*$ (C10-C12) for compound 2.

# Comparsion of the Nature of Hyperconjugative Interactions between Compound 1 and Compound 2 using the RHF/6-311G++(2d, 2p) Level

The hyperconjugative interactions of the compounds 1,2 using RHF/6.311G++(2d, 2p) level are illustrated in Table 4.

| Copound 1 intramolecular hyperconjugated interaction |      |                 | Copound 2 intramolecular hyperconjugated interaction |                       |             |      |                 |      |                       |
|--|------|-----------------|--|-----------------------|-------------|------|-----------------|------|-----------------------|
| Donor (i)  | Туре | Acceptor<br>(j) | Туре   | (E)kcal/mol RHF<br>C1 | Donor (i)   | Туре | Acceptor<br>(j) | Туре | (E)kcal/mol RHF<br>C2 |
| C34-<br>H35  | σ    | C46- C49        | π*   | 0.78                  | C34-<br>H35 | σ    | C4- N5          | π*   | 1.63                  |
| C41-H45  | σ    | C46- C49        | π*   | 0.99                  | C41-H44     | σ    | C40- C42        | π*   | 17.66                 |
| LP1N32   |      | N26- C28        | π*   | 88.25                 | LP1N32      |      | C1- N2          | π*   | 264.31                |

Table 4. The hyperconjugative interactions of the compounds 1,2 using RHF/6-311G++(2d, 2p).level.

From Table 4, it can be seen that the replacement of a hydrogen atom for compound 1 by a methyl group for compound 2 results an increased in the intramolecular hyper-conjugated interaction energies resulted from use of RHF with 311G++(2d, 2p) basis set for compound 2 compared to results of compound 1 from use the same method. For example, the intramolecular hyper-conjugated interaction energy from the donor  $\sigma(C34-H35)$  recorded 0.99kcal mol<sup>-1</sup> to  $\pi^*(C46-C49)$  for compound 1 and it was recorded 1.63 kcal mol<sup>-1</sup> to  $\pi^*(C4-N5)$  for compound 2. The Lone pair intramolecular hyperconjugated interaction energy from  $\sigma(LP1N32)$  to  $\pi^*(C1-N2)$  for compound 1 was recorded 88.25 kcal mol<sup>-1</sup> and it was recorded 264.31 kcal mol<sup>-1</sup> from  $\sigma(LP1N32)$  to  $\pi^*(C10-C12)$  for compound 2. These values in Tables 3,4 indicate

the impact of the replacement of a hydrogen atom by a methyl group which caused an increase in the intramolecular hyperconjugated interaction energies and stability of the molecule.

Comparison of Polarizaility ( $\alpha$ ), Hyperpolarizability ( $\beta$ ), and Anisotropic Polarizability ( $\Delta \alpha$ ) of the Title Compounds using the Two Different Levels B3LYP/6-311G++(2d, 2p) and RHF/6-311G++(2d, 2p) The polarizability( $\alpha$ ) ,hyper polarizability( $\beta$ ), and Anisotropic polarizability( $\Delta \alpha$ ) and HOMO-LUMO energy gaps of the title compounds using the two different levels B3LYP/6-311G++(2d, 2p) and RHF/6-311G++(2d, 2p) are represented in Table5.

| Method               | B3LYP/6-311G++(2d, 2p) |            | RHF/6-311G++(2d, 2p) |            |  |
|----------------------|------------------------|------------|----------------------|------------|--|
| Compounds            | Compound 1             | Compound 1 | Compound 1           | Compound 1 |  |
| a(esu)               | 53.24                  | 54.80      | 46.83                | 48.42      |  |
| $\Delta \alpha(esu)$ | 34.49                  | 32.35      | 27.45                | 25.35      |  |
| В                    | 17.63                  | 7.93       | 4.44                 | 3.81       |  |
| А                    | 7.62                   | 7.36       | 7.62                 | 7.36       |  |

**Table 5.** Polarizability ( $\alpha$ ), hyper polarizability ( $\beta$ ), and Anisotropic polarizability ( $\Delta \alpha$ ) of The Title Compounds

As it appears in Table 5, due to the presence of methyl group in compound 2 compare to compound 1, the polarizability( $\alpha$ ) increased from 53.24 esu for compound 1 to 54.80 esu for compound 2 by use of B3LYP/6-311G++(2d, 2p). However, the increase was larger at the RHF/6-311G++(2d, 2p) level. Since hyperconjugation intra molecular interaction energies is the property that induces polarizability in a molecule.(Msugh Targema, et al 2013). This was in a good agreement with our results of the polarizability and hyperconjugation intra molecular interaction energies for the two title compounds.

On the other hand, due to the presence of methyl group in compound 2 compare to presence of hydrogen in compound 1, the anisotropic polarizability ( $\Delta \alpha$ ) decreased from 34.49 esu for compound 1 to 32.35 esu for compound 2 by use of B3LYP/6-311G++(2d, 2p). However, the decrease was smaller at the RHF/6-311G++(2d, 2p) level. It can be noted (Table 5) that unlike the polarizability the hyperpolarizabilities exhibit rather wide range of variation by use of B3LYP/6-311G++(2d, 2p) level compare to use of RHF/6-311G++(2d, 2p) level. It was recorded 9.7 esu variation between compound 1 and compound 2 by use of B3LYP/6-311G++(2d, 2p) level. This variation was only 0.43 esu by use of RHF/6-311G++(2d, 2p) level. This was in a good agreement with K. Hatua\* and Prasanta K. Nandi study (2013).

# Comparsion of HOMO-LUMO Electro Properties Dependent of the Title Compounds using the Two Different Levels B3LYP/6-311G++(2d, 2p) and RHF/6-311G++(2d, 2p)



Figure 6. HOMO and LUMO plot of the title compound.



Figure 7. HOMO-LUMO of the title compounds using B3LYB/6-311G++(2d, 2p).and RHF/6-311G++(2d, 2p).

The HOMO- LUMO plots of the title compound are given in figures 5,6. The HOMO- LUMO results with electronic properties results are obtained in Table 6.

**Table 6.** The HOMO- LUMO and electronic properties of the title compounds using B3LYB/6-311G++(2d, 2p).andRHF/6-311G++(2d, 2p).

|             | rb3lyp /<br>6-311g++(2d,2p) |            | RHF /             |            |  |
|-------------|-----------------------------|------------|-------------------|------------|--|
| methods     |                             |            | 6-311g++ (2d, 2p) |            |  |
|             |                             |            |                   |            |  |
| compounds   | Compound 1                  | Compound 2 | Compound 1        | Compound 2 |  |
| HOMO        | -6.27                       | -6.25      | -8.14             | -8.11      |  |
| LUMO        | -1.63                       | -1.51      | 0.97              | 0.98       |  |
| $\Delta E$  | 4.65                        | 4.74       | 9.11              | 9.09       |  |
| χ           | -3.95                       | -3.88      | -3.58             | -3.57      |  |
| η           | 2.32                        | 2.37       | 4.56              | 4.54       |  |
| S           | 0.22                        | 0.21       | 0.11              | 0.11       |  |
| μ           | 3.95                        | 3.88       | 3.58              | 3.57       |  |
| ω           | 3.36                        | 3.18       | 1.41              | 1.40       |  |
| $\Delta En$ | 0.57                        | 0.48       | 0.10              | 0.11       |  |
| ΔEe         | 8.47                        | 8.25       | 7.27              | 7.24       |  |
| Pz          | -0.022                      | 0.086      | -0.047            | 0.093      |  |
| Vm          | 296.05                      | 378.82     | 350.26            | 334.52     |  |
| А           | 7.62                        | 7.36       | 7.62              | 7.36       |  |

With the study of frontier orbital HOMO-LUMO gap, one can easily investigate the electronic properties that depending on HOMO-LUMO gap. From Table 6, the change in the values of energy gaps indicated that the energy gaps increased by the replacement of a hydrogen atom for compound 1 with a methyl group for compound 2. For example when tittle compounds were optimized by using B3LYP/6-311G++(2d, 2p) basis set, the energy gap of compound 1 was 4.65eV and that value increased to 4.74eV for compound 2. In contrast results were obtained when the tittle compounds were optimized by using RHF/6-311G++(2d, 2p) level the energy gap decreased from 9.11 eV for compound 1 to 9.09eV for compound 2.

From the same Table 6, the electronegativity became less negative by the replacement of a hydrogen atom for compound 1 with a methyl group for compound 2 When the tittle compounds were optimized using B3LYP/6-311G++(2d, 2p), the electron negativity of compound 1 was -3.95 eV and it was -3.58eV for compound 2. Likewise when the tittle compounds were optimized using RHF/6-311G++(2d, 2p), the electronegativity became less negative. It was recorded -3.58eV for compound 1 and -3.57eV for compound 2. We can note from the same Table 6 that the softness did not so effect by the replacement of a example, when the tittle compounds were optimized using B3LYP/6-311G++(2d, 2p), compound 1 and compound 1 with a methyl group for compound 2 and the two tittle compounds had nearly values. For ev<sup>-1</sup> and 0.21 ev.<sup>-1</sup> respectively, and when the tittle compounds were optimized using RHF/6-311G++(2d, 2p), compound 1 had the same values 0.11 ev<sup>-1</sup>

From Table 6 the chemical potential  $\mu$  Electrophalicity index  $\omega$ , nuleofugality  $\Delta En$  and electrofugality  $\Delta Ee$  of the tittle compounds decreased by the replacement of a Hydrogen atom for compound 1 with a methyl group for compound 2. For example, the optimization of tittle compounds using B3LYP/6-311G++(2d, 2p) indicated that the chemical potential  $\mu$  of compound 1 was 3.95eV and decreased to 3.88 eV for compound 2. The electrophalicity index  $\omega$  was 3.36 eV for compound 1 and it decreased to 3.18eV for compound 2, electrofugality  $\Delta Ee$  was 8.47eV for compound 1 and it was recorded 8.25eV for compound 2. Nucleofugality  $\Delta En$  was recorded decreasing from 0.57eV for compound 1 to 0.48eV for compound 2.

As similar as use of B3LYP/6-311G++(2d, 2p) level, RHF/6-311G++(2d, 2p) level show a decrease in the values of all previse electro properties. The activity decreased from 7.62 to 7.36 by the replacement of a Hydrogen atom for compound 1 with a methyl group for compound 2. On the other hand, when the tittle compounds were optimized using B3LYP/6-311G++(2d, 2p) and RHF/6-311G++(2d, 2p) the natural atomic orbital occupancy  $p_z$  increased when Hydrogen atom for compound 1 replaced with a methyl group for compound 2.For instant when B3LYP/6-311G++(2d, 2p) was used the natural atomic orbital occupancy  $p_z$  was -0.022 for compound1 and it increased to 0.086 for compound 2 and when RHF/6-311G++(2d, 2p) was used the natural atomic orbital occupancy  $p_z$  was -0.047 for compound 1 and it increased to record 0.093 for compound 2.

#### 4. CONCLUSION

Through the optimization of 2-amino-3-[4-[4-(1-naphthale- 2ylmethylamino)-1,3,5-triazin-2-yl]phenyl]propanoic acid {Compound 1} and 2-amino-3-[4-[4-(1-naphthale- 2ylethylamino)-1,3,5-triazin-2-yl]phenyl]propanoic acid {Compound 2} by using Lee-Yang-Parr correlation functional (B3LYP) and Restricted Hartree-Fock (RHF) with the 6++311G(2d, 2p) basis set, our calculations indicate that the replacement of a Hydrogen atom for compound 1 by a methyl group for compound 2 results an increased in the intramolecular hyper-conjugated interaction energy and that change led to change in the other energy gap dependent values such as chemical potential, electronegativity, chemical hardness, electrophilicity index Polarizability ( $\alpha$ ), molar volume (V<sub>m</sub>), the first hyperpolarizability ( $\beta$ ) and the activity. From the natural atomic orbital occupancy analysis, it illustrated that the presence of methyl group as donating group in compound 2 caused change in p<sub>z</sub> from negative value for compound 1 to positive value for compound 2. So the quantum chemical calculation have great interest in creating a ways to predict the electronic properties of the compounds specially drugs compounds to find any drug –drug interaction or side effect and to predict new drug.

#### REFERENCES

- Hatua, K., & Nandi, P. K. (2013). Relationships between different-order polarizabilities and ground state dipole moment. Journal of Theoretical and Computational Chemistry, 12(01), 1250099.
- Jin, H., Cianchetta, G., Devasagayaraj, A., Gu, K., Marinelli, B., Samala, L., ... & Zang, Y. (2009). Substituted 3-(4-(1, 3, 5-triazin-2yl)-phenyl)-2-aminopropanoic acids as novel tryptophan hydroxylase inhibitors. *Bioorganic & medicinal chemistry letters*, 19(17), 5229-5232.
- Ouyang, L., He, G., Huang, W., Song, X., Wu, F., & Xiang, M. (2012). Combined structure-based pharmacophore and 3D-QSAR studies on phenylalanine series compounds as TPH1 inhibitors. *International journal of molecular sciences*, 13(5), 5348-5363.
- Prasad, O., Kumar, A., Narayan, V., Mishra, H. N., Srivastava, R. K., & Sinha, L. (2011). Quantum chemical study of molecular structure, non linear optical and vibrational properties of ortho and meta-fluorobenzaldehyde. J Chem Pharm Res, 3, 668-677.



# Quantum Chemical studies of Thiosemicarbazide, Phenyl isothiocyanate and their condensation 1-Phenyl1-2,5-dithiohydrazodicarbonamide as Corrosion Inhibitors for Copper in aqueous Sodium Chloride

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**Abstract:** Copper is the fifth most regular metal in the world which is valuable in unadulterated or alloying shape. Copper and its combinations are generally utilized in industries in light of some positive properties, for example, high electrical and thermal conductivity, mechanical workability and malleability. Copper and its combinations are exceptionally respected as a result of their wide application underway of wire, sheets and pipelines in electronic industries, heat exchangers and cooling towers. As per across the board utilization of copper in diverse industries, the issue of corrosion and corrosion protection of copper has pulled in a great deal of consideration and numerous investigations have been led to date on this issue are as yet continuous.

In this paper quantum chemical calculations based on density functional theory (DFT) method were performed on the selected thiosemicarbazide (InA), phenyl isothiocyanate (InB) and their condensation 1-phenyl1-2,5-dithiohydrazodicarbonamide (InC) used as corrosion inhibitors for copper in aqueous chloride solutions to determine the relationship between the molecular structure of inhibitors and inhibition efficiency. All different calculations were performed with of complete geometry optimization by using the standard Gaussian 9.0 software package. The structural parameters, such as energy and distribution of highest occupied molecular orbital (HOMO) and lowest unoccupied molecular orbital (LUMO), the charge distribution of the studied inhibitors, the absolute electronegativity ( $\chi$ ) values were calculated and correlated with inhibition efficiencies. The results showed that the inhibition efficiency of inhibitors increased with the increase in energy of HOMO and decrease in energy gap of frontier molecular orbital, and the areas containing N and S atoms are most possible sites for bonding the copper surface by donating electrons to the Cu. According to the results, the inhibition efficiencies of inhibitors follow the sequence : InC > InA > InB. The 1-phenyl1-2,5-dithiohydrazodicarbonamide molecules has the smallest energy gap and the highest reactivity, and therefore has the highest inhibition efficiency among the investigated inhibitors

**Keywords:** Copper, DFT Calculations, Thiosemicarbazide, Phenyl isothiocyanate, 1-phenyl1-2,5dithiohydrazodicarbonamide

## **1. INTRODUCTION**

With propelling industrialisation, the interest for and the quantity of copper amalgams has risen constantly. Today, copper amalgams have discovered a significant position among metallic materials because of their substantial assortment of metallurgical, physical and compound properties. A cutting edge modern culture utilizes copper in numerous zones (electrotechnology, building and development, mechanical designing, car, science, seaward applications, marine building, therapeutic purposes and numerous others).

It is noticed that nearness of heteroatoms, for example, nitrogen, sulfur, phosphorous in the natural compound atom improves its activity as copper corrosion inhibitor. This is clarified by the nearness of empty d orbitals in copper particle that structure coordinative bonds with molecules ready to give electrons <sup>(1)</sup>. Connection with rings containing conjugated bonds, electrons, is additionally present. In light of these outcomes an ever increasing number of mixes containing various heteroatoms and useful gatherings are created integrated since it is seen they are in charge of good properties with respect

to consumption restraint since they empower chemisorption. Additionally sub-atomic weight is bigger because of its helpful impact on physical adsorption <sup>(2)</sup>. There are endeavors to consolidate hypothesis and down to earth understanding from examinations of a few substances having comparative structure so as to discover models that would empower forecast of potential outcomes of recently blended mixes to go about as corrosion inhibitors.

From previous studies, Thiosemicarbazide and its derivatives are used as corrosion inhibitors for different metallic substrates <sup>(3)</sup>. Corrosion inhibitors are chemical compounds that are widely used in corrosion technology to protect structures from deterioration. Corrosion inhibitors offer protection by forming a protective film at the steel surface and reducing the ingress of aggressive ions into the concrete matrix. The inhibition efficiency of homologous series of organic substances, containing different heteroatoms, are in the following sequence; P > Se > S > N > O. Some amides and derivatives such as urea (U), thiourea (TU), thioacetamide (TA) and thiosemicarbazide (TSC) have been found to be potential inhibitors. A mixture of nitrogen and sulphur compounds is often found to be better than either type alone<sup>(5)</sup>. Several nitrogen and sulphur containing compounds have been reported to be effective inhibitors for different metals and the relationship between the amide molecular structures and their inhibition efficiencies have been studied in several research works. Aminoalcohols such as ethanolamine, dimethylethanolamine N,N-dimethylethanolamine can also be used for the protection of rebar in concrete in commercial corrosion inhibitors<sup>(4)</sup>.

In this study quantum chemical calculations based on density functional theory (DFT) method were performed on the selected thiosemicarbazide (InA), phenyl isothiocyanate (InB) and their condensation 1-phenyl1-2,5-dithiohydrazodicarbonamide (InC) used as corrosion inhibitors for copper in aqueous chloride solutions to determine the relationship between the molecular structure of inhibitors and inhibition efficiency by using the standard Gaussian 9.0 software package.

## 2. METHODOLOGY

Three organic inhibitors (thiosemicarbazide (InA), phenyl isothiocyanate (InB) and their condensation 1-phenyl1-2,5dithiohydrazodicarbonamide (InC)) as shown in Figure (1) were used in this study. All different calculations will be performed with of complete geometry optimization by using the standard Gaussian 9.0 software package. The molecular optimization was achieved using the functional B3LYP density functional theory (DFT) formalism, having an electron basis set : B3LYP/6-311G (d,p), 6-311G2d3p in gas phase and water phase. The structural parameters, such as energy and distribution of highest occupied molecular orbital (HOMO) and lowest unoccupied molecular orbital (LUMO), the charge distribution of the studied inhibitors, the absolute electronegativity ( $\chi$ ) values were calculated and correlated with inhibition efficiencies.



Figure (1) structure of organic inhibitors

## 3. RESULTS AND DISSCUSION

Thiosemicarbazide (InA), phenyl isothiocyanate (InB) and their condensation 1-phenyl1-2,5dithiohydrazodicarbonamide (InC)) were optimized to ground state with minimum energy and the optimized geometry, frontier molecular orbital's density distribution (HOMO and LUMO), the energy gap between ELUMO and EHOMO ( $\Delta E$ ), Chemical Potential ( $\mu$ ), hardness ( $\eta$ ), softness ( $\sigma$ ), electronegativity ( $\chi$ ), global electrophilicity ( $\omega$ ). All HOMO and LUMO are listed in Table (1- 4) in gas and water phase , while HOMO and LUMO are listed in Table (5). It is that clear the HOMO for InC was higher than InA and InB while LUMO for InC was lower than InA and InB that mean the InC much more favor electron donations to vacant d-orbitals of (Cu) during donor-acceptor interactions than InA and  $InB^{(7)}$ . In light of the the frontier molecular orbital hypothesis,  $E_{HOMO}$  is regularly corresponded with the electron giving capacity of the particle and ofers impressive understanding about the destinations or sections of the assessed atoms which can fill in as electron giver amid the adsorption on the metal surface while  $E_{LUMO}$  is related with the electron tolerating capacity of the particle and lower the esteem better is the capability of the atom to get electrons from the metal surface<sup>(8-10)</sup>.

From Figure (9), it is noticed that the energy gap between ELUMO and EHOMO ( $\Delta E$ ) was lowest in(InC), while ( $\Delta E$ ) for (InB) was lower than (InA) with bases set (6-311G(d,p). From figure (10), it is noticed that ( $\Delta E$ ) for (InC) was the lowest, while ( $\Delta E$ ) for (InA) was lower than (InB) with bases set (6-311G2d3p). The gap energy ( $\Delta E_{LUMO-HOMO}$ ) is a proportion of reactivity of a particle. Thus, an atom with negligible partition vitality are related with high synthetic reactivity and high hindrance efficiency, as the excitation vitality to expel an electron from the last involved orbital will be low. A smaller gap energy signifes high soundness for the compound in the complex framed with the copper surface<sup>(13)</sup>.

It is that clear from figure (10 - 11) it is noticed that the electron distribution of HOMO orbitals for (InA) are localized on S,N,C atoms ,while LUMO localized on 1S,2N,3C,4C,5C,6C,7C,8C,9C. For (InB) HOMO are localized 1S,2N,3C,4C,5C,6C,7C,8C,9C,while LUMO are localized S,N,3C,4C,5C,6C,7C,8C. For (InC) HOMO are localized S,2N,3C,4C,5C,6C,7C,8C,9C,20S While LUMO are localized S,2N,3C,4C,5C,6C,7C,8C,9C,15N and this is the reason the (InC) was lowest enrgy gap and lead to be more attraction with copper surface<sup>(11-12).</sup>

From Figure (13), it is noticed that the correlation plot between theoretical energy gap and experimental inhibitor efficiency bases set 6-311 G(2d,3p) was  $R^2 = 0,8247$ , while in Figure (14) the correlation plot between theoretical energy gap and experimental inhibitor efficiency bases set 6-311G(d,p) was  $R^2 = 0,2483$ ,that mean the correlation plot with bases set 6-311 G(2d,3p) much more acceptable than with bases set 6-311G(d,p) and this is the main reason why it was used two bases set in this investigation<sup>(14-15)</sup>.

| 6-311G(d,p) | 1-TSC-G  | 2-PISOTHCY - | 3b-phecar -G |
|-------------|----------|--------------|--------------|
|             |          | G            |              |
| LUMO+5      | 0,08696  | 0,06413      | 0,03346      |
| LUMO+4      | 0,07848  | 0,03645      | 0,01527      |
| LUMO+3      | 0,06139  | 0,03138      | -0,00932     |
| LUMO+2      | 0,05468  | -0,00200     | -0,02453     |
| LUMO+1      | 0,00897  | -0,03163     | -0,04898     |
| LUMO        | 0,00477  | -0,05629     | -0,05730     |
| HOMO        | -0,20712 | -0,23843     | -0,21278     |
| HOMO-1      | -0,21176 | -0,25772     | -0,22056     |
| HOMO-2      | -0,28060 | -0,28033     | -0,23115     |
| HOMO-3      | -0,31147 | -0,30027     | -0,24097     |
| HOMO-4      | -0,36345 | -0,36985     | -0,27660     |
| HOMO-5      | -0,40323 | -0,38381     | -0,29429     |

Table (1) HOMO and LUMO for three compounds in Gas Phase (6-311G(d,p)

Table (2) HOMO and LUMO for three compounds in Water Phase (6-311G(d,p)

| 6-311G(d,p) | 1-TSC-W | 2-PISOTHCY - | 3b-phecar -W |
|-------------|---------|--------------|--------------|
|             |         | W            |              |
| LUMO+5      | 0,10776 | 0,05944      | 0,04498      |
| LUMO+4      | 0,07764 | 0,04306      | 0,03338      |
| LUMO+3      | 0,06805 | 0,03523      | -0,01788     |
| LUMO+2      | 0,05662 | -0,00714     | -0,01971     |

| LUMO+1 | 0,03037  | -0,02677 | -0,03016 |
|--------|----------|----------|----------|
| LUMO   | -0,00495 | -0,05611 | -0,04444 |
| HOMO   | -0,22981 | -0,24237 | -0,23572 |
| HOMO-1 | -0,23147 | -0,26571 | -0,23719 |
| HOMO-2 | -0,28243 | -0,27568 | -0,24028 |
| HOMO-3 | -0,30028 | -0,30126 | -0,24317 |
| HOMO-4 | -0,37942 | -0,36459 | -0,27123 |
| HOMO-5 | -0,41068 | -0,38146 | -0,27586 |

Table (3) HOMO and LUMO for three compounds in Gas Phase (6-311G(2d,2p)

| 6-311G2d3p | 1-TSC-G  | 2-         | 3b-phecar -G |
|------------|----------|------------|--------------|
|            |          | PISOTHCY - |              |
|            |          | G          |              |
| LUMO+5     | 0,04043  | 0,00368    | -0,01100     |
| LUMO+4     | 0,01440  | 0,00322    | -0,01682     |
| LUMO+3     | 0,00717  | -0,00705   | -0,02778     |
| LUMO+2     | 0,00113  | -0,01475   | -0,03082     |
| LUMO+1     | -0,00449 | -0,03699   | -0,05371     |
| LUMO       | -0,03230 | -0,05954   | -0,06050     |
| НОМО       | -0,21033 | -0,23944   | -0,21445     |
| HOMO-1     | -0,21613 | -0,25813   | -0,22350     |
| HOMO-2     | -0,28556 | -0,28208   | -0,23188     |
| HOMO-3     | -0,31659 | -0,30128   | -0,24263     |
| HOMO-4     | -0,36855 | -0,37107   | -0,27887     |
| HOMO-5     | -0,40734 | -0,38550   | -0,29586     |

Table (4) HOMO and LUMO for three compounds in Water Phase (6-311G(2d,2p)

| 6-311G2d3p | 1-TSC-W  | 2-PISOTHCY - | 3b-phecar -W |  |  |  |  |
|------------|----------|--------------|--------------|--|--|--|--|
|            |          | W            |              |  |  |  |  |
| LUMO+5     | 0,03286  | 0,01321      | 0,00060      |  |  |  |  |
| LUMO+4     | 0,02960  | 0,00440      | -0,01306     |  |  |  |  |
| LUMO+3     | 0,01590  | -0,00801     | -0,02466     |  |  |  |  |
| LUMO+2     | 0,00909  | -0,01209     | -0,02732     |  |  |  |  |
| LUMO+1     | -0,01324 | -0,03186     | -0,03456     |  |  |  |  |
| LUMO       | -0,01355 | -0,05902     | -0,04607     |  |  |  |  |
| HOMO       | -0,23104 | -0,24315     | -0,23623     |  |  |  |  |
| HOMO-1     | -0,23368 | -0,26587     | -0,23768     |  |  |  |  |
| HOMO-2     | -0,28676 | -0,27706     | -0,24138     |  |  |  |  |
| HOMO-3     | -0,30552 | -0,30190     | -0,24475     |  |  |  |  |
| HOMO-4     | -0,38265 | -0,36564     | -0,27370     |  |  |  |  |
| HOMO-5     | -0,41389 | -0,38270     | -0,27720     |  |  |  |  |

| 6-311G(d,p) | 1-TS     | C-Cu          | 2-PISOT  | HCY -Cu       | 3b-phecar -Cu |               |  |  |
|-------------|----------|---------------|----------|---------------|---------------|---------------|--|--|
|             | Alpha    | beta orbitals | Alpha    | beta orbitals | Alpha         | beta orbitals |  |  |
|             | orbitals |               | orbitals |               | orbitals      |               |  |  |
| LUMO+5      | 0,00425  | -0,00218      | -0,03134 | -0,03313      | -0,02549      | -0,03262      |  |  |
| LUMO+4      | -0,00326 | -0,00945      | -0,03588 | -0,06292      | -0,03765      | -0,05116      |  |  |
| LUMO+3      | -0,01200 | -0,02001      | -0,06673 | -0,07404      | -0,05201      | -0,05518      |  |  |
| LUMO+2      | -0,02754 | -0,06601      | -0,07643 | -0,07840      | -0,05669      | -0,05743      |  |  |
| LUMO+1      | -0,06673 | -0,07771      | -0,08262 | -0,08219      | -0,06239      | -0,06599      |  |  |
| LUMO        | -0,08026 | -0,07792      | -0,08459 | -0,10032      | -0,08124      | -0,09624      |  |  |
| НОМО        | -0,11373 |               | -0,13480 |               | -0,14462      |               |  |  |
| HOMO-1      | -0,16418 | -0,16056      | -0,17572 | -0,17457      | -0,15648      | -0,15064      |  |  |
| HOMO-2      | -0,16736 | -0,16307      | -0,18673 | -0,17931      | -0,16641      | -0,16290      |  |  |
| HOMO-3      | -0,16941 | -0,16419      | -0,18793 | -0,18227      | -0,16746      | -0,16497      |  |  |
| HOMO-4      | -0,17069 | -0,16739      | -0,18812 | -0,18494      | -0,16786      | -0,16643      |  |  |
| HOMO-5      | -0,17079 | -0,16918      | -0,18933 | -0,18594      | -0,16949      | -0,16689      |  |  |

Table (5) HOMO and LUMO for three compounds intraction with copper in gas phas



Figure (7) HOMO and LUMO for three compounds intraction with Copper



Figure (8) HOMO and LUMO for three compounds intraction with copper and Copper alone



Figure (9) HOMO and LUMO for three compounds (6-311G(d,p)



Figure (10) HOMO and LUMO for three compounds (6-311G(2d,2p)





Figure (11) Homo and Lumo for copper composite with three compound

|              | A DE DE DE DE DE DE DE DE DE DE DE DE DE |                  |
|--------------|--|------------------|
| 1-TSC-CU-ESP | 2-PISOTHCY-CU-ESP                        | 3b-phecar-CU-ESP |



## Figure (12) Electrostatic Potential Images

| Molecule                                  | Method      | HOMO<br>(au) | LUMO<br>(au) | Convert<br>unit<br>(eV) | E <sub>HOMO</sub> | E <sub>lumo</sub> | I    | A     | Energy<br>gap | Absolut<br>e<br>hardne<br>ss (ŋ) | softnes<br>s (S) | Electronegativity | chemical potential (μ ) | Global<br>electro<br>philicit<br>y<br>index(<br>w) | global<br>molecula<br>r<br>nucleoph<br>ilicity (ε) | Nucleofugalit<br>y | Electrofugal<br>ity |
|---|-------------|--------------|--------------|-------------------------|-------------------|-------------------|------|-------|---------------|----------------------------------|------------------|-------------------|-------------------------|--|--|--------------------|---------------------|
|   |             |              |              |                         | (eV)              | (eV)              | (eV) | (eV)  | ΔE<br>(eV)    | (eV)                             | $(eV^1)$         | χ (eV)            | μ (eV)                  | ω  | (eV <sup>-1</sup> )                                | (eV)               | (eV)                |
| Thiosemicarbazide - G                     | 6-311G(d,p) | -0,2071      | 0,00477      | 27,212                  | -5,636            | 0,130             | 5,64 | -0,13 | 5,766         | 2,883                            | 0,347            | 2,753             | -2,753                  | 1,315  | 0,76069  | 0,00292            | 5,50919             |
| Thiosemicarbazide - W                     | 6-311G(d,p) | -0,2298      | -0,00495     | 27,212                  | ·6,253            | -0,135            | 6,25 | 0,13  | 6,119         | 3,059                            | 0,327            | 3,194             | -3,194                  | 1,667  | 0,59975  | 0,00297            | 6,39116             |
| Phenyl-isothiocyanate- G                  | 6-311G(d,p) | -0,2384      | -0,05629     | 27,212                  | -6,488            | -1,532            | 6,49 | 1,53  | 4,956         | 2,478                            | 0,404            | 4,010             | -4,010                  | 3,244  | 0,30824  | 0,47338            | 8,49318             |
| Phenyl-isothiocyanate- W                  | 6-311G(d,p) | -0,2424      | -0,05611     | 27,212                  | -6,595            | -1,527            | 6,60 | 1,53  | 5,068         | 2,534                            | 0,395            | 4,061             | -4,061                  | 3,254  | 0,30732  | 0,45995            | 8,58207             |
| 1-pheny1-2,5-dithiohydrazodicarbonamide-G | 6-311G(d,p) | -0,2128      | -0,0573      | 27,212                  | -5,790            | -1,559            | 5,79 | 1,56  | 4,231         | 2,115                            | 0,473            | 3,675             | -3,675                  | 3,192  | 0,31333  | 0,57463            | 7,92394             |
| 1-pheny1-2,5-dithiohydrazodicarbonamide-W | 6-311G(d,p) | -0,2357      | -0,04444     | 27,212                  | ·6,414            | -1,209            | 6,41 | 1,21  | 5,205         | 2,603                            | 0,384            | 3,812             | -3,812                  | 2,791  | 0,35823  | 0,28095            | 7,90455             |

Table 6. The calculated quantum parameters for studied inhibitors obtained using DFT with method 6-311G (d,P)

| Molecule                                   | Method     | HOMO<br>(au) | LUMO<br>(au) | Convert<br>unit<br>(eV) | E <sub>HOMO</sub> | E <sub>luno</sub> | I    | A    | Energy<br>gap | Absolut<br>e<br>hardne<br>ss (ŋ) | softnes<br>s (S)    | Electronegativity | chemical potential (μ ) | Global<br>electro<br>philicit<br>y<br>index(<br>ω) | global<br>molecula<br>r<br>nucleoph<br>ilicity (ε) | Nucleofugalit<br>y | Electrofugal<br>ity |
|--|------------|--------------|--------------|-------------------------|-------------------|-------------------|------|------|---------------|----------------------------------|---------------------|-------------------|-------------------------|--|--|--------------------|---------------------|
|  |            |              |              |                         | (eV)              | (eV)              | (eV) | (eV) | ΔE<br>(eV)    | (eV)                             | (eV <sup>-1</sup> ) | χ (eV)            | μ (eV)                  | ω  | (eV <sup>-1</sup> )                                | (eV)               | (eV)                |
| Thiosemicarbazide - G                      | 6-311G2d3p | -0,2103      | -0,0323      | 27,212                  | -5,723            | -0,879            | 5,72 | 0,88 | 4,844         | 2,422                            | 0,413               | 3,301             | -3,301                  | 2,250  | 0,44454  | 0,15947            | 6,76182             |
| Thiosemicarbazide - W                      | 6-311G2d3p | -0,231       | -0,01355     | 27,212                  | -6,287            | -0,369            | 6,29 | 0,37 | 5,918         | 2,959                            | 0,338               | 3,328             | -3,328                  | 1,871  | 0,53440  | 0,02297            | 6,67866             |
| Phenyl-isothiocyanate- G                   | 6-311G2d3p | -0,2394      | -0,05954     | 27,212                  | -6,516            | -1,620            | 6,52 | 1,62 | 4,895         | 2,448                            | 0,409               | 4,068             | -4,068                  | 3,380  | 0,29584  | 0,53622            | 8,67194             |
| Phenyl-isothiocyanate- W                   | 6-311G2d3p | -0,2432      | -0,05902     | 27,212                  | -6,617            | -1,606            | 6,62 | 1,61 | 5,010         | 2,505                            | 0,399               | 4,111             | -4,111                  | 3,373  | 0,29643  | 0,51479            | 8,73732             |
| 1-pheny1-2,5-dithiohydrazodicarbonamide -G | 6-311G2d3p | -0,2145      | -0,0605      | 27,212                  | -5,836            | -1,646            | 5,84 | 1,65 | 4,189         | 2,095                            | 0,477               | 3,741             | -3,741                  | 3,341  | 0,29935  | 0,64697            | 8,12880             |
| 1-pheny1-2,5-dithiohydrazodicarbonamide-W  | 6-311G2d3p | -0,2362      | -0,04607     | 27,212                  | -6,428            | -1,254            | 6,43 | 1,25 | 5,175         | 2,587                            | 0,387               | 3,841             | -3,841                  | 2,851  | 0,35075  | 0,30372            | 7,98555             |

Table 7. The calculated quantum parameters for studied inhibitors obtained using DFT with method 6-311G (2d,2p)



Figure (13) Correlation between Theoretical Energy gap and Experimental inhibitor efficiency Bases set 6-311 G(2d,2p)



Figure (14) Correlation between Theoretical Energy gap and Experimental inhibitor efficiency Bases set 6-311 G(d,p)

#### 4. CONCLUSION

1-The results showed that the inhibition efficiency of inhibitors increased with the increase in energy of HOMO and decrease in energy gap of frontier molecular orbital, and the areas containing N and S atoms are most possible sites for bonding the copper surface by donating electrons to the Cu.

2-The theoretical study was approved with experimental work

3-According to theoretical study, the inhibition efficiencies of the inhibitors follow the sequence:

1-phenyl-2,5-dithiohydrazodicarbonamide > Thiosemicarbazide > phenyl isothiocyanate

#### REFERENCES

- J.B. Matos, L.P.Pereira, S.M.L.Agostinho, O.E.Barcia, G.G.O.Cordeiro, E.D'Elia, Journal of electroanalytical chemistry 570 (2004) 91).
- 2. G. Moretti, F.Guidi, Corrosion science 44 (2002) 1995.
- 3. (A. Lalitha, S. Ramesh and S. Rajeswari , Surface protection of copper in acid medium by azoles and surfactantsElectrochimica Acta, (2005)
- 4. A. Lalitha, S. Ramesh and S. Rajeswari , Surface protection of copper in acid medium by azoles and surfactantsElectrochimica Acta, (2005).
- 5. Ameena Mohsen Al-Bonayan, Inhibiting Effect of Thiosemicarbazide and 4-Phenyl Thiosemicarbazide Towards the Corrosion of Carbon Steel in H3PO4 Solutions, Int. J. Electrochem. Sci., 10 (2015) 589 601.
- Lukovits, I., Kálmán, E. & Zucchi, F. Corrosion inhibitors-correlation between electronic structure and efficiency. Corrosion. 57, 3–8 (2001).
- 7. Khaled, K. F. Molecular simulation, quantum chemical calculations and electrochemical studies for inhibition of mild steel by triazoles. Electrochim. Acta 53, 3492 (2008).
- J. Radilla, G.E. Negrón, DFT study of the adsorption of the corrosion inhibitor 2-mercaptoimidazole onto Fe(100) surface, Electrochim. Acta 112 (2013) 577–586.
- 9. G. Bereket, E. Hür, C. Öğretir, Quantum chemical studies on some imidazole derivatives as corrosion inhibitors for iron in acidic medium, J. Mol. Struct. Theochem. 578 (2002) 79–88.
- I.B. Obot, D.D. Macdonald, Z.M. Gasem, Density functional theory (DFT) as a powerful tool for designing new organic corrosion inhibitors. Part 1: an overview, Corros. Sci. 99 (2015) 1–30.
- T. Arslan, F. Kandemirli, E.E. Ebenso, I. Love, H. Alemu, Quantum chemical studies on the corrosion inhibition of some sulphonamides on mild steel in acidic medium, Corros. Sci. 51 (2009) 35–47.
- A. Ongun Yüce, B. Doğru Mert, G. Kardaş, B. Yazıcı, Electrochemical and quantum chemical studies of 2-amino-4-methyl-thiazole as corrosion inhibitor for mild steel in HCl solution, Corros. Sci. 83 (2014) 310–316.
- 13. R.M. Issa, M.K. Awad, F.M. Atlam, Quantum chemical studies on the inhibition of corrosion of copper surface by substituted uracils, Appl. Surf. Sci. 255 (2008) 2433–2441.
- N.A. Wazzan, F.M. Mahgoub, DFT calculations for corrosion inhibition of ferrous alloys by pyrazolopyrimidine derivatives, Open J. Phys. Chem. 4 (2014) 6–14.
- H. Fan, S. Li, Z. Zhao, H. Wang, Z. Shi, L. Zhang, Inhibition of brass corrosion in sodium chloride solutions by self-assembled silane films, Corros. Sci. 53 (2011) 4273–4281.


# Breast Cancer Classification: An Adaptive Neuro-Fuzzy Inference System Approach

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Abstract: Nowadays, in various classification applications, data sets are generally used as information matrixes. These applications solve problems in many fields such as medicine, computer, engineering and natural sciences. Recently, almost all of these areas have focused on new techniques to be an alternative to the old methods. As a classification application, machine learning methods of artificial intelligence models are used and highly successful results are obtained. Breast cancer, one of the most important cancer types of our time, is the most common type of cancer among women. Breast cancer is a malignant tumor and occurs in the breast cells. Scientists have many data sets on breast cancer diagnostic methods and predict what factors can determine cancer on these clusters. At this point, classification methods are included in the subject. Artificial Neural Network and Fuzzy Logic algorithms came together to create a hybrid algorithm and named Adaptive Neuro-Fuzzy Inference System (ANFIS) algorithm. In this study, an existing breast cancer data set was used by ANFIS classification method. The results of the training and test data sets used in the classification were compared as error rate and accuracy values. As a result of the findings, ANFIS method was reported to be a good classifier in the diagnosis of breast cancer.

**Keywords :** ANFIS, Adaptive Neuro-Fuzzy Inference System, Breast Cancer, Classification, Artificial Neural Networks, Artificial Intelligence.

# **1. INTRODUCTION**

As with all cancers, early diagnosis and treatment in breast cancer is important for patients. For this purpose, as well as developing medical methods, estimation systems developed by using technology have been developed. On the data sets formed by data obtained from routine consultation and blood analysis, predictions can be made about whether the patients are healthy by using machine learning [1] methods, which is a sub-branch of artificial intelligence [2].

In a study published by Patrício et al. In 2018, a breast cancer prediction model was developed and evaluated based on anthropometric data and parameters that could be collected from routine blood analysis [3]. In the study, the predictive accuracy value obtained by using machine learning algorithms Logistic Regression [4], Random Forest [5], Support Vector Machines [6] is 95%. Monte Carlo Approach [7] was used during the study and the values of the model such as Sensitivity, Specificity and AUC [8] were calculated. At the end of the study, the sensivity range is 82% to 88%, the specificity range is 85% and 90%, and the AUC is 0.87 and 0.91. In this study, Patrício et al. found the data set safe in the study published in 2016 by Crisóstomo et al. and applied their own methods on this data set [9]. The data set was published on the UCI Machine Learning Repository web page [10]. Glucose, Insulin, HOMA, Leptin, Adiponectin, Resistin, MCP-1, Age and Body Mass Index values in this data set has a total of 116 samples.

With the Adaptive Neuro Fuzzy Inference System (ANFIS) method to be used in this study, estimation studies have been done on many natural sciences from past to present in the literature. Some of those; energy, socio-economic, economics, medicine, physics. Based on these estimation achievements, successful results were obtained in studies on breast cancer data sets.

In their study in 2005, Song et al. [11] predicted that a human-like decision process could be adapted to the diagnosis by using artificial intelligence algorithms in the detection of breast cancer. Anfis has developed a diagnostic system because it is an artificial intelligence algorithm that has the advantages of both fuzzy inference and neural networks. In this system, with the decision of the genetic algorithm, decision tree and correlation coefficient, the input of the data in the data set is given and a high accuracy value has been obtained by increasing the success performance.

In a study conducted in 2008 by Das et al. [12], he developed a neuro-fuzzy system based on a genetic algorithm to reach a solution by treating breast cancer in a computer-aided environment. In the proposed method, the basic features generated from images of tumor lesions seen in the chest were made into a data set for classification. At this stage, a genetic algorithm was used as a feature extraction method to narrow the number of properties in the data set with many features. Finally, the classification was studied with the method of ANFIS and the accuracy estimate of this proposed technique was specified as 87%.

In a study conducted in 2012 by Huang et al. [13], they compared the success of CBR (case-based reasoning) classifiers based on logistic regression model with ANFIS (artificial neural network), ANFIS (adaptive neuro-fuzzy inference system) and decision tree models based on PSO (particle swarm optimizer) based breast cancer data set. The results were demonstrated by CBR-based ANN classification with 83.60% accuracy, PSO-based ANN classification with 91.10% accuracy, and 92.00% accuracy with ANFIS classification method.

In a study published in 2012 by Fatima et al. [14], The University of California, Irvine (UCI) was developed by using the Anfis method as a diagnostic method using the data of the Wisconsin Breast Cancer Diagnosis (WBCD) database. As a result of the study, they showed that they obtained the best result in the same dataset in the literature with an accuracy value of 98.25%.

In a study conducted in 2013 by Shleeg et al. [15], they used Mamdami and Sugeno type fuzzy inference models with a model they developed on a breast cancer data set. When they compared the performances of the two types, they showed that using Sugeno was more advantageous than the Mamdani type.

In a study published in 2014 by Lalli et al. [16], he mentioned the use of thermography as the most effective method in the diagnosis of breast cancer and upon this, he created a data set on the images of the temogram. This data set has been used to measure success performance by using Anfis classification method and it is mentioned that this method can be used in breast cancer tumor.

The purpose of this study, the most important features of the data were found in the data set by using the method of Glucose, Insulin, HOMA, Leptin, Adiponectin, Resistin, MCP-1, Age and Body Mass Index. Afterwards, classification error rates and accuracy values were calculated by using the Adaptive Network Based Fuzzy Logic Inference System (ANFIS) which is an algorithm formed by the combination of Artificial Neural Networks and Fuzzy Logic.

# 2. MATERIAL AND METHODS

The artificial neural network algorithm, which is inspired by the biological nervous system, is capable of analyzing highperformance and complex events. Fuzzy logic algorithms, which are not suitable for all kinds of problems, are an algorithm which performs the modeling of fuzzy logic sets as a result of the analysis of problems, variables and the formation of parameters such as logic relations. A hybrid algorithm has been developed by using two algorithms. The basis of this algorithm, known as the ANFIS method, has the properties of both artificial neural networks [17] and fuzzy logic [18]. After defining input and output sets, the system is modeled with fuzzy logic. In the modeling phase, the optimization stage is realized by the learning methods of artificial neural networks.

In the basic principle of ANFIS architecture, it has been noted that the inputs and outputs comply with the rule "if – then" in accordance with the fuzzy inference system. For this reason, the first order is expressed in the Sugeno fuzzy rule model [19];

## If $x A_i$ and $y B_i$ Then $f_i = p_i x + q_i y + r_i$

The ANFIS operating principle consists of five separate layers, each with a different task. These layers work with as many neurons as the number of input sets. At the same time, this method is used in conjunction with Sugeno and Tsukamoto inference methods [20]. Figure 1 shows the two-input and two-order fuzzy logic system by modifying the Sugeno method.



Figure 1: Two inputs and two rules fuzzy logic - Sugeno method.

The rules of the first order two input sugeno method are determined as *If*  $x A_1$  *and*  $y B_1$  *then*  $f_1 = p_1 x + q_1 y + r_1$  and *If*  $x A_2$  *and*  $y B_2$  *then*  $f_2 = p_2 x + q_2 y + r_2$ . Consequently, the average of the weights of each rule determined by the method is calculated by

$$f = \frac{w_1 f_1 + w_2 f_2}{w_1 + w_2} = \overline{w}_1 f_1 + \overline{w}_2 f_2.$$



#### Figure 2: Flowchart of ANFIS Model

In the flowchart of ANFIS model shown in Figure 2, the data is initially blurred by the fuzzy system. For the learning stage of the fuzzy system created, the number of iterations (epoch) and the number of tolerances (errors) are specified, then the learning process continues until it reaches the tolerance point. Finally, the datasets are tested with independent data to calculate their accuracy.

Figure 3 shows the ANFIS architecture. The nodes in the second and third layers of this architecture are static, and the nodes in the remaining layers are adaptable with both the model parameters and the variables during the training [21].



#### Figure 3: ANFIS Architecture.

*Layer 1:* In this layer, known as the fuzzy layer, the output nodes are rated by each input node using the Gaussian membership function [22]. Each node in the layer is a function calculated by Equation 1.

$$(\mu_{Ai}(x), \mu_{Bi}(y)) \tag{I}$$

In Figure 3, x represents the input values, and Ai represents the fuzzy set of the node. The  $\mu$  Ai (x) has a value between 0 and 1 as a result of the Gauss function.

Layer 2: In this layer, known as the layer of rules, as seen in Equation 2, each node found consists of the product of the membership level from the previous layer.

 $w_i = \mu_{Ai}(x) \cdot \mu_{Bi}(y)$ (II) Layer 3: Nodes in this layer, known as the normalization layer, accept all nodes from the previous layer as the input value, as seen in Equation 3, and the normalization value of each value is calculated.

$$\overline{w_i} = \frac{w_i}{\Sigma w_i} \tag{III}$$

Layer 4: Each node in this layer, known as the clearing layer, is now named as the result parameter by calculating as in Equation 4.

 $\overline{W_{i}} \cdot f_{i}$ Layer 5 : It is known as the total layer and as seen in Equation 5, a single node in this layer is concluded as the output value of the values from the whole model.

$$x_0 = \frac{\sum w_i \cdot f_i}{\sum w_i} \tag{V}$$

ANFIS uses least-squares estimation methods [23] with back propagation [24] or hybrid learning algorithm as a learning method [19]. The error rate of learning methods is based on back-spreading.

The Breast Cancer Coimbra dataset, published on the UCI Machine Learning Repository website, consists of 116 samples of Glucose, Insulin, HOMA, Leptin, Adiponectin, Resistin, MCP-1, Age and Body Mass Index attributes [10].

Artificial intelligence and sub-branch studies, the data set has the right attributes to be trained as a result of the model can be obtained efficiently from the model. For this reason, the best features of the data set were used in ANFIS management, and the Boruta R Package, a feature selection (Feature Selection) [25] package for the r programming language using the Boruta Algorithm [26], was used [27]. As a result of this feature, the most important attribute of the data set which has 9 attributes is 5 and the details are shown in Figure 4. The five most important attributes shown in the figure are: Age, BMI, Glucose, HOMA and Resistin.



Figure 4 : Top 5 features of the data set resulting from attribute extraction.

In this study, anfis plug-in developed by Jang in Matlab software was used for ANFIS model in 1993. The error rate of the new data set consisting of Age, BMI, Glucose, HOMA and Resistin values is calculated with this extension.

#### **3. EXPERIMENTAL RESULTS**

In this study, Matlab R2018a version was used to perform the ANFIS network. The data set is divided into two as a training and test set, with a random ratio of 85 percent. As seen in Figure 6, the designed system has 5 inputs and 1 output according to the 5 independent attributes determined by the feature extraction method. The dataset is created as a column matrix and the first five columns are the input values of the system, while the last column consists of the output values.

(IV)



Figure 6 : Output of the network prepared for the processing process.

In the first step of the network, five independent variables are entered into the system. In the second stage, these input values are assigned to the 3 membership functions by a system of membership. In the next column, there are 243 rules using membership functions and a value is generated from each rule. Thanks to the resulting output, the membership function values have been converted to a single output value.

Figure 7 shows the general structure of the neural fuzzy inference system of the breast cancer prediction model.



Figure 7 : Fuzzy Inference System for breast cancer prediction model.

Figure 8 shows the graphs of all membership functions of this fuzzy inference system. Accordingly, the age value [24 86], the BMI value [18.37 36.79], the Glucose value [70 201], the HOMA value [0.4674 25.05] and the Resistin value [3.21 82.1].





Equation 6 was used to test the accuracy of the model. Here, the accuracy of the model must be between 0 and 1. With Target, the value of the data set after the test training and the threshold if the data set represents the error rate after training.

$$Accuracy = \frac{\sum_{i=1..N} (1 - (target_i - thershold(f(\overline{x_i}))))^2}{N}$$
(VI)

The membership functions in the matlab - anfis plugin in Table 1; Triangle membership function (trimf), trapezoid membership function (trapmf), bell shaped membership function (gbellmf), full symmetric gauss membership function (gaussmf), gauss membership function (gauss2mf),  $\pi$  membership function (pimf), full symmetric sigmodial membership

function (dsigmf) and sigmodial membership function. According to the table, the method used is hybrid and the error tolerance is 0, the membership function (mF) 3 and the number of cycles (epoch) 100.

| Membership<br>Function<br>Type<br>(MF Type) | Membership<br>Function<br>Dimensions<br>(Number of<br>MF) | Error<br>Tolarance | Epoch | Option<br>Method | Epochs<br>Error | Testing<br>Error | Accuracy |
|---|---|--------------------|-------|------------------|-----------------|------------------|----------|
| Trimf                                       | 33333   | 0                  | 100   | Hybrid           | 0.1937          | 2.2793           | 91.5004  |
| Trapmf                                      | 33333   | 0                  | 100   | Hybrid           | 0.2921          | 4.3079           | 93.2185  |
| Gbellmf                                     | 33333   | 0                  | 100   | Hybrid           | 0.2114          | 1.9348           | 89.0722  |
| Gaussmf                                     | 33333   | 0                  | 100   | Hybrid           | 0.1968          | 1.9148           | 89.7174  |
| Gauss2mf                                    | 33333   | 0                  | 100   | Hybrid           | 0.2336          | 1.5478           | 84.9024  |
| Pimf  | 33333   | 0                  | 100   | Hybrid           | 0.3008          | 12.1698          | 97.5278  |
| Dsigmf                                      | 33333   | 0                  | 100   | Hybrid           | 0.1947          | 4.4657           | 95.6394  |
| Psigmf                                      | 33333   | 0                  | 100   | Hybrid           | 0.1947          | 4.4657           | 95.6394  |

Table 1. Hybrid values obtained by ANFIS function.

In Figure 9, it is seen that the actual output values in the test data set overlap with the predicted values at a rate of 97.5%. The training error of the created model (epochs error) is 0.30085 in RMSE.



Figure 9 : Estimation of the actual output values and the estimation values for the test data set.

# 4. CONCLUSION

Many artificial intelligence algorithms have been developed on the classification and selection of breast cancer, one of the most common causes of death diagnosis among women. Genetic algorithm, fuzzy logic, artificial neural networks, image processing methods are frequently used in these developed algorithms. The purpose of developing these algorithms is to diagnose the disease on real cases. This is a solution for estimation of high accuracy rates.

In this study, a high performance was achieved by using the ANFIS algorithm on the data set for diagnosis of breast cancer. Breast cancer prediction was performed from the error rate results. When the results were compared, the error rates range from 0.193 to 0.30, while the test error rates were between 1.54 and 12.16.

When the data obtained from the study were analyzed, it was observed that the error values belonging to the membership function  $\pi$  gave the best results as a result of the accuracy calculation. The data obtained from the other functions varied between 84.9% and 97%.

In conclusion, it is concluded that ANFIS method will be successful in order to make a successful interpretation about the diagnosis of the disease on the selected breast cancer. It will be able to make future diagnostics with ANFIS model which is formed after obtaining input variables with different scenarios in future studies. Furthermore, thanks to the network structure of ANFIS, much more successful estimation results can be obtained from the data size of the sample numbers than the data size.

#### **5. REFERENCES**

- [1] Goldberg, David E., and John H. Holland. "Genetic algorithms and machine learning." Machine learning 3.2 (1988): 95-99.
- [2] Bench-Capon, Trevor JM, and Paul E. Dunne. "Argumentation in artificial intelligence." Artificial intelligence 171.10-15 (2007): 619-641.
- [3] Patrício, Miguel, et al. "Using Resistin, glucose, age and BMI to predict the presence of breast cancer." BMC cancer 18.1 (2018):
   29.
- [4] Harrell, Frank E. "Ordinal logistic regression." Regression modeling strategies. Springer, Cham, 2015. 311-325.
- [5] Díaz-Uriarte, Ramón, and Sara Alvarez De Andres. "Gene selection and classification of microarray data using random forest." BMC bioinformatics 7.1 (2006): 3.
- [6] Joachims, Thorsten. "Text categorization with support vector machines: Learning with many relevant features." European conference on machine learning. Springer, Berlin, Heidelberg, 1998.
- [7] Peczak, P., Alan M. Ferrenberg, and D. P. Landau. "High-accuracy Monte Carlo study of the three-dimensional classical Heisenberg ferromagnet." Physical Review B 43.7 (1991): 6087.
- [8] Lobo, Jorge M., Alberto Jiménez-Valverde, and Raimundo Real. "AUC: a misleading measure of the performance of predictive distribution models." Global ecology and Biogeography 17.2 (2008): 145-151.
- [9] Crisóstomo, Joana, et al. "Hyperresistinemia and metabolic dysregulation: a risky crosstalk in obese breast cancer." Endocrine 53.2 (2016): 433-442.
- [10] Breast Cancer Coimbra Data Set : https://archive.ics.uci.edu/ml/datasets/Breast+Cancer+Coimbra#
- [11] Song, Hee-Jun, Seon-Gu Lee, and Gwi-Tae Park. "A methodology of computer aided diagnostic system on breast cancer." Control Applications, 2005. CCA 2005. Proceedings of 2005 IEEE Conference on. IEEE, 2005.
- [12] Das, Arpita, and Mahua Bhattacharya. "GA based neuro fuzzy techniques for breast cancer identification." Machine Vision and Image Processing Conference, 2008. IMVIP'08. International. IEEE, 2008.
- [13] Huang, Mei-Ling, et al. "Usage of case-based reasoning, neural network and adaptive neuro-fuzzy inference system classification techniques in breast cancer dataset classification diagnosis." Journal of medical systems 36.2 (2012): 407-414.
- [14] Fatima, Bekaddour, and Chikh Mohammed Amine. "A neuro-fuzzy inference model for breast cancer recognition." International Journal of Computer Science & Information Technology 4.5 (2012): 163.
- [15] Shleeg, Alshalaa A., and Issmail M. Ellabib. "Comparison of Mamdani and Sugeno fuzzy interference systems for the breast cancer risk." International Journal of Computer, Information Science and Engineering 7.10 (2013): 387-391.
- [16] Lalli, G., et al. "A development of knowledge-based inferences system for detection of breast cancer on thermogram images." Computer Communication and Informatics (ICCCI), 2014 International Conference on. IEEE, 2014.)
- [17] Zurada, Jacek M. Introduction to artificial neural systems. Vol. 8. St. Paul: West publishing company, 1992.)
- [18] Ross, Timothy J. Fuzzy logic with engineering applications. John Wiley & Sons, 2005.
- [19] JANG, J.-S. R. (1993), "ANFIS: Adaptive-Network-Based Fuzzy Inference System", Ieee Transactions on Systems, Man and Cybernetics, 23(3), 665-685.
- [20] Wang, Hua O., Kasuo Tanaka, and Mark Griffin. "Parallel distributed compensation of nonlinear systems by Takagi-Sugeno fuzzy model." Fuzzy Systems, 1995. International Joint Conference of the Fourth IEEE International Conference on Fuzzy Systems and The Second International Fuzzy Engineering Symposium., Proceedings of 1995 IEEE Int. Vol. 2. IEEE, 1995.
- [21] Yılmaz M., Çomaklı Ö. and Haşıloğlu, A.S., 2002. Kanallarda zamana bağlı zorlanmış ısı taşınımının bulanık-sinir ağı (neurofuzzy) ile tahmini, GAP IV. Mühendislik Kongresi Bildiriler Kitabı, 06-08 Haziran 2002, Şanlıurfa, Turkey.
- [22] Balzar, Davor, and Hassel Ledbetter. "Voigt-function modeling in Fourier analysis of size-and strain-broadened X-ray diffraction peaks." Journal of Applied Crystallography 26.1 (1993): 97-103.
- [23] Pradhan, A. K., A. Routray, and Abir Basak. "Power system frequency estimation using least mean square technique." IEEE transactions on power delivery 20.3 (2005): 1812-1816.
- [24] Hagan, Martin T., and Mohammad B. Menhaj. "Training feedforward networks with the Marquardt algorithm." IEEE transactions on Neural Networks 5.6 (1994): 989-993.
- [25] Guyon, Isabelle, and André Elisseeff. "An introduction to variable and feature selection." Journal of machine learning research 3.Mar (2003): 1157-1182.
- [26] Kursa, Miron B., and Witold R. Rudnicki. "Feature selection with the Boruta package." J Stat Softw 36.11 (2010): 1-13.
- [27] Package 'Boruta' : https://cran.r-project.org/web/packages/Boruta/Boruta.pdf [Erişim Tarihi : 04.01.2019]



# **ORAL PRESENTATION**

# Novel Deposition Techniques in Memristor Fabrication

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**Abstract:** Thin film deposition represents one of the critical processes in the integrated circuit and memory device fabrication. With the growing interest in memristor technologies, future electronics have the tendency to utilize memristors in data storage and neuromorphic circuits. Memristor consists of an active layer sandwiched by two metal electrodes. The characteristics of each layer determine the operational capabilities and endurance of the device. Therefore, development of a robust fabrication scheme is crucial to extend the use of memristors. In this research, a memristor fabrication technique that combines wet atomic layer deposition (W-ALD), electrodeposition and sputtering will be presented with the evaluation of advantages and disadvantages. The active layer is determined to be TiOx and anodic electrodeposition on Ti is performed for the TiOx thin film deposition. In order to transfer the fabrication scheme from the laboratory environment to the industry, especially the oxygen content in the TiOx needs to be reproducible and anodic electrodeposition provides advantages in terms of cost and reproducibility. Pt, Cu, and Al metals are worked out at the bottom and top electrodes of the memristor. Pt is not oxidized easily during the operation of the memristor; therefore, it is extensively worked out in the literature. Wet ALD and galvanic displacement processes are demonstrated to fabricate Pt electrodes. The use of Pt is compared with the use of other metals, in terms of cost and feasibility in fabrication. In the presentation, particular emphasis will be given to the W-ALD and galvanic displacement for the deposition of noble metals. Consequently, the study presented here demonstrates several process flows for memristor fabrication.

Keywords: Nanofabrication, Memristor, Resistive Switching, Electrodeposition, Galvanic Displacement.



# Comparison of Short and Long Termed Asr Test Methods in Grain Dispersion Optimized Fly Ash Mortar Bars

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**Abstract:** Reinforced concrete has an important role in today's buildings. Reinforced concrete elements lose strength with physical and chemical harmful effect if the structural elements are unprotected or bad designed. One of the harmful effects is Alkali-Silica Reaction (ASR) proceeding by the forming of silica gels produced from the reaction of alkali oxides and reactive aggregate in cement used in the production of concrete. Those silica gels have a high capacity of water absorption properties. Gels get expanded in the humid environment by absorbing water and cause stress in the concrete. Cracks occur in the concrete as a result of those gels and deteriorating starts. These reactions name as an alkali-silica reaction. ASTM C 1260 is being the most preferred standard in ASR studies because of its short term duration to get results. ASTM C 1260 and ASTM C 227 test methods used to measure the alkaline activity are compared by preparing cement mortar bars containing optimized fly particle size distribution. Test specimens containing up to 20% replacement with and without optimized fly ash particle size distribution (OFA) are prepared separately for two of the test methods. Those specimens, stored in 80±2°C 1N NaOH solution for 14 days in compliance with ASTM C 1260, were kept in 38°C water for 360 days curing while the specimens were avoided from any contact to water in compliance with ASTM C 227.

As a result, plain specimens (HSN, NSN) are considered as harmful ASR according to both of the test methods. While the specimens HN5, HN10, HO5 are found acceptable region according to ASTM C 1260, the samples NN5, NN10, NO5 are determined as harmful region according to ASTM C 227. The specimens HN15, NN15, HN20, NN20, HO10, NO10, HO15, NO15, HO20, NO20 are determined harmless region according to both of the methods.

Keywords: Alkali-Silica Reaction, Fly ash, Particle Size Distribution, Length Change, ASTM C 1260, ASTM C 227

# **1. INTRODUCTION**

Reinforced concrete has an important role in today's buildings. Reinforced concrete elements lose strength with physical and chemical harmful effect if the structural elements are unprotected or bad designed. One of the harmful effects is Alkali-Silica Reaction (ASR) proceeding by the forming of silica gels produced from the reaction of alkali oxides and reactive aggregate in cement used in the production of concrete. Those silica gels have a high capacity of water absorption properties. Gels get expanded in the humid environment by absorbing water and cause stress in the concrete. Cracks occur in the concrete as a result of those gels and deteriorating starts. These reactions name as an alkali-silica reaction. In order to examine the effect of ASR, ASTM C 227, which is a 1-year test method, was used in the early 1950s. ASTM C 1260, results in just 14 days, was officially accepted in 1994 (Rogers, 1999). In the studies (Andiç and Ramyar, 2004), the effect of fly ash and blast furnace slag on the alkali silica reaction was investigated. As a result of these studies, it was observed that blast furnace slag and fly ash reduced the effect of ASR, but fly ash was more effective in reducing ASR effect than blast furnace slag. Yıldırım et al. stated that ASR formation is less in the mortar bars having fly ash than having blast furnace slag. They used ASTM C 227 test methods in their study about the ASR on crushed aggregate produced in Sakarya region. According to the results obtained from both methods, specimens classified as the harmless region.

To this end, ASTM C 1260 and ASTM C 227 standard test methods were compared on cementitious mortar bar having with and without optimized fly ash replacement.

### 2. MATERIAL AND METHODS

#### Material

In this study, cementitious mortar bars are prepared with fly ash as obtained from the factory, and particle size distributions (PSDs) of fly ash optimized for 5%, 10%, 15% and 20% additive ratios. Fuller-Thompson (year) have made a correlation for PSD optimization of aggregates. This correlation is given in the Equation 1. This method was carried out for fly ash particle size distribution.

$$P(D) = \frac{D^{q}}{D_{maks}^{q}} x100$$
 (1)

Where;

P(D) : Total percentage of material smaller than the diameter of the sieve opening

D : is the diameter of sieve opening

 $D_{maks} \quad : is the maximum particle diameter of fly ash$ 

q : is distribution modulus

Sevim and Demir (2019a and 2019b) obtained the best mechanical, durability and physical properties for q=0.4. q=0.4 dispersion modulus has been preferred in this study, too. Maximum particle size diameter of fly ash is determined  $63\mu m$  and required PSD analysis made according to the equation and results are given in Table 1. Table 2 lists the mortar mixtures' coding for those produced with and without fly ash having optimized.

Table 1. Required quantities for fly ash PSD (g)

| Additive Ratio | )     |       | Sieve Size | Sieve Size (µm) |       |  |
|----------------|-------|-------|------------|-----------------|-------|--|
| (%)            | 0-25  | 25-38 | 38-45      | 45-53           | 53-63 |  |
| 5              | 15.20 | 2.77  | 1.26       | 1.30            | 1.47  |  |
| 10             | 30.40 | 5.54  | 2.52       | 2.60            | 2.94  |  |
| 15             | 45.60 | 8.31  | 3.77       | 3.90            | 4.41  |  |
| 20             | 60.80 | 11.09 | 5.03       | 5.2             | 5.88  |  |

Table 2. Codes of fly ash-blended cement mortar mixtures for different properties

| Properties                     | Mixture ID. |
|--------------------------------|-------------|
| Plain, ASTM C1260              | HSN         |
| Fly Ash, ASTM C 1260           | HN          |
| Optimized Fly Ash, ASTM C 1260 | НО          |
| Plain, ASTM C 1227             | NSN         |
| Fly Ash, ASTM C 1227           | NN          |
| Optimized Fly Ash, ASTM C 1227 | NO          |

In this study, aggregate gradation analysis, given in ASTM C 1260 and ASTM C 227, were used. Aggregate gradation analysis, given in the both methods are the same, are given in Table 3.

 Table 3. Aggregate gradation analysis

| Sieve Diameter    | Cumulative Passing (%) |
|-------------------|------------------------|
| 4.75 mm - 2.36 mm | 100                    |
| 2.36 mm - 1.18 mm | 90                     |
| 1.18 mm - 600 μm  | 65                     |
| 600 μm - 300 μm   | 40                     |
| 300 μm - 150 μm   | 15                     |

Cement used in the study has more than 0.6% alkali oxide ratio as given in ASTM C 1260 and ASTM C 227 because the ASR effect is going to be examined. Chemical properties of the materials used in the study are given in the Table 4.

| Chemical Composition (%)       | Cement | Aggregate | FA    |
|--------------------------------|--------|-----------|-------|
| SiO <sub>2</sub>               | 18.79  | 73.05     | 57.11 |
| Al <sub>2</sub> O <sub>3</sub> | 5.05   | 7.31      | 19.27 |
| Fe <sub>2</sub> O <sub>3</sub> | 2.54   | 3.34      | 9.21  |
| CaO                            | 63.28  | 5.46      | 5.31  |
| MgO                            | 2.23   | 0.68      | 2.03  |
| K <sub>2</sub> O               | 0.83   | 1.49      | 2.39  |
| Na <sub>2</sub> O              | 0.28   | 0.06      | 0.64  |
| SO <sub>3</sub>                | 3.44   | -         | 0.13  |
| $Cr_2O_3$                      | 0.03   | -         | 0.02  |
| $Mn_2O_3$                      | 0.06   | -         | 0.08  |
| TiO <sub>2</sub>               | 0.26   | -         | 0.90  |
| Loss of ignition               | 3.20   | -         | 3.24  |
| $Na_2O+0.658 \times K_2O$      | 0.82   | -         | -     |

**Table 4**. Chemical properties of cement, aggregate and fly ash.

#### Methods

Aggregate/cement ratio is being given 2.25 in both standards, but while w/c ratio is given 0.6 in ASTM C 260, ASTM C 227 doesn't give defined ratio. According to ASTM C227, specimens have more than 0.1% length change expansion are classified "harmful", specimens whose length change expansion is below 0.1% are classified "harmless" in terms of ASR. According to ASTM C 1260, classification is as follows: length change expansion below 0.1% "harmless", between 0.1 and 0.2% "acceptable", over 0.2% "harmful" in terms of ASR.

## 3. RESULTS AND DISCUSSION

In this study, while length change expansions according to ASTM C 1260 were found as follows: HRN 0.210%, HN5 0.188%, HN10 0.112%, HN15 0.073%, HN20 0.044%, HO5 0.160%, HO10 0.087%, HO15 0.050%, HO20 0.005%; according to ASTM C 227, length change expansions according to ASTM C 1227 were found as follows: NRN 0.176%, NN5 0.125%, NN10 0.107%, NN15 0.097%, NN20 0.092%, NO5 0.113%, NO10 0.098%, NO15 0.090%, NO20 0.082%. Length change expansions of cementitious mortar bars having different fly ash additive ratio are given in Figure 1.

Plain specimens HSN and NSN were classified as harmful according to both test methods. While specimens HN5, HN10, and HO5 were found acceptable according to ASTM C 1260, specimens NN5, NN10, and NO5 were classified as harmful according to ASTM C 227. Specimens HN15, NN15, HN20, NN20, HO10, NO10, HO15, NO15, HO20, and NO20 were found harmless according to both test methods



Substitution Rate(%)

Figure 1. Length change expansions of cementitious mortar bars having different fly ash additive ratio.

# 4. CONCLUSION

As a result, when we compare these two test methods, there is a duration time difference caused by curing requirements. Besides their curing durations are different, their result evaluations are different too. While ASTM C 227 takes a long time about one year, it takes 14 days to get results with ASTM C 227 test method. According to the results got from this study, the difference between the two test methods is that ASTM C 227 classifies the specimens as acceptable but ASTM C 1260 classifies as harmful.

#### REFERENCES

- Rogers, C. A., Multi-laboratory study of the accelerated mortar bar test (ASTM C 1260) for alkali-silica reaction, Cement, concrete and aggregates, 21.2 (1999): 185-194.
- Funk, J.E., Dinger, D.R., Funk, J.E.J., Caol Grinding and Particle Size Distribution Studies for Coal Water Slurries at High Solids Content. Final Report, Empire State Electric Energy Research Corporation (ESEERCO), New York, 1980.
- Ramyar K., Andiç, Ö.; Uçucu Kül İnceliği ve Kullanım Oranının Alkali-Silis Reaksiyonuna Etkisi. Türkiye İnşaat Mühendisliği On Yedinci Teknik Kongre ve Sergisi, 2004.
- Yıldırım, K., & Sümer, M., Alkali Silika Reaksiyonunun Azaltılmasında Dört Farklı Mineral Katkı Kombinasyonunun Optimizasyonu. Sakarya University Journal Of Science, 22(5), 1-1, 2018.
- İpek, M., K. Yılmaz, and G. Sert. "Sakarya Bölgesinde Üretilen Kırmataş Agregaların Alkali Agrega Reaksiyonunun İncelenmesi, 5." Uluslar arası İleri Teknolojiler Sempozyumu (IATS '09), 2009, 13-15.
- Sevim, Ö., Demir, İ. (2019). Physical and permeability properties of cementitious mortars having fly ash with optimized particle size distribution. Cement and Concrete Composites, 96, 266-273.
- ASTM C227-03, Standard Test Method for Potential Alkali Reactivity of Cement-Aggregate Combinations (Mortar-Bar Method), ASTM International, West Conshohocken, PA, 2003.
- ASTM C1260-14, Standard Test Method for Potential Alkali Reactivity of Aggregates (Mortar-Bar Method), ASTM International, West Conshohocken, PA, 2014.



# Estimation of External and Internal Indices for Radiological Evaluation of the Use of Soils as a Structural Building Material

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Abstract: Structural building materials such as soil, brick, concrete etc. used in homes, schools and workplaces contain naturally occurring radionuclides. Building materials are a secondary source of radon in indoor environments. Soil containing natural radionuclides (U-238, Th-232, Ra-226, Rn-222, K-40 etc.) is a direct radiation source in the environment. Hence, it is importance to evaluate the usability of soil as a building material from a radiological point of view because of its major influence on the terrestrial ecosystem and humans. In this study, external (radium equivalent activity index and activity concentration index) and internal indices (alpha index and internal hazard index) related to exposure to members of the public living in Karabük city center located in Western Black Sea of Turkey were estimated for forty soil samples. The estimated values of all indices were compared with the international recommended limits or criteria. The results reveal that the average values of indices do not exceed the recommended limits.

Keywords: Soil, natural radioactivity, radiological hazards, external and internal risk indices, building materials

## **1. INTRODUCTION**

Individuals may be exposed to ionizing radiation emitted from natural radionuclides (primordial and cosmogenic radionuclides) in two different ways: (1) individuals may be exposed to gamma-rays emitted from the radionuclides in indoor and outdoor, which is called external irradiation and (2) individuals may be exposed to ionizing radiation such as alpha and beta emitted from the radionuclides through respiration (such as inhalation of radioactive gas) and digestion (such as the consumption of foods containing natural radionuclides), which is called internal irradiation (UNSCEAR 2008). Natural primordial radionuclides, whose half-times are comparable to the age of the world, are in the uranium (U-238) and thorium (Th-232) natural radioactive series, and radioactive potassium isotopes (K-40) (UNSCEAR 2008). Also, building materials are also a secondary source of radon in indoor environments such as homes, school and work places. Long-term exposure to radon and its short-lived decay products increase the risk of the lung cancer.

In this study, radium equivalent activity index, activity concentration index as external indices and alpha index and internal hazard index as internal indices were estimated to assess the radiation dose to individuals living in Karabük city center located in Western Black Sea of Turkey and usage of soil samples as building materials from a radiological point of view. The results were compared with criteria or limit values.

## 2. MATERIALS AND METHODS

Forty soil samples were collected from Karabük city is located in the Western Black Sea region in the Northwest of Turkey. The radioactivity measurements were carried out by using a gamma-ray spectrometer with a high-resolution coaxial p-type vertical HPGe detector (ORTEC GEM50P4-83). The detector resolution is 1.9 keV at full width half maximum of the 1332.5 keV gamma-ray photopeak from <sup>60</sup>Co and has a relative efficiency of 50% relative to NaI (Tl) detector.

Radium equivalent activity index ( $Ra_{eq}$ ) was developed as a common radiological index to represent the activity concentrations of Ra-226, Th-232 and K-40 measured in the soil samples and is given as follows (Beretka and Mathew 1985):

$$Ra_{eq} = 370 Bq/kg \cdot \left(\frac{A_{Ra}}{370 Bq/kg} + \frac{A_{Th}}{259 Bq/kg} + \frac{A_{K}}{4810 Bq/kg}\right)$$
(1)

where  $A_{Ra}$ ,  $A_{Th}$  and  $A_K$  are the activity concentrations of Ra-226, Th-232 and K-40 in Bq/kg, respectively. The maximum value or upper limit of  $Ra_{eq}$  index for dwellings or homes must be less than 370 Bq/kg to limit the dose received to 1.5 mSv/y. Activity concentration index (I) was proposed by the European Commission to assess the excess gamma radiation originating from building materials. It is widely used at the investigation level for practical monitoring purposes and calculated as follows (EC 1999):

$$I = \frac{A_{Ra}}{300 \text{ Bq/kg}} + \frac{A_{Th}}{200 \text{ Bq/kg}} + \frac{A_K}{3000 \text{ Bq/kg}}$$
(2)

 $I \le 1$  corresponds to the annual effective dose less than or equal 1 mSv, while  $I \le 0.5$  corresponds to the annual effective less than or equal 0.3 mSv for materials used in bulk amounts such as soil, concrete, bricks etc. (EC 1999). Alpha index (I<sub>A</sub>), which can be used to assess the excess alpha radiation due to inhalation of radon escaped from construction materials, is calculated using by the next formula (Righi and Bruzzi 2006):

$$I_{A} = \frac{A_{Ra}}{200 \,\text{Bq}/\text{kg}} \tag{3}$$

When the activity concentration of  $^{226}$ Ra in a building material exceeds the value of 200 Bq/kg, it is possible that the radon exhalation from this material could cause indoor radon concentration exceeding 200 Bq/m<sup>3</sup>. It is very important to remark that there is no distinction between materials used in bulk materials and materials with restricted use in the alpha index determination (Righi and Bruzzi 2006). Internal hazard index (I<sub>IHI</sub>), which can be used to assess radiation hazard respiratory organs due to radioactive inert gas radon (Rn-222) and its short-lived secondary products is determined by the next formula (Krieger 1981):

$$I_{\rm IHI} = \frac{A_{\rm Ra}}{185 \,{\rm Bq/kg}} + \frac{A_{\rm Th}}{259 \,{\rm Bq/kg}} + \frac{A_{\rm K}}{4810 \,{\rm Bq/kg}}$$
(4)

For the safe use of materials in the construction of buildings,  $I_{IHI}$  should be less than unity.

#### 3. RESULTS AND DISCUSSION

#### Results

The values of radium equivalent activity index, activity concentration index, alpha index and internal hazard index estimated for soil samples are given in Table 1. The values of  $Ra_{eq}$ , I, I<sub>A</sub> and I<sub>IHI</sub> varied from 41 to 161 Bq/kg, 0.1 to 0.6, 0.1 to 0.5 and 0.2 to 0.7, respectively.

Table 1. The values of Raeq, I, IA and IIHI

| Sampla codo | External indi | ces | Internal indices |      |  |
|-------------|---------------|-----|------------------|------|--|
| Sample code | Raeq (Bq/kg)  | Ι   | IA               | IIHI |  |
| SOIL1       | 105           | 0.4 | 0.1              | 0.4  |  |
| SOIL2       | 83            | 0.3 | 0.1              | 0.3  |  |
| SOIL3       | 77            | 0.3 | 0.2              | 0.3  |  |
| SOIL4       | 161           | 0.6 | 0.5              | 0.7  |  |
| SOIL5       | 66            | 0.2 | 0.1              | 0.2  |  |
| SOIL6       | 92            | 0.3 | 0.1              | 0.3  |  |
| SOIL7       | 99            | 0.4 | 0.1              | 0.3  |  |
| SOIL8       | 67            | 0.3 | 0.1              | 0.2  |  |
| SOIL9       | 41            | 0.1 | 0.1              | 0.2  |  |
| SOIL10      | 85            | 0.3 | 0.1              | 0.3  |  |
| SOIL11      | 66            | 0.3 | 0.1              | 0.2  |  |
| SOIL12      | 158           | 0.6 | 0.2              | 0.6  |  |
| SOIL13      | 86            | 0.3 | 0.2              | 0.3  |  |
| SOIL14      | 104           | 0.4 | 0.2              | 0.4  |  |

Table 1 continued

| SOIL15 | 79  | 0.3 | 0.2 | 0.3 |
|--------|-----|-----|-----|-----|
| SOIL16 | 95  | 0.3 | 0.1 | 0.3 |
| SOIL17 | 77  | 0.3 | 0.2 | 0.3 |
| SOIL18 | 108 | 0.4 | 0.2 | 0.4 |
| SOIL19 | 145 | 0.5 | 0.2 | 0.5 |
| SOIL20 | 68  | 0.2 | 0.1 | 0.3 |
| SOIL21 | 78  | 0.3 | 0.1 | 0.3 |
| SOIL22 | 99  | 0.4 | 0.2 | 0.4 |
| SOIL23 | 104 | 0.4 | 0.2 | 0.4 |
| SOIL24 | 65  | 0.2 | 0.2 | 0.3 |
| SOIL25 | 123 | 0.5 | 0.1 | 0.4 |
| SOIL26 | 112 | 0.4 | 0.2 | 0.4 |
| SOIL27 | 101 | 0.4 | 0.2 | 0.4 |
| SOIL28 | 107 | 0.4 | 0.1 | 0.4 |
| SOIL29 | 131 | 0.5 | 0.2 | 0.4 |
| SOIL30 | 73  | 0.3 | 0.2 | 0.3 |
| SOIL31 | 45  | 0.2 | 0.1 | 0.2 |
| SOIL32 | 58  | 0.2 | 0.1 | 0.2 |
| SOIL33 | 72  | 0.3 | 0.2 | 0.3 |
| SOIL34 | 49  | 0.2 | 0.1 | 0.2 |
| SOIL35 | 90  | 0.3 | 0.2 | 0.3 |
| SOIL36 | 128 | 0.4 | 0.3 | 0.5 |
| SOIL37 | 60  | 0.2 | 0.1 | 0.2 |
| SOIL38 | 84  | 0.3 | 0.1 | 0.3 |
| SOIL39 | 84  | 0.3 | 0.1 | 0.3 |
| SOIL40 | 123 | 0.4 | 0.3 | 0.5 |

### Discussion

All known radiological parameters for external and internal exposure estimated for each soil sample to assess the radiological hazards associated with usage of soil samples as structural building materials. The average value of  $Ra_{eq}$ , I, I<sub>A</sub> and I<sub>IHI</sub> was found as 91 Bq/kg, 0.3, 0.2 and 0.3, respectively. These parameters should be used only as screening tools for ready-to-use building materials. Any actual decision on restricting the use of a material should be based on a separate dose assessment. Turkey applies a strict criterion based on a dose in the range 0.3-1 mSv/y for structural building materials. These values are lower than the limit or criterion value of 370 Bq/kg, 1, 1, and 1 for  $Ra_{eq}$ , I, I<sub>A</sub> and I<sub>IHI</sub>, respectively.

#### 4. CONCLUSION

Consequently, this study indicated that the examined soil samples used as building materials would not cause any significant radiological risk.

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#### REFERENCES

Beretka J & Mathew P J (1985). Natural radioactivity of Australian building materials, industrial wastes and by-product. Health. Physics, 48: 87-95.

EC (European Commission) 1999 Radiation protection 112- Radiological protection principles concerning the natural radioactivity of building materials. Directorate- General Environment, Nuclear Safety and Civil Protection.

Krieger R (1981). Radioactivity of construction materials. Betonwerk Fertigteil Technology, 47: 468-473.

Righi S & Bruzzi L (2006). Natural radioactivity and radon exhalation in building materials used in Italian dwellings. Journal of Environmental Radioactivity, 88: 158-170.



# Comparison of Short and Long Termed Asr Test Methods in Grain Dispersion Optimized Blast Furnace Slag Mortar Bars

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**Abstract:** Concrete is a building material consisting of cement, aggregate and water. If the alkaline ratio in cement is high and aggregate contains reactive silica within a humid environment, then alkali-silica gels occur. When alkali-silica gels occur, alkali-silica reaction (ASR) starts and ASR gels proceed. Those gels, causes internal stress and map type cracks in concrete, have a high capacity of water absorption. Serious degradation problems may occur if the structure isn't protected from ASR effect, which is one of the dangerous durability problems. It is used ASTM C 1260 and ASTM C 227 standard test methods for ASR detection. While ASTM C 1260 is mostly preferred since it has 14-days short test duration, ASTM C 227 is preferred less because it has 12-months test duration. In addition, the phase of preparing test specimens is the same in both methods, but the curing methods are different.

In this study, the ASR effect on cementitious mortar bars was studied according to ASTM C1260 and ASTM C227. All results obtained these methods were compared and evaluated. The particle size distribution (PSD) of granulated blast furnace slag (GBFS) was optimized by using the help of vacuum sieve and then, cementitious mortars, having granulated blast furnace slag replacement with and without particle size distribution optimization, were prepared. According to ASTM C1260, specimen length change expansion ratios can be below 0.1%, between %0.1-0.2%, and above 0.2% and these are classified as harmless, acceptable, and harmful, respectively. ASTM C227 classifies the length change expansion ratio of samples as below and above 0.1%, these are called as harmless and harmful, respectively.

**Keywords:** Alkali-Silica Reaction, granulated blast furnace slag, particle size distribution, ASTM C 1260, ASTM C 227, length change expansion

# 1. INTRODUCTION

The design and protection of reinforced concrete structures have an important place in terms of sustainability. Serious damages can occur due to bad design and durability problems. One of the durability problems is the alkali-silica reaction, which affecting concrete sustainability. The alkali-silica reaction occurs when the alkali-oxide content of cement is more than 0.6%, the aggregate used in concrete production is reactive and there is enough moisture in the environment which will start reactions. Alkali-silica reaction produces ASR gels causing cracks and therefore, loss of strength and permeability. These gels absorb a high amount of water, increase the concrete internal stress, and cause expansion, then results in a destruction in the concrete. ASTM C 1260 and ASTM C 227 test methods are being used for determining the ASR effect. While ASTM C 1260 is mostly preferred since it has 14-days short test duration, ASTM C 227 is preferred less because it has 12-months test duration. In addition, the phase of preparing test specimens is the same in both methods, but the curing methods are different.

Pozzolans are decreasing the Ph ratio by binding the CH gels. With the help of Ph, it is increased the solubility of the silica and prevented the occurrence of alkali-silica gel. In some studies, it's been observed that the use of 30% Class C fly ash and 5% silica powder reduced the ASR effect and GBFS can decrease the ASR effect in non-hazardous degrees. To this end, ASTM C 1260 and ASTM C 227 standard test methods were compared on cementitious mortar bar having with and without optimized granulated blast furnace slag replacement.

### 2. MATERIAL AND METHODS

#### Materials

In this study, cementitious mortar bars are prepared with granulated blast furnace slag as obtained from the factory, and particle size distributions (PSDs) of GBFS optimized for 5%, 10%, 15% and 20% additive ratios. Fuller-Thompson (1907) have made a correlation for PSD optimization of aggregates. This correlation is given in the Equation 1. This method was carried out for GBFS particle size distribution.

$$P(D) = \frac{D^{q}}{D_{maks}^{q}} x 100$$
(1)

Where;

P(D) : Total percentage of material smaller than the diameter of the sieve opening

D : is the diameter of sieve opening

 $D_{maks} \quad : is the maximum particle diameter of fly ash$ 

q : is distribution modulus

Sevim and Demir (2019a and 2019b) obtained the best mechanical, durability and physical properties for q=0.4. q=0.4 dispersion modulus has been preferred in this study, too. Maximum particle size diameter of fly ash is determined  $63\mu m$  and required PSD analysis made according to the equation and results are given in Table 1. Table 2 lists the mortar mixtures' coding for those produced with and without fly ash having optimized.

Table 1. Required quantities for GBFS PSD (g)

| Additive  |       |       | Sieve Size(µm) |       |       |
|-----------|-------|-------|----------------|-------|-------|
| Ratio (%) | 0-25  | 25-38 | 38-45          | 45-53 | 53-63 |
| 5         | 15.20 | 2.77  | 1.26           | 1.30  | 1.47  |
| 10        | 30.40 | 5.54  | 2.52           | 2.60  | 2.94  |
| 15        | 45.60 | 8.31  | 3.77           | 3.90  | 4.41  |
| 20        | 60.80 | 11.09 | 5.03           | 5.2   | 5.88  |

Table 2. Codes of GBFS-blended cement mortar mixtures for different properties

| Properties                  | Mixture ID. |
|-----------------------------|-------------|
| Plain, ASTM C1260           | AR          |
| GBFS, ASTM C 1260           | AN          |
| Optimized GBFS, ASTM C 1260 | AO          |
| Plain, ASTM C 1227          | LR          |
| GBFS ASTM C 1227            | LN          |
| Optimized GBFS, ASTM C 1227 | LO          |

In this study, aggregate gradation analysis, given in ASTM C 1260 and ASTM C 227, were used. Aggregate gradation analysis, given in the both methods are the same, are given in Table 3.

Table 3. Aggregate gradation analysis

| Sieve Diameter    | Cumulative Passing (%) |
|-------------------|------------------------|
| 4.75 mm - 2.36 mm | 100                    |
| 2.36 mm - 1.18 mm | 90                     |
| 1.18 mm - 600 μm  | 65                     |
| 600 μm - 300 μm   | 40                     |
| 300 μm - 150 μm   | 15                     |

Cement used in the study has more than 0.6% alkali oxide ratio as given in ASTM C 1260 and ASTM C 227 because the ASR effect is going to be examined. Chemical properties of the materials used in the study are given in the Table 4.

| Chemical<br>Composition (%)              | Cement | Aggregate | BFS   |
|--|--------|-----------|-------|
| SiO <sub>2</sub>                         | 18.79  | 73.05     | 42.4  |
| $Al_2O_3$                                | 5.05   | 7.31      | 12.56 |
| Fe <sub>2</sub> O <sub>3</sub>           | 2.54   | 3.34      | 1.03  |
| CaO                                      | 63.28  | 5.46      | 35.04 |
| MgO                                      | 2.23   | 0.68      | 5.11  |
| K <sub>2</sub> O                         | 0.83   | 1.49      | 0.75  |
| Na <sub>2</sub> O                        | 0.28   | 0.06      | 0.14  |
| $SO_3$                                   | 3.44   | -         | 0.37  |
| $Cr_2O_3$                                | 0.03   | -         | 0.01  |
| $Mn_2O_3$                                | 0.06   | -         | 0.95  |
| TiO <sub>2</sub>                         | 0.26   | -         | 0.86  |
| Loss of ignition                         | 3.20   | -         | -     |
| Na <sub>2</sub> O+0.658×K <sub>2</sub> O | 0.82   | -         | -     |

Table 4. Chemical properties of cement, aggregate and GBFS.

#### Methods

In order to determine the ASR effect, ASTM C 1260 and ASTM C 1227 test methods were used.

In these methods, while the mortar bar samples should be waited for 14-days in 1N NaOH solution in  $80\pm2$  oC according to ASTM C 1260, specimens should be waited over water with  $38\pm2$  oC, without any contact of water according to ASTM C 1227. Aggregate/cement ratio is being given 2.25 in both standards, but while w/c ratio is given 0.6 in ASTM C 1260, ASTM C 1227 doesn't give defined ratio. According to ASTM C 1227, specimens have more than 0.1% length change expansion are classified "harmful", specimens whose length change expansion is below 0.1% are classified "harmless" in terms of ASR. According to ASTM C 1260, classification is as follows: length change expansion below 0.1% "harmless", between 0.1 and 0.2% "acceptable", over 0.2% "harmful" in terms of ASR.

# 3. RESULTS AND DISCUSSION

## Results

In this study, while length change expansions according to ASTM C 1260 were found as follows: AR 0.210%, AN5 0.195%, AN10 0.169%, AN15 0.148%, AN20 0.107%, AO5 0.163%, AO10 0.147%, AO15 0.129%, AO20 0.077%. length change expansions according to ASTM C 1227 were found as follows: LR 0.176%, LN5 0.134%, LN10 0.124%, LN15 0.121%, LN20 0.107%, LO5 0.127%, LO10 0.122%, LO15 0.102%, LO20 0.095%. Length change expansions of cementitious mortar bars having different GBFS additive ratio are given in Figure 1.



Figure 1. Length change expansions of cementitious mortar bars having different GBFS additive ratio

#### 4. CONCLUSION

AR and LR were classified as harmful according to both of the methods. Samples prepared accordingly to ASTM C 1260, while AO20 was found harmless, the other samples were found acceptable in terms of ASR. . Samples prepared accordingly to ASTM C 1227, while LO20 was determined as harmless, the other samples were determined as harmful.

Cementitious mortar bar samples containing 20% GBFS replacement with optimized PSD were found harmless according to both methods, while other samples found as acceptable according to ASTM C 1260 and found as harmful according to ASTM C 227. These findings show that It can be the difference between the test methods.

#### REFERENCES

F. Bektaş, Preventive Measures Against Alkali-Silica Reaction. Yüksek Lisans Tezi, Orta Doğu Teknik Üniversitesi, Ankara, 2002.

Shehata, M.H., Shashiprakash, S.G., Thomas, M.D.A., "Alkali Aggregate Reaction and Fly Ash", Sixth NCB International Seminar on Cement and Building Materials, 1999.

Swamy, R.N., The Alkali-Silica Reaction in Concrete. Van Nostrand Reinhold,

#### New York, 1992.

- Tosun, K., Yazıcı, H., Baradan, B., "Uçucu Kül ve Silika Tozunun Alkali Silika Reaksiyonuna Etkisinin İncelenmesi", Türkiye İnşaat Mühendisliği 16. Teknik Kongresi, Ankara, 2001.
- ASTM C227-03, Standard Test Method for Potential Alkali Reactivity of Cement-Aggregate Combinations (Mortar-Bar Method), ASTM International, West Conshohocken, PA, 200.
- ASTM C1260-14, Standard Test Method for Potential Alkali Reactivity of Aggregates (Mortar-Bar Method), ASTM International, West Conshohocken, PA, 2014.
- Funk, J.E., Dinger, D.R., Funk, J.E.J., Caol Grinding and Particle Size Distribution Studies for Coal Water Slurries at High Solids Content. Final Report, Empire State Electric Energy Research Corporation (ESEERCO), New York, 1980.
- Sevim, Ö., Demir, İ. (2019). Physical and permeability properties of cementitious mortars having fly ash with optimized particle size distribution. Cement and Concrete Composites, 96, 266-273.
- Fuller, W.B., Thompson, S.E., The laws of proportioning concrete. ASCE J. Transport., 59, 67-143, 1907.



# **ORAL PRESENTATION**

# Dynamic Programming in Vehicle Routing Problems and its Application in the **Pharmaceutical Industry**

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Abstract: Due to the increasing number of vehicles nowadays, the intensity of the traffic is also an important problem in the urban transportation. The most important criterion in the pharmaceutical sector is to meet the needs of people in the shortest time and with the least cost. Vehicle routing problems help companies to offer the most suitable routes. The routing problems can find application area in pharmacies and service sector in addition to production enterprises such as iron and steel industry. In this case study, an alternative solution has been developed with the help of "dynamic programming" method to distribute the pharmaceuticals to pharmacies in Karabük. In this case, the aim is to decrease the transportation distance. In the route determined, the shortest distance is reached increasing the number of customers served.

Keywords: Vehicle routing, dynamic programming, pharmaceutical industry, urban transport, transportation

# **1. INTRODUCTION**

Logistics systems are in the supply chain from the first purchasing stage to the final stage of the distribution and storage of products or services. The logistics systems are associated with vehicle routing problems, as it consists of many constraints such as the delivery of products and services at minimum cost and distribution as soon as possible. Therefore, in order to find solutions to the problems encountered in logistics systems, vehicle routing problems have been used for a long time. Vehicle routing allows customers to distribute and pickup products or services from a specific or multiple warehouse. In order to respond to the needs of emerging and growing systems, the solutions to complex routing problems can also become quite long and complex. According to the complexity of the problem, genetic algorithms, artificial neural networks can be applied as the heuristic solution methods, dynamic programming, branch and bound are of the exact solution methods (Patir, 2009).

Dynamic programming is one of the exact solution methods of vehicle routing problems; it is a technique that aims to get the optimal solution within each group by dividing the problem into smaller groups. One of the most important features of this method is that each of the decisions given in the first stage in dynamic programming affects the decisions in the following stages. Dynamic programming method can be applied to a variety of problems such as continuous, discrete, time independent, linear and nonlinear. Using the dynamic programming method in vehicle routing problems which are NP-Hard provides save the solution time, some of the possible solution combinations, not eligible, are not considered in the solution, and dynamic programming algorithm save the solution time. These features reveal the advantages of dynamic programming method (Yılmaz, 1999).

Various problems such as budgeting, inventory problems, production planning can be solved by dynamic programming method in the production environment. In this study, the dynamic programming method is applied to a vehicle routing problem in pharmaceutical service sector rather than manufacturing sectors.

Vehicle routing problems are the problems of companies that they distribute the products or services one or several warehouses and a certain number of customers to minimize the travel distance. In vehicle routing problems, the distribution is started from the warehouse and the same route is monitored and returned to the warehouse (Atasagun, 2015). In general, some limitations can be added to this problem, such as vehicle capacity, path length, customers who need to be serviced at a specific time period. Vehicle routing problems are the problems of traveling salesman problems adding multiple tools and constraints.

In vehicle routing problems, several basic conditions should be met such as the customer demand, the total customer demand should be satisfied on a distribution route, the capacity of the vehicle should not be exceeded on that route (Düzakın, 2009).

Vehicle routing problems are encountered in our daily lives such as gasoline diesel distribution, money distribution, postal services, delivery of cargo companies, product distribution of raw materials to manufacturers and transportation of passengers by means of transportation (Keskintürk et al., 2015). In these problems, there is a flow from the producer to the customer, as well as a reverse flow from the customer to the manufacturer.

Nowadays, adding new constraints to the vehicle routing problems from the real life variates the classical vehicle routing problems which are not enough on meeting the needs of the complex real systems. The most common variant of the vehicle routing problem is the capacity limited vehicle routing problem. Medicine delivery to pharmacies within a certain time from drug stores can be given as an example of vehicle routing problems with time windows. In this type of problem, customers expect to receive services in a certain time. Especially in big cities, the traffic density occurring at certain times may cause some uncertainties on vehicle routing problems. These uncertainties have an impact on the arrival time or average speed of the vehicles and therefore the classical vehicle routing problems become insufficient under uncertainties. The distance constrained, periodic, stochastic, multi-warehouse, symmetric, asymmetric, open-ended, closed-ended, dynamic and static vehicle routing problem are some of the other types of the problems (Koç, 2012).

There are many methods used to solve vehicle routing problems. It is possible to collect these methods in three main title as exact methods, heuristic solution methods and meta-heuristic solution methods. Exact solution methods guarantee the best solution. However, the solution time increase incrementally depending on the size of the problem. In general, small and medium-sized problems can be solved practically (Keskintürk et al., 2015). The four well-known exact solution methods are branch and bound algorithm, branch and cut algorithm, cutting plane and dynamic programming. The well-known meta heuristic solution methods are particle swarm optimization, artificial bee colony, ant colony, simulated annealing, genetic algorithm and tabu search algorithm. In this study, we investigate the advantages of using dynamic programming approach.

The rest of the paper is organised as follows. In section 2, the vehicle routing problem encountered in pharmaceutical industry is introduced, the dynamic programming approach is applied to solve the problem and, the results of the numerical analysis are presented. In section 3, the managerial implications and the future research directions are discussed.

# 2. MATERIAL AND METHODS

The only distribution warehouse of a pharmaceutical company operating in Karabük meets the pharmaceutical needs of pharmacies on the daily customer demands. The company distributes medicines to four main regions: Safranbolu, 5000 Evler, 100. Y1 and Karabük (City centre). Karabük (City centre) has also divided into three regions. The company tries to provide services as soon as possible by combining the orders received from the automation systems of 67 pharmacies. In this study, an alternative solution is proposed to the medicine distribution of the pharmaceutical warehouse. The aim is to reach the shortest distance by increasing the number of customers served.

The solution approach is the dynamic programming method which is one of the exact solution methods of vehicle routing problems. Dynamic programming approach decompose the large problem into smaller sub-problems and creates an optimal solution for each part as the basis of subsequent parts, repeatedly (Ersöz, 2015). Each problem has its own unique structure; while starting the solution with the dynamic programming approach, the way in which the problems are decomposed, and the number of sub-problems can vary. The main reason of this variation is that the dynamic programming cannot be standard for all the problems. Therefore, the application of dynamic programming method in complex problems requires expertise (Avc1, 2012). Dynamic programming is a method that can produce solutions to the business problems such as cargo loading, marketing and investment, distribution and routing, inventory, capital budgeting, price strategy determination, production planning (Sevinç, 2008).

In our problem, the distribution regions are determined at first. Then, in order to start the solution with the dynamic programming approach, the distances of the regions to each other are presented in a distance matrix. The network diagram of Karabük is organised with the generated distances matrix. The distance matrix used in the application is given in Table 1.

Table 1. Inter-regional distances matrix (in meters)

| Counties   | Safranbolu | 5000 Evler | 100. Yıl | Merkez | 200 Evler | Esentepe |
|------------|------------|------------|----------|--------|-----------|----------|
| Safranbolu | _          | 3600       | 4500     | 8900   | 9400      | 8400     |
| 5000 Evler | 3600       | _          | 3300     | 6500   | 7000      | 6100     |
| 100. Yıl   | 4500       | 3300       | _        | 6100   | 6600      | 5600     |
| Merkez     | 8900       | 6500       | 6100     |        | 2600      | 2200     |
| 200 Evler  | 9400       | 7000       | 6600     | 2600   | —         | 1400     |
| Esentepe   | 8400       | 6100       | 5600     | 2200   | 1400      | _        |

Figure 1. The transportation networks



The network diagram in Figure 1 shows all the possible routes between the depot and the  $7^{th}$  region Safranbolu. To solve the problem by dynamic programming approach, the network is divided into three stages. Figure 2 shows the three stages of the problem and the transportation distances are calculated for each stage, separately.

Figure 2. The tree stages in the transportation network



In the dynamic programming approach, there are two kinds of solution techniques as forward iterations and backwards iterations. In this study, both the forward iterations and the backward iterations techniques are used as follows;

#### **Forward Iteration**

The general idea is to calculate the cumulative shortest distances from the initial node to the last node of the first stage and use the cumulative result as the input of the next step (Taha, 2000).

STEP 1: In the first step, the starting nodes 2, 3, and 4. are connected to nodes with a single arrow. The shortest distances between the nodes are determined as follows:

Shortest distance to node 2 = 2978 meters

Shortest distance to node 3 = 1473 meters

Shortest distance to node 4 = 3950 meters

STEP 2: In the second step, the shortest distances to the 5th and 6th nodes are determined cumulatively. At the beginning of the solution, before the 5th node, the 6th node is evaluated. There are three possible routes ((2,5), (3,5) and (4,5) to reach node 5 and there are two possible routes ((3,6), (4,6)) to reach the 6th node.

(Shortest distance to node 5) =  $\min \begin{cases} 7000 + 2978 = 9978 \\ 6500 + 1473 = 7973 \\ 6100 + 3950 = 10050 \end{cases}$  = 7973 meters (from node 3)

(Shortest distance to node 6) = min  $\begin{cases} 6100 + 1473 = 7573\\ 5600 + 3950 = 9550 \end{cases}$  = 7573 meters (from node 3)

STEP 3: In the last step, the arrival node is accessible from both the 5th and the 6th. The following data is obtained by considering the distances from the 2nd node and the 6th node.

(Shortest distance to node 7) = min  ${7973 + 3600 = 11573 \\ 7573 + 4500 = 12173} = 11573$  (from node 5) Thus, it has been achieved that the route 1-3-5-7 is the optimum route.

#### **Backward Iteration**

In the backward iteration technique, there is a reverse flow from the 7<sup>th</sup> node to the 1st node, against the forward iteration technique.

STEP 3: In step 3 (X4 = 7), for 5th node and 6th node (X3 = 5 and 6), there is no alternative route to choose because the nodes are connected only by one way, and the results of step 3 are summarized as follows:

| X3  | d (X3, X4) | Optimu  | ım Solution      |
|-----|------------|---|------------------|
|     | $X_4 = 7$  | <b>F</b> <sub>3</sub> ( <b>X</b> <sub>3</sub> ) | $\mathbf{X_4}^*$ |
| 5th | 3600 m     | 3600 m  | 7th              |
| 6th | 4500 m     | 4500 m  | 7th              |

Table 2. Solution table of step 3

STEP 2: There is no alternative route from node 2 to node 6 in step 2. Together with F3 (X3) in step 3, suitable alternatives are shown in the following table:

#### Table 3. Solution table of step 2

| $\mathbf{X}_2$ | <b>d</b> ( <b>X</b> <sub>2</sub> , <b>X</b> <sub>3</sub> ) | Optimum           | Solution  |     |
|----------------|--|-------------------|---|-----|
|                | $X_3 = 5 		 X_3 = 6$                                       |                   | <b>F</b> <sub>2</sub> ( <b>X</b> <sub>2</sub> ) | X3* |
| 2th            | 3600+7000=10600 m  | -                 | 10600 m   | 5th |
| 3th            | 3600+ 6500=10100 m   | 4500+6100=10600 m | 10100 m   | 5th |
| 4th            | 3600+6100=9700 m   | 4500+5600=10100 m | 9700 m  | 5th |

STEP 1: There are three possible routes from 1st stage (1,2), (1,3) and (1,4). The following table is obtained using the results found in step 2:

Table 4. Solution table of step 1

| <b>X</b> 1 |                    | $d(X_1, X_2) + F_2(X_2)$ |                   | Optimum S                                   | olution          |
|------------|--------------------|--------------------------|-------------------|---|------------------|
|            | $X_2 = 2$          | $X_2 = 3$                | $X_2 = 4$         | $\mathbf{F}_{1}\left(\mathbf{X}_{1}\right)$ | $\mathbf{X_2}^*$ |
| 1th        | 10600+2978=13578 m | 10100+1473=11573 m       | 9700+3950=13650 m | 11573 m                                     | 3th              |

The calculations based on the forward iteration technique and the backward iteration technique show that the shortest route is 1-3-5-7 and the shortest distance between the 1st and 7th nodes is 11573 meters.

#### **3.RESULTS AND DISCUSSION**

In the study, an alternative solution is proposed to the vehicle routing problem of a pharmaceutical warehouse in Karabük by dynamic programming approach. With this alternative solution, each vehicle serves increasing number customers with the shortest possible distance. In dynamic programming, a special effort is needed to solve the problems by using a suitable combination of programming parameters. The fact that the situations and decision variables take large values and it causes that the size of the scheduling problem increase considerably. The dynamic programming approach can not be applied by a general algorithm such as simplex method. Therefore, it is difficult to apply using commercial software programs. All these factors are some of the disadvantages encountered in the application of dynamic programming method (Özdemir, 2002).

Creating the network diagram during the application is difficult with the increasing number of decision variables. Due to the lack of a standard modelling of dynamic programming problems, in our approach, the problem is divided into the lower stages and the optimal route is confirmed by using both forward and backward recursion techniques together. Although the dynamic programming provides an optimal solution for the small sized problems, it can be supported by an algorithm integrating several real-life conditions of the vehicle routing problem for industrial applications.

# 4. REFERENCES

- Avcı, K., 2012. Dinamik Programlama Tekniğinin Çok Aşamalı Stok Kontrol Problemlerine Tatbik Edilmesi, Yüksek Lisans Tezi, İstanbul Üniversitesi Sosyal Bilimler Enstitüsü, İstanbul.
- Atasagun, G.C., 2015. Zaman Bağımlı Eş Zamanlı Topla Dağıt Araç Rotalama Problemi, Yüksek Lisans Tezi, Selçuk Üniversitesi Fen Bilimleri Enstitüsü, Konya.
- Düzakın, E. ve Demircioğlu, M., 2009. Araç Rotalama Problemleri ve Çözüm Yöntemleri, Çukurova Üniversitesi İİBF Dergisi, 13(1): 68-87.
- Ersöz, G., 2015. Çok Amaçlı Dinamik Programlama Modelinin Ulaştırma Sorunlarına Uygulanması, Yüksek Lisans Tezi, Dokuz Eylül Üniversitesi Sosyal Bilimler Enstitüsü, İzmir.
- Keskintürk, T., Topuk, N. ve Özyeşil, O., 2015. Araç Rotalama Problemleri ile Çözüm Yöntemlerinin Sınıflandırılması ve Bir Uygulama, İşletme Bilimi Dergisi, 3(2): 77-107.
- Koç, Ç., 2012. Zaman Bağımlı Araç Rotalama Problemi, Yüksek Lisans Tezi, Selçuk Üniversitesi Fen Bilimleri Enstitüsü, Konya.
- Yılmaz, E., 1999. Dinamik Programlama Yöntemi ve Üretim Planlama Probleminde Bir Uygulama, Yüksek Lisans Tezi, İstanbul Üniversitesi Sosyal Bilimler Enstitüsü, İstanbul.
- Özdemir, A., 2002. Envanter Sorunlarının Çözümünde Dinamik Programlama Modelinin Uygulanması, Yüksek Lisans Tezi, Dokuz Eylül Üniversitesi Sosyal Bilimler Enstitüsü, İzmir.
- Patır, S., 2009. Dinamik Programlama ve Bir Ecza Deposunun Şehir İçi İlaç Dağıtımına Alternatif Bir Çözüm Önerisi", İktisadi ve İdari Bilimler Dergisi, 23(2): 63-79.
- Sevinç, C., 2008. Lojistik Planlama Çok Ürünlü Çok Kademeli İkmal Zincirindeki Depolarda Bulundurulması Gerekli Minimum Malzeme Miktarının Dinamik Programlama Yöntemiyle Optimizasyonu, Yüksek Lisans Tezi, Dokuz Eylül Üniversitesi Sosyal Bilimler Enstitüsü, İzmir.
- Taha, A. H. 2000. Yöneylem Araştırması (1.Baskı). (Çevirenler: Ş. Alp Baray, Şakir Esnaf). İstanbul: Literatür Yayıncılık.



# Failure Mode and Effects Analysis: An Application in the Textile Industry

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Abstract: In this study, Failure Mode and Effects Analysis (FMEA) which is a technique widely applied in manufacturing enterprises, has been discussed. FMEA is a technique that enables the system to detect and prevent any failures, faults and problems that may occur in a system before it reaches the customer. With the analysis, the risk priority coefficients for the failures are calculated. The production processes for the ready-made clothing is monitored and an FMEA application is made in the Textile Industry. Considering the results of the analysis, the inner arm, elbow press, and pat pat press failures are pointed out. In order to eliminate these failures, an automation system suitable for improving the quality functions and the correct use of the machines in the relevant band are suggested for the employees trained. With the suggestions, we aim to improve the quality and process functions of the company significantly and to minimize the failures caused by the carelessness.

Keywords: Failure mode and effect analysis (FMEA), quality, process improvement, process FMEA, textile industry

# **1. INTRODUCTION**

The ready-to-wear is a sector that has kept the economy alive and has a significant effect in the development of the country. In the competitive production environment, all companies should give attention to the concept of quality and policies. In the past, quality has been applied with traditional methods in production or service, however nowadays, the rational techniques are used to reach the solution of problems, systematically.

Statistical methods such as Pareto Analysis and Control Graphics are common techniques in quality and process improvement. In addition, changing, developing standards and increasing problems reveal the need for the different techniques. Since FMEA identifies potential failures, the causes and effects of the failures, extracts failure priorities, facilitates problem tracking, it is well-known among the techniques used in quality and process improvement.

In this study, FMEA technique is explained at first. In the industrial application, the number of the failures and the causes of the failures have been determined, the scores have been given for the Risk Priority Number (RPN) and managerial implications are discussed. As a result of the application, the severity, frequency and catchability of the failures are decreased.

As an example, Aircraft Model 299 of the US Army in 1935, was hard to use and a complex equipment. After the first flight, the plane crashed, and two of the five crew members died. The checklists started to be used in order to decrease the complexity. On the first flight after the checklist application, the pilots flew 1.8 million miles. As another example, John Hopkins Hospital in the United States used control lists in order to prevent catheter-related infections. The control lists reduced the rate of catheter-related infections from 11% to zero in the first ten days of the application. 43 infections and 8 deaths are prevented, and two million dollars was saved. With the checklists used in the medical and military fields, the inattention was prevented. Textile is a labour-intensive sector. In our study, it is stated that the digital checklists can be used in an automation system and can reduce the failure rates caused by the carelessness of the employees. We believe that the application of Industry 4.0 applications, which become widespread nowadays, can be applied in the labour-intensive sectors (Gawande, 2011).

In the literature, a piston-making manufacturing process and its failures are considered by FMEA technique and the reasons of failures are found. The effects of failure types are scored over 10 points in terms of severity, catchability and frequency factors. As a result, risk priority (RPN) values of the failure types are calculated. In order to reduce the RPN values according to the causes and effects of these failures, several suggestions on the problem solutions are presented and it has been shown that the technique improve the company's quality functions (Çevik and Aran, 2009). In line with

Çevik and Aran 2009, the failures in the textile sector are examined, Process FMEA technique is applied and effects are estimated in our study.

In our case study, an FMEA team is created and the factors that produce failures in the enterprise are identified. The elimination of planting failures from these factors is determined in a priority. FMEA steps are organised to correct sewing failures. The production process is monitored during 12 weeks after the application. As a result of the suggestions, 4.1% plant failure in the jean pants production line and 5.2% sewing failure in the shirt production line is achieved. In addition, 966.3 minutes gain is obtained at the time of troubleshooting. Further, in order to minimize these failures, an automation system should be used considering the warnings of the employees (Yücel, 2006).

# 2. MATERIAL AND METHODS

Textile and ready-to-wear sector are considered for FMEA application. The aim of FMEA is to increase the competitiveness of quality and production efficiency by identifying the most common failures in the production process and focusing on the common failures. The failures generated during jacket production are studied. Risk Priority Number (RPN) is calculated by scoring frequency, catchability and severity of the failures. The failures are prioritized and then the reasons for the failures are discussed. The failures on the production process are: A, the inner arm; B, elbow press; C, pat pat press; D, Stain; E, rope draw; F-arm sewing; G, the front arm; H, the arm breaks; I, attachment; J, back-side seams; K, the back arm; L strap.

There are four FMEA types: FMEA, Design FMEA, Service FMEA, Process FMEA. Most of the failures in the production process are caused by the employee. Process FMEA is used in order to prevent and analyse the failures in the production process. Process FMEA enables us to analyse the processes, tools, processes and the effects of manpower and to determine their weaknesses.

The following steps are; (i) The type of FMEA to be determined. (ii) A process flow chart is created. (iii) Potential failures are identified for each process. (iv) The causes of the failure are determined for each failure. (v) The severity, frequency of occurrence and catchability of each failure determined and scored. (vi) Risk Priority Number is calculated by multiplying the determined scores. (vii) Solutions that eliminate the prioritized failures are followed in line with Sabir and Bebekli, 2015.

At the beginning of the study, the data is gathered in 2 weeks. Based on the data, Pareto Diagram is drawn to prioritize failures.



**Figure 1.** Pareto Diagram for failures in jacket production **Table 1.** Failures ordered by RPN values

| FAILURE EFFECT VALUES                 | F1 | С | F2 | RPN |
|---------------------------------------|----|---|----|-----|
| the Inner Arm                         | 5  | 8 | 9  | 360 |
| Elbow Press                           | 5  | 8 | 7  | 280 |
| Outside The Stitching Of The Arm Part | 5  | 9 | 6  | 270 |
| Stitching Of Arm Compartment Burst    | 6  | 9 | 5  | 270 |
| Pat Pat Press                         | 5  | 8 | 6  | 240 |
| Fastening                             | 6  | 8 | 5  | 240 |

| Stain                 | 6 | 5 | 6 | 180 |
|-----------------------|---|---|---|-----|
| Injury                | 6 | 3 | 5 | 90  |
| Rear Arm              | 5 | 3 | 5 | 75  |
| Back-Side Seams       | 6 | 4 | 3 | 72  |
| Front arm Frustration | 7 | 1 | 7 | 49  |
| Clothes Tree          | 5 | 3 | 1 | 15  |

F1: Forcefulness C: Catchability F2: Frequency RPN: Risk Priority Number

10-point scale is used to determine the forcefulness, catchability and frequency values. Historical data is used to determine the values. Violence, catchability and frequency values are determined to calculate the Risk Priority Number (RPN) (Kadıoğlu et al, 2009). As the forcefulness increases between 1-10, the effect of the consequences of the failure increases. As the catchability increases between 1-10, the level of detection of the failure decreases. As the frequency increases 1-10, the frequency of the failure increases. Failures identified for violent, catchable and frequency values are listed in Table 1. In the table, the failures in the first seven are very clearly separated from the others. The seven failures should be addressed, and precautions should be taken. In general, FMEA measures are taken as a priority for the failures of RPN $\geq$ 100.

#### Causes of the failures

1. The inner arm: It is a common failure for the factory, and it has a low catchability. The failure occurs when the machine is not set correctly, the wrong or incomplete method is applied, or the type of fabric (cotton, etc.) is different.

2. Elbow press: The frequency and catchability of the failure in the ironing line is high. It needs to be repaired. The causes of the failures are the same as the inner arm.

3. Outside of the stitching of an arm part: Installation is a failure in line 2 with high frequency. The detection of the failure is challenging.

4. Stitching of an arm: The degree of catchability of the failure and the rate of waste are very high. The machine has a failure caused by incorrect setting of the sewing and running carelessness.

5. Pat Pat Press: This failure is similar to the failure in the inner arm failure. The only factor that differs is the fabric type. In some fabrics the failure is not observed, but for some type of fabrics causes the failure inevitably.

6. Fastening: The failures can only be detected by the visual monitoring method and the frequency is high. The failure factors are machine and fabric variety. The machine may not hold the sewing setting for the product, and it can hold the running machine more than necessary. The sewing point can go out the frame. The place where the attachment held can be skipped. The selection of the colour of the rope used for the fastening process may be inaccurate or the process can harm the fabric.

7. Stain: The stain is a failure that occurs on the ironing line or any manufacturing process. The failure is based on machine, human, material and environment. During the manufacturing process, stains are contaminated from the machine and the failure rate varies from the material used. The carelessness of the employee results in the mess, or the environmental impacts such as the inconvenient working environment can cause the failure.

#### **Precautions for Failures**

(1) The machines in the factory are quite old and almost 20 years old, the machine-related problems are common. The precautions caused by the machine are as follows; In order to ensure the correct and complete setting of the machine, an employee of the corresponding line should be trained on the machines, employee qualification can be measured by various interviews and employees can be placed on the corresponding machines according to their expertise. (2) For the elbow press, the machine and the human factor for the internal arm failure are the same. (3) All the measures recommended for the inner arm for the machine can also be applied to the failures of the outside of the arm. Due to the severity of the failure, the product can also be disassembled. The machine's sewing setting should be made by an employee who controls the machine. (4) Stitching out of the arm is a failure with a high degree of catchability and is very difficult to visualize. It depends on the employee's attention. The same recommendations are made with the out-of-the-way failure stitching the arm. (5) Pat pat press is the machine that the same precautions should be taken with the internal arm. In terms of the

fabric, employees should careful work on the fabric type which is a complex factor. (6) Attachment is a failure caused by employee carelessness. The precaution is the automation system. (7) Stain can be contaminated from the machine, the working environment and the worker. Therefore, the employee should always keep his / her working environment clean and tidy. Cleaning of work entries and cleaning should be done at the end of the process.

# **Automation System**

An automation system has been designed to control events at the station. It is a system in which production stages, processing times and materials (fabric type, rope type etc.) of each product can be followed. The system will be able to describe the method to be applied, the incoming station and the outgoing station. It will be an automation system that contains warning instructions for actual failures.

The data required for the system must be stored in the program. The data should reach the employee. A separate barcode number is generated for each product model. The necessary procedures, the attention points, materials to be used and semi-finished products is recorded to the system.

The barcode, which is on the incoming product to the station, is introduced to the system with the help of barcode reader. The worker chooses the operation of his own station (arm, lining, body, tailor, etc.). The operations of the product in the station, fabric, etc. materials, the method to be applied, processing times and the lot size are reflected on the digital display. The employee makes the necessary preparations for production by looking at the digital display. Pressing the button next to the screeen starts the production process. Each operation remains on the screen during the standard operation time and the next operation is completed. Warnings are taken on the screen to avoid any failure in the application technique or carelessness. When the employee needs to stop the system (break, operation failure, etc.), the red button on the screen prints. After the production process of the incoming batch is completed, production is terminated by pressing the button next to the screen. The production time of the model is saved in the system. Finally, the station that the product will go to is reflected on the digital display and the process ends for the station. This cycle continues for each station in this way.

# 3. RESULTS AND DISCUSSION

The manufacturing process of the jacket is examined by FMEA technique. As a result of the examination, 12 failure types are determined. 12 risk priority coefficients are calculated when considering the effects, causes and the precautions to determine the types of these failures before the customer level. Since 7 of these values meet  $\geq 100$  criteria, we decide that a priority should be considered. With the suggestions, a significant decrease in the severity and frequency of failures can be achieved.

Automation System can provide a significant improvement on quality. However, the system requires a long time period to establish and operate. Most of the failures are based on the employee inattention, the machine settings and the incorrect application of the method. Every failure-free product increases the quality of the product. The effective use of the fabric and yarn will reduce the failures. Reducing the failures can save time. As the failures are reduced, the number of repair work on the machine is reduced, so the machine can be used effectively. Machine efficiency may increase in terms of production.

## 4. CONCLUSIONS

As a result of the study, we discuss preventing the carelessness in the textile industry, which is the labour intensive sector. We decrease the production failures caused by the incorrect application of the production methods on changing the product models. An automation system is suggested to decrease the production failures. The use of checklists mentioned in this study has implemented to prevent the carelessness, misapplication of the productions methods and to decrease the costs. We also minimize the failures by integrating the use of checklists and the automation system.

For the future research, it can be possible to perform a fully automated system instead of some manual operations and labour-intensive processes. Nowadays, robots and automation systems used in the production environment in many sectors. Using robots and automation systems may decrease the production failures.

# REFERENCES

- Çevik, O., and Aran, G., 2009 Kalite İyileştirme Sürecinde Hata Türü Etkileri Analizi (FMEA) ve Piston Üretiminde Bir Uygulama, 8(16): 241-265.
- Gawande, A., 2011. Checklist Manifesto: İşi Doğru Yapmanın Basit Bilimi, Domingo Yayınevi, İstanbul.
- Kadıoğlu et al., 2009. Makine İmalatı Yapan Bir İşletmede Tasarım Hata Türü ve Etkileri Analizi İle Hata Kaynaklarının Belirlenmesi ve Kalitenin İyileştirilmesi, 11(1): 42-55.
- Sabır, E. C., and Bebekli, M., 2015. Hata Türleri ve Etkileri Analizinin, HTEA, Tekstil Boya-Terbiye İşletmelerinde Kullanımı, Çukurova Üniversitesi Mühendislik-Mimarlık Fakültesi Dergisi, 28(1): 157-162.

Yücel, Ö., 2007. Konfeksiyon Üretiminde Hata Türleri ve Etkileri Analizi, 17(2): 126-131.



# Inventory Management and Economic Production Quantity In Small And Medium Sized Enterprises

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**Abstract:** Deterioration of the perishable products lead to waste of production, time, labour and the related costs. In small and medium-sized enterprises, the importance of the inventory managements is not well understood. The waste and the related costs can be reduced by an appropriate inventory policy. In this study, the inventory management, which is used in manufacturing industry has been implemented in a small and medium-sized patisserie. The amount of waste and the sales revenue obtained for pastries which are the perishable products, and economic production quantity is determined according to the number of daily pastries sold. The fixed economic production quantity is a strategy looking for an economic answer to the question of when and how much will be produced. As a result of the study, it has been calculated that a total of 35 cakes are wasted on average for all types of pastry and the economic production quantity is defined to decrease the waste. As a result of this study, it is observed that the waste is decreased by 12.5%

Keywords: Inventory management, economic production quantity, inventory profile, fast moving perishable consumer goods

# **1. INTRODUCTION**

The Supply Chain is the system that involves the transformation of materials and parts from raw material to product and all stages of the products including recycling until they reach the retailer or the consumer. It is part of the stages of procurement, supplies, transformation of products and transportation of completed products to customers. The components of a supply chain are actors, resources and activities. Actors are people and organizations within the supply chain. Producers, suppliers, customers, wholesalers, retailers, warehouses and service providers are the actors of the supply chain. Resources are material, capital and human resources. Activities cover product/service conversions, business transactions and cycles within the chain.

Supply Chain Management provides both up and down information and material coordination in the chain. The aim is the high quality and low-cost products. Therefore, the quality and the production failures are due to the processes in management. One of the management processes is inventory management. Inventory management enables the company to determine the products and materials in terms of the production plan and stock quantity target. Inventory should be traced through a system and stock movements should be examined in the chain.

In inventory management of short-lived products, stock renewal by considering only the quantity as in traditional products causes inefficiencies. In addition to the quantity of short-lived products, the remaining life of the product must be considered. The remaining life of the products should be periodically checked, and short-lived products should be avoided and deteriorated.

In this study, we aim to regulate the production waste and the related costs in the small and medium sized enterprises. In our daily life we use various foods such as bread, milk and pastry with shelf life. There are various research in the literature. Interested readers can be directed to Akkerman et al. 2010 for the review paper on perishable products. Sarathi et al. (2014) examined the effect of short-lived products on the price and stock amount in the two-stage supply chain as producer and retailer. It has been shown that there is a significant increase in profit by working in coordination with information sharing among the supply chain.

#### 2. MATERIAL AND METHODS

In inventory management, the inventory level can be monitored through an inventory management system to determine the level of inventory and to determine when the stocks need to be renewed and to determine the order / production quantities. The main purpose of inventory management systems is to ensure that the required piece is ready at the desired time and at the desired location. Inventory control systems differ depending on such factors as business / organizational structure, product structure and management approaches. Economic Production Quantity Model is an analysing method used in inventory management.

The Classic Economic Production Quantity model reveals the fact that production will be made up to optimal production quantity. The basic assumptions of this model are as follows (Eroğlu, 2002):

- Demand and production speed are constant.
- Production speed is greater enough to meet the demand.
- Production preparation cost is fixed and known.
- Unit production costs are fixed and known.

In batch type production systems, the production of the same type is carried out collectively in a certain volume. In order to determine the suitable lot size and to make the minimum production costs, it is calculated by considering the relevant data such as the amount of goods to be produced in each lot, demand, stock level and production rate (Karaöz, *M*,2003).

$$Q = \sqrt{(2 * D * S/(H * ((1 - \frac{demand}{production \ rate})))}$$
(1)

The aim of Equation (1) is to find the amount of economic production that makes the total cost minimum.

$$\text{Total Cost} = \left(\left(\frac{q}{2}\right)^* \text{H}\right) + \left(\left(\frac{D}{q}\right)^* \text{s}\right) + \text{D}^* \text{C}$$
(2)

Equation (2) shows the total cost formula.

#### 3. RESULTS AND DISCUSSION

In this case study, research have been carried out on pastries, it is observed that the pastries produced in the plant in fixed amounts and some of the pastries produced not sold. It causes the waste of material, working effort and an appropriate inventory management strategy should be selected. For the strategy, the amounts of pastry, the costs of the pastries, the incomes of the pastries and the quantities are observed. According to the observations, 6 different types of pastry are produced and sold. The materials used in the enterprise are supplied weekly or monthly. The shelf life of the supplied products is controlled at certain times because they are short. A priority is given to products that are approaching the date of consumption, as well as allowing re-ordering instead of decreasing stocks.

240 pieces are produced in each day Each tray has 20 pieces. 40 different types of pastry are produced from each kind of pastry. There is a demand for bread between 07:00 and 12:00 hours at the weekdays, between 10:00 and 14:00 hours at weekend. It was observed that on average, 80 customers arrive at weekdays and 110 customers arrive at the weekend. All the pastries are not bought by the customers. Pastries remaining are waste.

|                     |                 | 54    |            |  |
|---------------------|-----------------|-------|------------|--|
| Varieties of Pastry | Unit Sale Price | Piece | Total Cost |  |
|                     |                 |       |            |  |
| Plain P.            | 0.19 TL         | 40    | 7.6 TL     |  |
| Cheesy P.           | 0.43 TL         | 40    | 17.10 TL   |  |
| Cheesed P.          | 0.28 TL         | 40    | 11.25 TL   |  |
| Olive P.            | 0.28 TL         | 40    | 11.3 TL    |  |
| Potato P.           | 0.27 TL         | 40    | 10.6 TL    |  |
| Dill P.             | 0.46 TL         | 40    | 18.45 TL   |  |

Table1. Total Cost Table

| Total | 2.63 TL | 240 | 76.3 TL |  |
|-------|---------|-----|---------|--|

Table 1 shows the total cost table according to the number of pastries produced daily. A total of 76.3 TL per day are calculated to produce pastry.

| Varieties of Pastry | Unit Sale Price | Piece | Total Cost |
|---------------------|-----------------|-------|------------|
| Plain P.            | 1.25 TL         | 40    | 50 TL      |
| Cheesy P.           | 1.25 TL         | 40    | 50 TL      |
| Cheesed P.          | 1.25 TL         | 40    | 50 TL      |
| Olive P.            | 1.25 TL         | 40    | 50 TL      |
| Potato P.           | 1.25 TL         | 40    | 50 TL      |
| Dill P.             | 1.50 TL         | 40    | 60 TL      |
| Total               | 7.75 TL         | 240   | 310 TL     |

Table2. Pastry Production Total Income Table

In table 2, total income statement is given according to the number of pastries produced daily. In a single day, a total of 310 TL income is calculated from the pastry.

 Table 3. Current Status Table

| Varieties Of Pastry | Waste Quantities (Piece) | Quantity Produced (pcs) |
|---------------------|--------------------------|-------------------------|
|                     |                          |                         |
| Plain P.            | 109                      | 1200                    |
| Cheesy P.           | 111                      | 1200                    |
| Cheesed P.          | 101                      | 1200                    |
| Olive P.            | 108                      | 1200                    |
| Potato P.           | 102                      | 1200                    |
| Dill P.             | 6                        | 1200                    |

Table 3. shows monthly production amounts and monthly amount of waste for each kind of pastry.

Table 4. Specified Quantity of Manufacturing Quantity and Economic Production Table

|            | Varieties of Pastry |           |          |            |           |         |  |
|------------|---------------------|-----------|----------|------------|-----------|---------|--|
|            | Plain P.            | Cheesy P. | Olive P. | Cheesed P. | Potato P. | Dill P. |  |
| Quantity   | 1200                | 1200      | 1200     | 1200       | 1200      | 1200    |  |
| Produced   |                     |           |          |            |           |         |  |
| Economic   |                     |           |          |            |           |         |  |
| Production | 1012                | 966       | 1054     | 962        | 987       | 1103    |  |
| Quantity   |                     |           |          |            |           |         |  |

The quantities produced and the economic production quantities are compared in Table 4.

Table 5. Comparison of The Costs

|   | Varieties of Pastry |            |            |            |            |           |
|---|---------------------|------------|------------|------------|------------|-----------|
|   | Plain P.            | Cheesy P.  | Olive P.   | Cheesed P. | Potato P.  | Dill P.   |
| Current Cost                                  | 3087.54 TL          | 7661.36 TL | 4783.28 TL | 4786.7 TL  | 4653.36 TL | 7444.3 TL |
| Cost by<br>Economic<br>Production<br>Quantity | 2805.7 TL           | 6352.4 TL  | 4137.7 TL  | 4137.7 TL  | 3987.7 TL  | 6797.4 TL |

The costs are minimized by applying the economic production quantity as in Table 5.



Figure 1. The Histogram showing the difference between the current situation and the proposed approach

The difference between the quantity produced for each pastry and 40 pieces economic production quantity is illustrated in Figure 1.

#### 4. CONCLUSIONS

Economic production quantity model is used to calculate the amount of pastry to be produced daily. 240 varieties of pastry are produced daily, and 6 varieties are considered in this study. Independent demand of customers is taken into consideration, it was observed that a total of 37 snacks per day are wasted. With the economic production quantity model for the short-lived, non-stackable products, the amounts to be produced per day for each variety are determined.

In this study, an alternative solution has been proposed with the economic production quantity model which is one of the inventory methods. The cost and the unnecessary working efforts are reduced by decreasing the waste. We show the applicability of the results in an industrial case. In the case, the results are tested, and the number of pastries produced and wasted are decreased by meeting the customer demand. The decrease in waste is directly related to the materials in stock. The application of the method also decreases the total inventory costs of the enterprise.

#### REFERENCE

- Akkerman, R., Farahani, P., & Grunow, M. (2010). Quality, safety and sustainability in food distribution: a review of quantitative operations management approaches and challenges. Or Spectrum, 32(4), 863-904.
- Sarathi, G. P., Sarmah, S. P., Jenamani, M., 2014. An integrated revenue sharing and quantity discounts contract for coordinating a supply chain dealing with short life-cycle products. Applied Mathematical Modelling, 38(15-16), 4120-4136.
- Eroğlu, A., 2002. Deterministik Envanter Modelleri, Fakülte Kitabevi, 182, Isparta.
- Karaöz, M., 2003. Öğrenme ve farklı talep fonksiyonlarını içeren ekonomik üretim miktarı model önerileri, Doktora tezi, Süleyman Demirel Üniversitesi, İsparta



# Position Weighted Balancing Technique for Assembly Lines and its Application in Service Systems

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**Abstract**: In this study, it has been shown that the assembly line balancing techniques known in the manufacturing systems can be applied in service systems. Nowadays, the services that are provided for health sector to individuals in the hospitals, both institutional and private, can be disrupted. Lack of staff and lack of good planning lead to the problem. We aim to investigate the distribution of the meal in a hospital cafeteria to the patient and their relatives to analyse the current problems. The food served to patients and their relatives is considered as a product. And the problem is solved as a one straight assembly line model by using the Position Weighted Balancing Technique. The model provides the staff who are assigned for each task to reach out to the patients in time, by helping the other waiting jobs. Here, a waiting period for the personnel is reduced and a system that allows the patient to reach the meal in the shortest time is provided.

Keywords: Assembly line balancing, position weighted balancing technique, service systems, idle time, one straight assembly line model

## **1. INTRODUCTION**

The assembly line is a production process consisting of workstations in which the work required to produce a particular product takes place. Assembly lines are connected to each other in production systems by conveyor and automatic, manual, rail material handling systems. At each workstation, a group of jobs, cycle time (time between two product outputs), precedency constraints (technological restrictions) are analysed using predetermined processing times. Assembly line balancing problems are encountered in service systems such as tax office and passport office, bank and hospital as seen in manufacturing systems. The tasks are divided into elements of work and performed by separate workers with industrialization. Faster, mass and cheaper production could be made. In this study, assembly line balancing problems used in production and service systems are discussed. The assembly line balancing minimizes idle times and balancing losses. Line balancing costs are reduced with minimum workstation.

Services are a series of activities. They cannot be tested before they are purchased or consumed (Lewis and Boom, 1983). Consumer behaviour changes with social life, technological developments and increasing level of prosperity. These changes emphasize the importance of the service sector (Yetiş, 2001). When the assembly line production for a product is designed, the problem of balancing the processing times for the workstations in the production line occurs. The aim is to run the assembly line efficiently. For this reason, during the production process, it is necessary to distribute the operations to the stations in such a way that no spare time is left to the assembly employees. Assembly line balancing is the minimization of processing time differences between workstations under existing constraints. At this point, the layout of the production systems, line balancing problem arises (Başkak, 1998). The assembly line balancing problem is the assignment of tasks to sequential stations, with the tasks required for the assembly, the times that these tasks take and the priority relationships to be given, with a performance measure optimized (Gökçen, 1994). Interested readers can be directed to the literature review (Sivasankaran and Shahabudeen 2014). In the following sections, the assembly line balancing application and the future research directions are discussed.

## 2. MATERIAL AND METHOD

Assembly line balancing problems are divided into two types: type-1 and type-2. In type-1 assembly line balancing problems, the cycle time is given, and the number of stations is minimized. If the production speed is a fixed parameter and the product demand can be estimated, the solution of the type-1 problem is considered to organize an assembly line. In type-2 problem, the number of workers, time, priority, order of each task is known. The cycle time is calculated to

balance the assembly lin. In type-2 problems, the number of stations is known, and the cycle time is calculated. If there are changes in the production process, demand pattern, the production speed can be increased, and the type-2 problem is addressed.

In this study, catering services of a hospital were discussed. A fixed-menu for each patient is considered a product. Hospital service is available at certain times. There are disruptions in service due to the insufficient number of personnel, the shortcomings in planning and the idle waiting of the personnel. It is aimed to minimize the idle time of the staff when assignments are made to workstations by minimizing cycle time.

C: Cycle time

T: Available service time

ÜS: Number of products to be served

| $C = T / \ddot{U}S$  | (1) |
|--|-----|
| Equation 1 calculates the cycle time.  |     |
| $n_{min}: \sum t_i / C$  | (2) |
| Equation 2 calculates the minimum number of staffs required.                                 |     |
| $n_{possible}: t_i > (c/2)$  | (3) |
| The equation calculates the number of jobs with a duration greater than half the cycle time. |     |

| $D(\%)=(n*C-\sum ti)/(n*C)$ Equation 4 calculates the loss of balance. | (4 | I) |
|--|----|----|
|--|----|----|

The location-weighted balancing technique is used in order to eliminate the lack of service due to the lack of staff and the planning shortcomings in a hospital catering service. Table 1 describes the operation descriptions of the hospital cafeteria service process. Table 2 shows the total standard times for those operations according to the order of operations.

| Operations | Operation Description                              |
|------------|--|
| 1          | Staff monthly menu planning                        |
| 2          | Determination of the daily food need               |
| 3          | Picking up suitable materials                      |
| 4          | Preparation and cooking                            |
| 5          | Sampling and control of food                       |
| 6          | Segmentation                                       |
| 7          | Dispersion on Tabldot                              |
| 8          | Service preparation                                |
| 9          | Separation of staff meals                          |
| 10         | Distribution of staff meals                        |
| 11         | Removal of service                                 |
| 12         | Collection of empty trays                          |
| 13         | Kitchen cleaning                                   |
| 14         | Cleaning of waste resulting from food distribution |

Table 1. Operation definitions
Table 2. standard time table

| Op Number     | 1     | 2     | 3     | 4     | 5     | 6     | 7     |
|---------------|-------|-------|-------|-------|-------|-------|-------|
| Time (minute) | 1,395 | 2,731 | 2,551 | 3,181 | 1,608 | 4,184 | 2,326 |
| Op Number     | 8     | 9     | 10    | 11    | 12    | 13    | 14    |
| Time (minute) | 4,914 | 1,899 | 3,211 | 3,304 | 6,411 | 3,181 | 0,001 |

In the technological priority diagram showing the precedency relationship of tasks, work items follow each other due to the technical characteristics of the installation. The precedencies are grouped under priority relationships and displayed with a graph. The diagram shows the relationship of two work items linked by an arrow. The numbers in the circles show the numbers of work items, the numbers above the circles show the processing times of the work items.



Figure 1. Precedency relationship of tasks

Information required to create position weighted balancing matrix is as follows. The hospital is organized with capacity of 460 beds. 3 meals in a day are served in the hospital. The food served is 520125 pieces per year. The annual service will be taken equal to the annual demand. The cafeteria serves 365 days in a year. The daily working time is 9 hours. Cycle time is the maximum time of a product can be processed at any station during its production, Equation 1 is used to calculate the cycle time. The minimum number of staffs is required. The balance loss shows the extent to which processors or stations are deployed.

T = 365 days \* 9 hours / day \* 60 minutes / hour = 3285000 min / hour

460 \* 3 = 1380 pieces per day

1380 \* 365 = 503700 pieces of products given per year

From Equation 1, 3285000 / 503,700 = 6.511 min / piece

 $\sum t_i = 1.395 + 2.731 + 2.551 + 3.181 + 1.608 + 5.555 + 2.326 + 4.914 + 1.899 + 3.211 + 3.304 + 6.411 + 3.181 + 0.001 = 42.667$ 

From Equation 2, 42.667 / 6.511 = 6.553 ~ 7 units

From Equation 3, 6.511 / 2 = 3.255

Number of work item = 4 (116, 122, 127, 131)

 $n_{enaz} = Enb$  (7; 4) = 7 pieces

From Equation 4, D (%) = (7 \* 6.511 - 42,.67) / (7 \* 6.511) = 6.38%

| Table 3. loca | ation we | eight ba | lance | matrix |
|---------------|----------|----------|-------|--------|
|---------------|----------|----------|-------|--------|

| Op. number | Op.time<br>(minute) | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 | 13 | 14 | Position<br>weight |
|------------|---------------------|---|---|---|---|---|---|---|---|---|----|----|----|----|----|--------------------|
| 1          | 1.395               |   | 1 | + | + |   | + |   | + | + |    | +  | +  | +  | +  | 35.117             |
| 2          | 2.731               |   |   | 3 | + |   | + |   | + | + |    | +  | +  | +  | +  | 33.722             |

| 3  | 2.551 |  | 2 | + | + | + | +  | +  | +  | +  | 30.992 |
|----|-------|--|---|---|---|---|----|----|----|----|--------|
| 4  | 3.181 |  |   | 4 | + | + | +  | +  | +  | +  | 28.442 |
| 5  | 1.608 |  |   | 5 | + | + | +  | +  | +  | +  | 26.871 |
| 6  | 4.184 |  |   |   | 6 | + | +  | +  | +  | +  | 25.262 |
| 7  | 2.325 |  |   |   | 7 | + | +  | +  | +  | +  | 22.933 |
| 8  | 4.914 |  |   |   |   | 8 | +  | +  | +  | +  | 22.917 |
| 9  | 1.899 |  |   |   |   |   | 9  | +  | +  | +  | 18.003 |
| 10 | 3.211 |  |   |   |   |   | 10 | +  | +  | +  | 16.104 |
| 11 | 3.304 |  |   |   |   |   |    | 11 | +  | +  | 9.591  |
| 12 | 6.411 |  |   |   |   |   |    |    | 12 | +  | 3.181  |
| 13 | 3.181 |  |   |   |   |   |    |    |    | 13 | 0      |
| 14 | 0.001 |  |   |   |   |   |    |    |    |    |        |

Table 3 shows the position weights for each work item. The position weight of the work item is calculated by adding its own process time and the process times of subsequent jobs. Work items are sorted by decreasing position weights. Work items are assigned to workstations. Assignment is made starting from the highest position weight. The largest position-weighted work item is assigned to the first station. The idle time of the workstation is calculated by subtracting the total of assigned times from the cycle time. The largest position weight is selected from the remaining work items. The work item is checked and assigned to the station. The list of reserved jobs is checked. The precedency will not be distorted if work items with no predecessor are assigned. The duration of work items is compared with the idle time of the station. If the process time is less than the unassigned time, the work item is assigned to the station and the unused station time is recalculated. The process is performed when the process time is unused.

## 3. RESULTS AND DISCUSSION

As a result of the weighted balancing technique we performed in the hospital cafeteria at Table 4, the operation time is realized as 6.553 minutes. If more than one personnel are assigned (6,511 = 7 personnel), it is to (7 \* 7 \* 365 = 17885 man \* hour) = 12817 hours of service effort in a year, 17885-12817 = 5068 hours is idle.

| Station n. | Item n. | Position weight | Process time | Cumulative process time | Idle time |
|------------|---------|-----------------|--------------|-------------------------|-----------|
| 1          | 1       | 35.117          | 1.395        | 1.395                   | 5.115     |
|            | 2       | 33.722          | 2.731        | 4.126                   | 2.025     |
|            | 5       | 26.871          | 1.608        | 5.734                   | 0.777     |
| 2          | 3       | 30.992          | 2.551        | 2.551                   | 3.861     |
|            | 4       | 28.442          | 3.181        | 5.732                   | 0.899     |
| 3          | 6       | 25.262          | 4.184        | 4.184                   | 2.327     |
|            | 7       | 22.933          | 2.326        | 6.511                   | 0         |
| 4          | 8       | 22.917          | 4.914        | 4.914                   | 1.597     |
|            | 9       | 18.003          | 1.597        | 6.511                   | 0         |
| 5          | 10      | 16.104          | 3.211        | 3.211                   | 3.383     |
|            | 11      | 12.895          | 3.304        | 6.511                   | 0         |
| 6          | 12      | 9.591           | 6.411        | 6.411                   | 0.1       |
|            | 14      | 0.001           | 0.001        | 6.412                   | 0.099     |
| 7          | 13      | 3.181           | 3.181        | 3.181                   | 3.331     |

### Table 4. Balancing results

### **4. CONCLUSIONS**

In this study, it has been shown that assembly line balancing in manufacturing systems is also applied in service systems. As in production systems, the standard services offered to the customer in service systems can be divided into sections. Work order is also important in the service systems and production system. When the standard service is considered as a service line, all of the approaches for the assembly line can also be applied to the service line. In the study, a balancing study was performed in a hospital cafeteria where catering services were provided. The problem is solved by simulating

a single model assembly line balancing problem. As a result, it has been shown that assembly line balancing methods can also be applied to service systems.

#### REFERENCES

- Başkak M. 1998. Çok Modelli/Ürünlü Montaj Hatlarının Dengelemesi İçin Yeni Bir Model ve Çözüm Yöntemi, Doktora Tezi, İstanbul Teknik Üniversitesi Fen Bilimleri Enstitüsü, İstanbul.
- Gökçen, H. 1994. Karışık Modelli Deterministlik Montaj Hattı Dengeleme Problemleri İçin Yeni Modeller, Doktora Tezi, Gazi Üniversitesi Fen Bilimleri Enstitüsü, Ankara.
- Lewis, R.C., Booms, B.H., 1983. The Marketing Aspects of Quality in Emerging using the Ranked Positional Weight Technique. Journal of Industrial Engineering. 12 (6)
- Sivasankaran, P., Shahabudeen, P. 2014. Literature review of assembly line balancing problems. The International Journal of Advanced Manufacturing Technology, 73(9-12), 1665-1694.
- Yetiş, H., 2001. Hizmet Kalitesinin Servqual Modeli ile Ölçülmesi, Yüksek Lisans Tezi, Gazi Üniversitesi, Fen Bilimleri Enstitüsü, Ankara.



# Statistical Process Control and its Application in the Textile Industry

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**Abstract:** One of the most widely used quality control techniques is the statistical process control. The application of this method in the Textile Industry is presented as an example of the applications in the manufacturing sector. The understanding and implementation of statistical quality control methods will help increasing the quality levels. In this study, the quality control department of a firm in the textile industry carries out monthly and annual statistical process control with quality control data generated in the production line. The elbow press is focused in the assembly line aiming at minimizing the error rates. Pareto Analysis, Spread Diagram, Control Charts and p - c diagrams are provided, and the error rates are examined.

Keywords: Quality control, statistical process control, decreasing the production failures, textile industry

## **1. INTRODUCTION**

Textile sector is one of the manufacturing sectors where quality variations are seen intensively due to its inherent features. Since there is a labour-intensive production against the developing technology, the human factor is on the base of different production and process errors. There is a significant contribution of statistical process control to science and industry; the production errors in terms of the predetermined specifications and their effects can be determined in the industrial applications. Therefore, companies avoid the cost of errors and save time. The main objective of all enterprises is to achieve maximum quality in their products and services, to sustain and improve the quality level continuously.

Quality improvement is a process that consists of the studies done by companies to determine and eliminate the factors that negatively affect the quality of their products or services to increase the level of customer satisfaction. Nowadays, statistical methods are used in quality and process improvement. These methods, which are widely used in Japan, are known as Ishakawa's seven main tools: Cause-Effect Diagram, Check Sheet, Control Chart, Histogram, Pareto Chart, Scatter Diagram, Run Chart. Kaoru Ishikawa argues that 95% of the problems in a business can be solved by seven techniques of quality control and he explains and exemplifies these techniques (Özer, 1990).

It can be said that Statistical Process Control is simply concerned with determining the variability in the process and minimizing the quality variability (Özcan, 2001). In the past, the solution to the quality problems can be reached by the classical methods, however recently, the systematic and modern quality control techniques are preferred in the production process. In this study, the seven basic tools are examined in a textile factory, to identify the most common mistakes in the production stage and to focus on these errors to increase the competitiveness with quality and production efficiency. The rest of the study is organised as follows: A literature review is presented in section 2. In section 3, the quality term is described, and the quality control problems are addressed. In section 4, the results of the case study and future research directions are discussed.

# 2. QUALITY AND QUALITY CONTROL

Quality focus on current and future needs and can be defined as design, availability, consumer satisfaction. Crosby said that quality is not what quality is: Quality is not absolute perfection. The way to achieve quality is not correction, but prevention. The quality performance standard is zero error, not the acceptable level of error. Quality is measured by non-compliance costs and cannot be measured with indices.

Quality control according to Feigenbaum; It is defined as the planning and coordination of the contributions of the departments for market research, design, research-development, manufacturing, sales and after-sales service to create a

certain quality level, which provides the desired quality characteristics. Quality control aims to improve quality and ensure the continuity of quality in order to meet customer requirements.

Quality control is not the sole responsibility of a department or a specific group of people in the enterprise. It is a community of activities at all stages of production, which is the responsibility of all employees from the highest level to the manager. Therefore, the concept of quality control is integrated with the concepts of total quality control and total quality management (Doğan, 1991).

#### **3. MATERIAL AND METHODS**

Statistical process control is a feedback mechanism that measures the quality of products as output from any process, determines whether they meet the desired specifications and, takes precautions. In this study, the errors that occur during the production of jackets in a production facility operating in the textile industry are discussed. For creating the quality control diagrams related, Minitab software is used. We give suggestions to improve the quality level accounting for the reasons of the errors.

i. Control table: The control table has a simple use for understanding how often the events occurred during the production.

Table 1. A Weekly Error of Jacket Production

| Production Bands      | Error Number |  |
|-----------------------|--------------|--|
| Body Line             | 48           |  |
| Lining Line           | 21           |  |
| Arm Line              | 31           |  |
| Assembly-1 Line       | 13           |  |
| Assembly -2 Line      | 142          |  |
| Ironing Line          | 264          |  |
| Final Control Process | 255          |  |
| Total Error           | 744          |  |

Table 2. Elbow Press Errors in The Iron Band

| Days      | Items Viewed | Error Number | Cumulative Percent (%) |  |
|-----------|--------------|--------------|------------------------|--|
| Monday    | 570          | 35           | %1.129                 |  |
| Tuesday   | 700          | 20           | %1.774                 |  |
| Wednesday | 550          | 30           | %2.742                 |  |
| Thursday  | 630          | 24           | %3.516                 |  |
| Friday    | 650          | 30           | %4.482                 |  |
| TOTAL     | 3100         | 139          |                        |  |

ii. Process Diagram: The company produces two type jackets and one type of pant. Process (flow) diagram of pants line as follows:



Figure 1. Process (flow) diagram of pants band



Figure 2. Process (flow) diagram of jacket band

iii. Histogram: The reason for drawing a histogram is to have information about the distribution of the interested variable.



Figure 3. Histogram of Error Numbers in Jacket Line

iv. Pareto Analysis: The errors can be classified in terms of their effect on product performance or fix costs. Pareto analysis shows which of the problems should be considered primarily. The diagram is applied by quality groups in order to direct the efforts to the most productive areas and to make the right decisions. Pareto analysis determines the order of solution and the priority the problems. Often, 80% of the problems are caused by 20% of errors.

The stages of a pareto analysis are as follows:

The time interval for collecting the data and the data required are determined. Data collection form is arranged. The data is saved in the form. Thus, pareto diagram is drawn using the data.



Figure 4. Pareto Analysis of Error Numbers in Production Line

From the Pareto Analysis in Figure 4, we can see that we must focus on the ironing line since most of the errors are found in the ironing line. In this way, we can discuss the Elbow Press which is on the ironing line.



Figure 5. The Pareto Analysis of the Error Numbers for the Elbow Press of a Jacket

v. Scatter chart: It is the diagram which is obtained by marking the data in terms of its characteristic related to one or some of the reasons. When this diagram shows the relationship between the two variables, one can predict how the other can act by controlling one another.



Figure 6. Scatter Chart

vi. Cause and Effect Diagram: For each step in the process, or for each problem, starting from the general reasons, the smallest detail is brought down, and the basic information is revealed in order to reveal the cause. The diagram is an effective tool in explaining what causes and what causes it.



Figure 7. Cause and Effect Diagram

vii. Control Charts: It is a graph showing the quality changes in the time. The measured values obtained from the samples taken at certain and equal time intervals from production. A control graph contains essentially three lines; Centre Line, Upper Control Limit, and Lower Control Limit (Kartal, 1999). The main function of the process control graphs is to show how a process works. In this way, process performance, customer expectations and the production process are improved.

C Chart:

| For each subgroup: $c_i$ = number of defects in subgroup i                  |     | (1)                    |
|---|-----|------------------------|
| For all data: $c = (Total number of defects) / (Total number of subgroups)$ |     | (2)                    |
| OD = (c = Average number of defects per subgroup                            |     | (3) $\ddot{U}KS = c +$ |
| 3√ c  | (4) |                        |
| $ACS = c + 3\sqrt{c}$   |     | (5)                    |

 $c_i$  in Equation (1) refers to the number of defects in subgroups. In Equation (2), we get c by dividing the number of defects by the number of sub-groups. In Equation (3), the expression is so called the mean number of faults in a subgroup. The formula in Equation (4) is the Upper Control Limit and the formula in Equation (5) is the Lower Control Limit.



**Figure 8.** C Chart of Quality Control Errors P Chart:

| For each subgroup: $p_i = (Number of defective products in the group) / n_i p_i = defective product rate$ | (6)  |
|---|------|
| For all data: $p = (Total number of defective products) / (Total number of groups)$                       | (7)  |
| OD = (p = Average defective product per group   | (8)  |
| UKS = p + 3spi  | (9)  |
| AKS = p -3spi   | (10) |

 $spi = \sqrt{p} (1-p) / n_i = the standard error of p$ 

Equation (6) refers to the number of defective products in  $p_i$  subgroups. In Equation (7), we obtain p by dividing the total number of defects by the total number of groups. In equation (8), the expression is so called the average number of defects. Equation (9) is the upper control limit and Equation (10) is the lower control limit.



Figure 9. P Control Chart of Quality Control Errors

### 4. RESULTS AND DISCUSSION

In terms of the error rates during production, the following suggestions are given for the textile company:

• It has been observed that, for all the lines in the production, the fixed seam shares given to the garment molds should be done by pull test and determined by sewing type.

• Every new model that will be produced in the jacket line where jacket models has different fabrics, carry a risk for dressmaking operators. In the same model and in high-volume orders, the risk is increasing. In small batch orders, the time taken by operators to get used to the model has decreased considerably. For this reason, it is suggested to conduct a model evaluation meeting with the personnel working on the production line and production managers for each new model. In this way, it is possible to determine the phases where error may occur and to make an error-free dressmaking.

• A detailed dressmaking instruction for each model should be prepared.

• A system analysis should be made in the enterprise accounting for the work/ study and the ergonomic factors.

• Cleaning of sewing machines by operators is suggested at the end of working hours. In order to increase the effectiveness of the machine maintenance department, the general maintenance activities should be systematically made monthly and quarterly.

#### REFERENCES

Bircan, H, Özcan S. 2001. "Otomotiv Yan Sanayide Uygulanabilen İstatistiksel Proses Kontrol Teknikleri vee Bir Uygulaması" 2. Ulusal Araştırmalar Sempozyumu, İ.T.Ü. Sosyal tesisleri Maçka-İstanbul.

DOGAN, Üzeyme. 1991. Kalite Yönetimi ve Kontrolü. İstiklal Matbaası, İzmir

- Gedik, H., 2003. İstatiksel Proses Kontrol Teknikleri ve Sivas Dikim Evi'nde Bir Uygulama, Cumhuriyet Üniversitesi Sosyal Bilimler Enstitüsü, Yayınlanmamış Yüksek Lisans Tezi, Sivas.
- Kartal, M., 1999. İstatistiksel Kalite Kontrol, Şafak Yayınları, Sivas.
- Özer, S., 1990. "Kalite Kontrolün Gelişimi ve İstatistiksel Proses Kontrol Teknikleri", Kalite, Sayı:7, Türkiye Şişe Cam Fabrikaları A.Ş. İstanbul.



# Enhanced Gas Sensing Properties of Indium Doped ZnO Thin Films by Fabricated Silar Method

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**Abstract:** In the present work, the effect of in doping on the various properties of the ZnO thin films was investigated. The pure and 5% indium doped ZnO thin films have been synthesized by SILAR method on glass substrates. The XRD study clearly showed the existence of the polycrystalline structure of the films. The morphological study indicated the formation of granular structures on the surface of the films. The band gap energy and the grain size values of 5% In-ZnO were 3.32 eV and 22.33 nm, respectively. Also, the sensitivity of ZnO thin film was improved with indium doping. Consequently, it was seen that indium incorporation made a significant change on the structural, morphological properties and enhanced gas sensing performance.

Keywords: ZnO, indium doped ZnO, thin film, gas sensor, SILAR

### **1. INTRODUCTION**

The effects of preparation conditions and doping on gas sensing the property of ZnO based thin films have been intensively studied because of their interesting functionalities such as less toxicity, low cost, optimal conductivity, stability, and electroacoustic transducer, etc.(Ganbavle et. al., 2016; Bindal et. al, 2013). Moreover, ZnO is a well-known sensor material for CO, NH<sub>3</sub>, volatile organic compounds (VOCs), etc., and the effects of preparing condition and doping on the sensor property have also been studied. On the other hand, various kinds of NO sensing materials, such as tin oxide (SnO<sub>2</sub>), tungsten oxide (WO<sub>3</sub>), titanium oxide (TiO<sub>2</sub>) and Zinc oxide (ZnO), etc., have been developed, and they are aimed at exhaust gas and environmental monitoring (Wang et. al., 2010). However, there are only a few studies on indium doped ZnO based sensors for NO sensing so far (Latyshev et. al., 2017).

ZnO can be doped with a wide variety of ions. The ZnO doping is achieved by replacing  $Zn^{2+}$  atoms with atoms of elements of higher valance such as  $In^{3+}$ ,  $Al^{3+}$ ,  $Sn^{4+}$ , and Pb<sup>4+</sup>. Several deposition techniques are used to grow indium doped zinc oxides (IZO) thin films such as sputtering (Latham et. al., 2010), hydrothermal synthesis (Cheng et. al., 2008), solgel (Ivanova et. al., 2010) and successive ionic layer adsorption and reaction (SILAR) (Yildirim et. al., 2012). Among them, the SILAR method is reported as a relatively new and less investigated method. As compared to the other methods of thin films mentioned above, the SILAR method is simple, flexible and requests an easy way to the synthesis of thin films. It does not demand good quality substrates and can be controlled at room temperature without the need for vacuum. Moreover, it is also effective of cost and it can accommodate to any substrate material or surface profile (Karaduman et. al., 2017).

In this paper, we have investigated the effect of an indium doping ratio (5% indium) on the structural, electrical and gas sensing properties of indium-doped zinc oxide thin films deposited by SILAR method.

### 2. MATERIALS AND METHODS

Before the successive ionic layer adsorption and reaction process (SILAR), the glass substrate was washing in soapy water and then rinsed in distilled water and dried in nitrogen gas. After washing, the substrates were ultrasonic cleaned in acetone for 10 minutes. Then the substrates were rinsed for 10 minutes in a (1:1) ethanol-water mixture. ZnO thin films were grown on glass substrates using SILAR method at room temperature and ambient pressure. To deposit ZnO thin

films, aqueous zinc-ammonia complex ions ( $[Zn(NH_3)_4]^{2+}$ ) and aqueous indium-ammonia complex ions ( $[In(NH_3)_4]^{3+}$ ) were chosen for the cation precursor, in which using analytical reagents of 0,1M ZnCl2 (pH $\approx$ 5,5) 0,1M InCl3 (pH $\approx$ 5,5) and concentrated ammonia 0.8 M NH3(aq) (25–28%) were used. The concentration values defined for the indium (In) solution was as 0.1 M and the molar ratio of in: NH3 is 1:10 obtained.

The gas-sensing properties of the sensors were measured with a gas-sensing characterization system (Karaduman et. al., 2017). The gas sensing experiments were performed in a test chamber with NO gas in the concentration range of 100 ppb-50 ppm by monitoring changes in the sensor resistance. In order to observe the behavior of different concentrations under the same conditions, the airflow rate must be kept constant. The gas and dry airflow rates were precisely manipulated using computer controlled mass flow controllers. A LakeShore 325 temperature controller was used to maintain a constant temperature. Silver wires were affixed to the top of the surface of the pellets with silver paste. The resistances of the samples were continuously monitored with a computer-controlled system by using a Keithley 2400 sourcemeter and data was collected in real-time using a computer with corresponding data acquisition hardware and software. The sensors under-test were fixed in the chamber and then purged by air. Initially, dry air flowed through the chamber, so the baseline sensor resistance could be recorded. The relative humidity was about 25 % and monitored by a Honeywell HIH-4000 humidity sensor. The sensing behavior is analyzed in terms of sensor sensitivity. The sensor sensitivity was calculated using the following equation 1:

$$Sensitivity = \frac{R_g}{R_a}$$
(1)

Where R<sub>a</sub> and R<sub>g</sub> are the resistivities in the air and upon exposure NO gas in air, respectively (Yoshida 2002).

## 3. RESULTS AND DISCUSSION

The polycrystalline structures of the pure ZnO and 5% In-ZnO thin films were determined by XRD measurements, as seen in Fig 1. The XRD pattern of the pure ZnO thin film shows sharp and strong diffraction peaks and these diffraction peaks indicate the existence of the hexagonal wurtzite structure of ZnO [Card No:36-1451] (Thambidurai et. al., 2014). The 2 $\theta$  values of the diffraction peaks are 31.8°, 34.5°, 36.3°, 47.5°, 56.7°, 62.9°, and 68.1°, and these peaks are corresponding to (100), (002), (101), (102), (110), (103) and (112), respectively. There is no impurity phase in the XRD patterns of the 5% In-ZnO film.



Figure 1. The XRD images of pure ZnO and 5% In-ZnO films.

The grain size D was evaluated using the Debye-Scherrer formula (Wang et. al., 2010), as given in equation 2:

$$D = \frac{0.94\lambda}{\beta \cos\theta} \tag{2}$$

Where  $\lambda$  is the wavelength of X-ray radiation, *c* and  $\beta$  is FWHM in radian. The grain size of the films were calculated using the FWHM for (002) peak. The grain size of ZnO decreased from 37.57 to 22.33 nm with indium doping of 5%. This behavior is due to the distortion caused by the substitution of In<sup>3+</sup> ions, which acts as an impurity in the ZnO (Wang et. al., 2010).



Figure 2. The SEM images of pure ZnO and 5% In-ZnO films.

Gas sensing properties are significantly affected by the surface morphology. For this, SEM analysis was performed to better understand the surface morphology of the thin films and SEM images of the films were given Fig. 2 (a-b). Depending on indium doping ratio, the variation in the morphology of the thin films is clearly seen from SEM images. All the films have dense and homogeneous surface morphology. It was seen that the pure ZnO and 5% In-ZnO thin films had a granular structure.

The optical properties of ZnO and 5% thin films were determined by using the optical absorption measurements. The optical direct bandgap of the thin films can be determined by extrapolation of the linear region of  $(\alpha hv)^2$  versus (hv) plots using Tauc equation (Gawali et. al., 2018):

$$\alpha = \frac{A}{h} (h - E_g)^{1/2} \tag{3}$$

where A is a constant,  $\alpha$  is the absorption coefficient,  $E_g$  is the optical bandgap and hv is a photon energy.



**Figure 3.**  $(\alpha h v)^2$  versus (hv) plots of ZnO and 5% In-ZnO thin films

Fig. 3 shows  $(\alpha hv)^2$  versus (hv) plots of ZnO and 5% In-ZnO. The bandgap values increased from 3.2 to 3.32 eV with indium dopant. This increase of the band gap of ZnO thin film explained by the Bursteine Moss shift. The result was in agreement with the literature (Thambidurai et. al., 2014).



Figure 4. The gas sensitivity of ZnO and 5% In-ZnO thin films for 50 ppm at 167 °C to NO gas.

Fig. 4 displays the gas sensitivity of ZnO and 5% In-ZnO thin films for 50 ppm gas concentration at an operating temperature of 167 °C. The sensitivity of ZnO and 5% In-ZnO thin films were calculated to be 2.23 and 10.1, respectively. It is clearly seen from Fig. 3, the sensitivity of the 5% In-ZnO is drastically higher than the ZnO. Therefore, the sensitivity of ZnO thin film has improved with indium doping. The gas sensing mechanism of metal oxide semiconductor sensors can be explained by changing conductivity or electrical resistance of the sensor surface. Adsorbed oxygen captures electrons from the conduction band of the semiconductor, resulting in molecular  $O_2^-$  form and subsequently atomic  $O^-$ ,  $O^{2-}$  forms (Hassan et. al., 2017). After the oxygen adsorption reaction on the outer surfaces of the sensor is exposed to NO, due to the attraction of additional electrons from conduction band, leading to an increase of the electron depletion layer and increasing the resistance of the sensor. Moreover, indium doping can enhance the reacting gas adsorption on the surface of the sensor, which also increases gas sensitivity by enhancing gas reactions.

#### 4. CONCLUSION

In summary, the structural, morphological, optical, and gas sensing properties of pure ZnO and 5% In-ZnO films prepared by SILAR method were investigated. The pure ZnO and 5% In-ZnO thin films are polycrystalline in nature. The band gap energy and the grain size values of 5% In-ZnO are 3.32 eV and 22.33 nm, respectively. Also, the sensitivity of ZnO thin film has improved with indium doping. Consequently, it was seen that indium incorporation makes a significant change and enhance on the structural, morphological, optical and gas sensing properties.

#### REFERENCES

- Bindal N., Sharma M., Sharma R., Upadhaya S.C., 2013. Influence of Substrate in Structural, Morphological, and Optical Properties of ZnO Films deposited by Successive Ionic Layer Adsorption and Reaction (SILAR) Method, International Journal of Advance Engineering Technology And Research 1: 1–7.
- Cheng C., Xu G., Zhang H., Luo Y., 2008. Hydrothermal synthesis Ni-doped ZnO nanorods with room-temperature ferromagnetism, Materials Letters 62:1617–1620.
- Ganbavle V. V., Inamdar S.I., Agawane G.L., Kim J.H., Rajpure K.Y., 2016. Synthesis of fast response, highly sensitive and selective Ni:ZNO based NO2 sensor, Chemical Engineering Journal 286: 36–47.
- Gawali S.R., Patil V.L., Deonikar V.G., Patil S.S., Patil D.R., Patil P.S., Pant J., 2018. Ce doped NiO nanoparticles as selective NO2gas sensor, Journal of Physics and Chemistry of Solids 114: 28–35.
- Hassan M.M., Khan W., Mishra P., Islam S.S., 2017. Naqvi A.H., Enhancement in alcohol vapor sensitivity of Cr doped ZnO gas sensor, Materials Research Bulletin 93: 391–400.

- Latham K., Plessis J. du, Kalantar-Zadeh K., Shafiei M., Rahmani M.B., Wlodarski W., Keshmiri S.H., 2010. Transition from n to p -Type of Spray Pyrolysis Deposited Cu Doped ZnO Thin Films for NO 2 Sensing, Sensor Letters 7: 621–628.
- Latyshev V.M., Berestok T.O., A.S. Opanasyuk, A.S. Kornyushchenko, V.I. Perekrestov, Nanostructured ZnO films for potential use in LPG gas sensors, Solid State Sciences. 67 (2017) 109–113.
- Ivanova T., Harizanova A., Koutzarova T., Vertruyen B., 2010. Study of ZnO sol-gel films: Effect of annealing, Materials Letters 64:1147–1149.
- Wang C., Yin L., Zhang L., Xiang D., Gao R., 2010. Metal oxide gas sensors: Sensitivity and influencing factors, Sensors 10: 2088– 2106.Yildirim M.A., Akaltun Y., Ates A., 2012. Characteristics of SnO2thin films prepared by SILAR, Solid State Sciences 14: 1282–1288.
- Karaduman I., Çorlu T., Yıldırım M.A., Ateş A., Acar S., 2017. Hydrogen Gas Sensing Characteristics of Nanostructured NiO Thin Films Synthesized by SILAR Method, Journal of Electronic Materials 46: 4017–4023.
- Yoshida Y., Matsuura Y., Inoue T., Ohtsuka K., Kajiyama Y., 2002. Metal oxide semiconductor NO2 sensor, Sensors and Actuators B: Chemical 25: 388–391.
- Thambidurai M., Kim J.Y., Kang C. mo, Muthukumarasamy N., Song H.J., Song J., Ko Y., Velauthapillai D., Lee C., 2014. Enhanced photovoltaic performance of inverted organic solar cells with In-doped ZnO as an electron extraction layer, Renewable Energy 66: 433–442.



# Investigation of Electrical Properties of Al-Doped ZnO Thin Films with Different Solvents

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**Abstract:** Aluminium doped zinc oxide thin films can be deposited by many physical and chemical methods. Every deposition method has its own advantages and disadvantages according to their use. In the literature, ZnO thin films fabricated using different deposition approaches. Among them, the sol–gel method is a simple method used for the preparation of different metal oxide thin films. In addition, the solvent that used in growing process changes electrical and morphological properties ZnO of thin films. Aluminum-doped Zinc Oxide (AZO) thin films were deposited by solgel spin-coating method using different solvents: ethanol and 2-methoxyethanol with TEA stabilizer. The electrical and morphology properties of films were characterized by IV, SEM and EDAX. The result revealed the effect of solvents on thin films' morphology and resistivity; the conductivity of 2-methoxyethanol sample is increased.

Keywords: Sol-Gel, solvent, stabilizer

# **1. INTRODUCTION**

In recent years, most researches in material sciences have been focusing on semiconductor materials with wide band gap. One of them is zinc oxide (ZnO), which exhibits multifunctionalities, due to its novel nonlinear optical properties. The advantages of ZnO thin films include abundance and non-toxicity of the ZnO material, low cost and quantum size effect. Therefore, ZnO is considered as a very promising functional electronic material due to its electrical, optoelectronic and luminescent properties. ZnO thin films are very attractive candidates for various fields of industrial and high-technological applications such as gas sensors, varistors, ultraviolet (UV) detectors, thin film transistors and thin-film solar cells, especially due to their wide band gap energy of 3.37 eV at room temperature and high mechanical and thermal stabilities. The doping of impurities such as Al, In, Ga, B resulted in high electrical conductivity. By the process of doping aluminum to ZnO, free electrons will be released when zinc is replaced by aluminum, so the increase of carrier concentration can improve the conductivity (Maache et.al., 2017).

ZnO thin films can be realized using different fabrication methods, including vapor phase processing approaches such as physical vapor deposition (PVD) and chemical vapor deposition (CVD) and wet solution processing approaches such as sol-gel processes. Different techniques result in different properties of the ZnO thin films. The structural, electronic transport and optical properties of the ZnO films are very sensitive to the preparation method and deposition conditions. The sol-gel spin coating process provides a simplified fabrication route for the ZnO layers, as it eliminates the need of vapor phase deposition equipment. Very recently, the sol-gel processes have been used as an inexpensive alternative for the fabrication of ZnO thin films. The sol-gel fabrication technique is particularly adapted to produce ZnO colloids and films in a simple, low cost and highly controlled way. As compared to other preparation methods, the spin coating method for the growth of the ZnO films has some important advantages such as its simplicity of technological runs and low cost of the used materials and equipment. In the sol-gel spin coating process, the properties of the ZnO films correlate with the sol-gel preparation conditions and parameters.

In this paper, a low-cost sol-gel spin coating technique was used to synthesize the ZnO films on glass substrates. The sol-gel spin coating fabrication process of the ZnO films is described. The influence of solvents on the material properties of the ZnO films was investigated. The effects of the solvents on the structural and optical properties of the sol-gel spin coated ZnO films will be discussed (Kamaruddin et. al., 2011).

### 2. MATERIALS AND METHODS

The spin coating technique was applied to deposit Al-doped ZnO (AZO) thin films on glass substrates. The films were derived from two different coating solutions prepared using two different combinations of solvents and with same stabilizer. Two organic solvents ethanol (ZA-26) and 2-Methoxyethanol (ZA-29) and stabilizer from the III group aliphatic amine triethylamine (TEA) were used to form the thin films. The coating solutions of the deposited films were synthesized by dissolving zinc acetate dihydrate, ZAD (ZnC4H6O4.2H2O) and aluminum nitrate nonahydrate (Al(NO3)3·9H2O) in the solvent, and then the stabilizer was added to the mixed solution. The concentration of ZAD in the solvent was 0.2 M/l, and the molar ratio of stabilizer to ZAD was kept at 1:1. The Al dopant concentration ([Al]/[Al + Zn]) was calculated to be 3 at.%. Al-doped ZnO samples prepared by sol-gel spin-coating method annealed at N2 environment. The data has been measured by Keithley 2400 Source Meter collected in real-time using a computer. The morphology of samples were observed by scanning electronic microscope (SEM).

#### **3. RESULTS AND DISCUSSION**

Surface morphology of ZnO thin films were investigated by scanning electron microscopy (SEM). Fig 1, below illustrates the SEM images of the AZO thin films coated by spin-coating technique with different solvents. The SEM images confirm that the surface morphology of the films mainly depends on the solvents. No cracks are observed from the film surfaces. The film has a porous nature due to solvent evaporation and dispersion agent during the heating process (Kumar et. al., 2017). The grain size of deposited films (ZA-26 and ZA-29) are about 1.453  $\mu$ m and 196.4 nm, respectively. When more grains are presented in the film, there are fewer grain boundaries. Therefore, the atoms are easily moved from one grain to another grain and also the electrical properties are improved. This fact is perfectly matched with the two-point probe results, where the electrical conductivity changed by solvents. The grain size increases with respect to change of solvents this may be due to the increase of crystallites (Kumar et. al., 2017).



Figure 1. ZA-26 and ZA-29 SEM

Here, the electrical properties are mainly depend on the different solvents. From the Fig 2, it is clearly visible that resistance alters with respect to different solvents due to reduction in the grain boundary scattering to free charge carriers and increase in carrier lifetime. We conclude from the electrical studies that thin film with ethanol solvent noted less resistivity than the film with 2-methoxyethanol.



Figure 2. IV graph of samples

From the illustrations below in Fig 3, we can see that the EDAX spectra of the sensors show the existence of Al, Zn and O elements in the film. The presence of Ca and Si elements in the spectra is due to the precursor materials in the synthesis process and the glass bottom.



Figure 3. EDAX of the samples

#### 4. CONCLUSION

The thin film deposition by sol-gel technique is a chain process where the quality and properties of the final product rely on the intermediate steps. Pure zinc oxide and aluminum doped zinc oxide films were prepared onto glass substrates by spin coating. They were characterized by using a variety of techniques. Morphology of the films showed that they depend on solvents and have porous nature. The EDAX spectra of the sensors show the existence of Al, Zn and O elements in the film. The electrical resistivity of the films strongly depended on the solvents as well. The film with 2-methoxyethanol (ZA-29) showed higher resistivity than the film with ethanol solvent.

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#### REFERENCES

- Kamaruddin, Sharul Ashikin, et al. "Zinc oxide films prepared by sol-gel spin coating technique." Applied Physics A 104.1 (2011): 263-268.
- Kumar, K. Deva Arun, et al. "Effect of solvents on sol-gel spin-coated nanostructured Al-doped ZnO thin films: a film for key optoelectronic applications." Applied Physics A 123.12 (2017): 801.
- Maache, M., T. Devers, and A. Chala. "Al-doped and pure ZnO thin films elaborated by sol-gel spin coating process for optoelectronic applications." Semiconductors 51.12 (2017): 1604-1610.



# Investigation of Corrosion Properties of Copper Carbide Surface Composites Produced on Copper Surface

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**Abstract:** In this study, aims to improve the corrosion properties of copper and to minimize its alteration of electrical properties. For this reason, copper carbide surface composite on electrolytic copper are coated with plasma spray coating method.  $B_4C$  was used as carbide. This carbide have been added 5%, 10% and 20% by weight to investigate the effect on corrosion properties. Potentiodynamic method was used for corrosion. Through the examinations of corrosion results of all samples, it is observed that the lowest current density is in the uncoated substrate material.

Keywords: Copper, plasma spray coating, surface coating, microstructure, corrosion

# Bakır Yüzeyinde Üretilen Bakır Karbür Yüzey Kompozitlerinin Korozyon Özelliklerinin İncelenmesi

Öz: Bu çalışmada, bakırın korozyon özelliklerinin geliştirilmesi ve elektriksel özelliklerinin ise minimum etkilenmesi hedeflenmiştir. Bu sebeple elektrolitik bakır üzerine bakır matriksli karbür takviyeli yüzey kompozitleri plazma sprey kaplama yöntemiyle kaplanmıştır. B<sub>4</sub>C kullanılmıştır. Bu karbürlerin miktarının korozyon özellikler üzerine etkisini araştırmak amacıyla her biri için ağırlıkça %5, %10 ve %20 olarak ilave edilmiştir. Korozyon için potansiyodinamik yöntem kullanılmıştır. Korozyon sonuçları irdelendiğinde tüm numuneler arasında en düşük akım yoğunluğunun kaplamasız altlık malzemede olduğu görülmektedir. Numunelerin kaplanmasıyla birlikte akım yoğunluğu artmaktadır.

Anahtar Kelimeler: Bakır, B<sub>4</sub>C, plazma sprey kapmala, yüzey kaplama, korozyon

# **1. INTRODUCTION**

Economic losses caused directly or indirectly by the corrosion cause high costs. In a sense, the economic losses of corrosion include corrosion-resistant materials, surface coatings, additions to aggressive environments in order to reduce the effectiveness of the environment and replacement of the deformed parts that cannot perform their duties with new ones. The studies have showed that non-metallic materials are similarly affected by environmental factors. For example, the methods used to describe the deformation of metal and its alloys as a result of stress corrosion can be applied successfully to glass, ceramic materials, polymers and compound materials. For this reason, the term corrosion is used in a way to cover the deformation of all materials having the quality of construction material due to the environmental effect. Studies have shown that non-metallic materials are similarly affected by environmental factors. For example; The methods used to describe the corrosion corrosion deterioration of metal and its alloys can be applied successfully to glass, ceramic materials, polymers and compound material and its alloys can be applied successfully to glass, ceramic materials, polymers and compound materials and its alloys can be applied successfully to glass, ceramic materials, polymers and compound materials. Therefore, the term corrosion is used to cover all materials bearing the quality of the building material by the environmental effects. Coatings are an engineering solution to improve the surface of materials against wear, corrosion, degredation and other surface properties (Doruk, 1982).

The development of coating technology plays an important role in the prevention of undesirable damages on the surfaces of materials such as abrasion and corrosion. Thermal spray methods are widely used in coating methods to prevent damage caused by abrasion. Another important feature of thermal spray coatings is that they can be re-coated in the same place after the abrasion of the coatings. Nowadays, there are various methods as thermal spray coating methods. In each of these methods, the coating materials to be sprayed and the temperatures they reach vary, and each method is used in different areas. (Mishra, vd., 2005; Islak, 2018; Yilbas ve Arif, 2007).

The most important place of use of thermal spray method is to increase resistance against abrasion and maintenance. In the biomedical fields, in the automotive sector, the thermal spray method in the maintenance sector and in the dental

sector is widely used. Coatings made with thermal spray methods are highly cost-effective because they are removable (Charles, 1994). HVOF (High-velocity oxy-fuel) and Plasma-Spray methods are used in many areas to improve the surface properties of machine parts and to reusable and use worn parts. Plasma Spray and HVOF methods from thermal spray coatings are preferred in areas where very good quality coatings are used (Islak, 2013).

## 2. MATERIAL AND METHODS

Pure copper plates at 20 mm x 130 mm x 5 mm and 99.9% purity was selected as the substrate. Cu and  $B_4C$  powders with -90+45  $\mu$ m and -45+15  $\mu$ m grain sizes respectively were coated on the substrate using a plasma spray method.  $B_4C$  was added to Cu in proportions of 5, 10 and 20% by weight. In the production of the coating layers, a Sulzer Metco 9-MB model plasma spray coating system with a power of 70 kW was used. The flow rate of the argon gas used to produce the plasma beam was set at 42,5 l/min in all coatings. Spraying was made at a distance of 70 mm. The coating powders are injected externally into the gun.





The coated samples were sanded with coarse grained and fine sandpaper for corrosion tests and cleaned in an ultrasonic bath. The corrosion measurements were performed using the Reference 3000 Potentiostat / Galvanostat / ZRA corrosion system given in Figure 2.2. Corrosion tests were conducted after keeping the samples at room temperature ( $25 \,^{\circ}$ C) in a NaCI solution of 3.5 wt.% (pH 7-8) for 1 hour. A conventional three-electrode cell was used for all electrochemical measurements. Ag/AgCl electrode was used as the reference electrode and a carbon graphite was used as the counter electrode. Potentiodynamic sweep was performed with a sweep of 1mV/s sweep rate in the range of  $\pm 0.25$  V potential range according to Eocp. Anodic and cathodic Tafel regions conducting extrapolation were used in order to determine the corrosion rates and corrosion potentials. Polarization resistance values were calculated from linear regions of current-potential curves close to corrosion potential.



Figure 2.2.: Corrosion testing system

# 3. RESULTS AND DISCUSSION

When examining optical images of the samples coated with B4C using plasma spray method (Figure 3.1) was found between the coating interfaces of the samples and coatings were found to be successful.



Figure 3.1 (a) Cu-5 B<sub>4</sub>C coating (b) Cu-10 B<sub>4</sub>C coating, (c) Cu-20 B<sub>4</sub>C coating.

Figure 3.2 shows potential change polarization curves of the samples determined in 3.5% NaCI corrosive medium. The results of potentiodynamic polarization measurements are summarized in Table 3.1. Corrosion potential (Ekor), anodic and cathodic Tafel curves ( $\beta a$  and  $\beta c$ ), corrosion resistance (Rp), corrosion rate and corrosion current (Ikor) were found from Tafel curves. Corrosion resistance was calculated by the Stern and Geary equation given below (Stern M.,Geary A. L., 1957).





$$I_{kor} = \frac{\beta_a x \beta_c}{2.303 x R_p (\beta_a + \beta_c)}$$

(3.1)

Corrosion rates in coatings generally depend on the amount of porosity and microcracks that occur in the characteristics of the coatings. Porosities and microcracks significantly reduce the corrosion resistance of coatings (Zhang and Kong; 2018).

| Numuneler | Ecor<br>(mV) | Icor (µAcm <sup>-2</sup> ) | βa (V/decade)        | βc (V/decade)         | Korozyon Hızı<br>(mpy) |
|-----------|--------------|----------------------------|----------------------|-----------------------|------------------------|
| Cu        | -177         | 1,96                       | 24,5 e <sup>-3</sup> | 55,2 e <sup>-3</sup>  | 1,195                  |
| B5        | -167         | 17,5                       | 15,7 e <sup>-3</sup> | 73,7 e <sup>-3</sup>  | 10,66                  |
| B10       | -143         | 31                         | 37,7 e <sup>-3</sup> | 290,8 e <sup>-3</sup> | 18,93                  |
| B20       | -165         | 51,5                       | 32,6 e <sup>-3</sup> | 58,1 e <sup>-3</sup>  | 31,34                  |

Table 3.1. Electrochemical results of B<sub>4</sub>C coating samples

When Figure 3.2 and Table 3.1 are examined, it is seen that the lowest current density is in uncoated material. Current density is increased with coating of samples. This may be due to the porosities that occur in the plasma spray coating and to the micro cracks that occur due to the thermal stress differences of the phases, to the deeper penetration of aggressive chlorine ions. Corrosion rates in coatings generally depend on the amount of porosity and microcracks, which are the characteristic features of the coatings. Porosities and microcracks significantly reduce the corrosion resistance of coatings (Kariofillis et al., 2006, Zhang and Kong; 2018).

#### 4. CONCLUSION

- Coating of Cu substrate with Cu-B<sub>4</sub>C powder mixture resulted in a significant increase in the hardness.
- Lowest current density is in uncoated material. Current density is increased with coating of samples.

#### REFERENCES

Charles, P. H. (1994). Thermal Spraying: Process, Preparation, Coatings and Application. Welding Journal, 73(4), 47-51.

Doruk, M., (1982). " Corrosion and Its Prevention", METU Publications, 20-29, Ankara.

- Islak, S., & Buytoz, S. (2013). Microstructure properties of HVOF-sprayed NiCrBSi/WCCo-based composite coatings on AISI 1040 steel. Optoelectronics and Advanced Materials-Rapid Communications, 7(11-12), 900-903.
- Islak, S., Çalıgülü, U., Hraam, H. R., Özorak, C., & Koç, V. (2018). Electrical Conductivity and Microstructure Properties of Cu-Mo Coatings, 1st International Symposium on Light Alloys and Composite Materials (ISLAC'18), March 22-24, 2018 Karabük, Turkey
- Kariofillis, G. K., Kiourtsidis, G. E., & Tsipas, D. N. (2006). Corrosion behavior of borided AISI H13 hot work steel. Surface and Coatings Technology, 201(1-2), 19-24.
- Mishra, S. B., Chandra, K., Prakash, S., & Venkataraman, B. (2005). Characterisation and erosion behaviour of a plasma sprayed Ni3Al coating on a Fe-based superalloy. Materials letters, 59(28), 3694-3698.
- Stern M., Geary A. L., (1957). Electro chemical Polarization I. A Theoretical Analysis of the Shape of Polarization Curves. Journal of the Electro Chemical Society 104 56-63.
- Yilbas, B. S., & Arif, A. F. M. (2007). Residual stress analysis for hvof diamalloy 1005 coating on Ti–6Al–4V alloy. Surface and Coatings Technology, 202(3), 559-568.
- Zhang, D., & Kong, D. (2018). Microstructures and immersion corrosion behavior of laser thermal sprayed amorphous Al-Nicoatings in 3.5% NaCl solution. Journal of Alloys and Compounds, 735, 1-12. Kirik, I., Ozdemir, N., Sarsilmaz, F., (2012). Microstructure and Mechanical Behaviour of Friction Welded AISI 2205/AISI 1040 Steel Joints. MP Material Testing, Vol. 54(10), pp. 683-688.



# Investigation of Corrosion Resistance of AISI 304 and AISI 316L Austenitic Stainless Steel Surface Coating with Cr<sub>3</sub>C<sub>2</sub> Powders Using HVOF Thermal Spray Coating Method

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Abstract: In this study, one of the thermal spray methods, HVOF spraying method was used. AISI 304 and AISI 316L austenitic stainless steels were used as base material. The samples were covered with  $Cr_3C_2$  by HVOF Thermal spray coating method. The coating thicknesses range from 0-100 $\mu$ m, 100-200 $\mu$ m, 200-300 $\mu$ m. Corrosion resistance of the samples were investigated after the coating process. Potentiodynamic method was used for corrosion. For the microstructure properties of the produced coatings, optical microscope and SEM analyzes were used. Corrosion resistance increased with the increase of coating thickness. It has been observed that AISI 304 stainless steel has higher corrosion resistance than AISI 316L stainless steel.

Keywords: Corrosion resistance, thermal spray coating, HVOF, stainless steel, Cr<sub>3</sub>C<sub>2</sub>

# 1. INTRODUCTION

Economic losses caused directly or indirectly by the corrosion cause high costs. In a sense, the economic losses of corrosion include corrosion-resistant materials, surface coatings, additions to aggressive environments in order to reduce the effectiveness of the environment and replacement of the deformed parts that cannot perform their duties with new ones. The studies have showed that non-metallic materials are similarly affected by environmental factors. For example, the methods used to describe the deformation of metal and its alloys as a result of stress corrosion can be applied successfully to glass, ceramic materials, polymers and compound materials. For this reason, the term corrosion is used in a way to cover the deformation of all materials having the quality of construction material due to the environmental effect (Doruk,1982, Günen, 2012).

Nickel is the second most important alloying element of stainless steels. The oxide layer on the surface of stainless steel forming in the environment with oxygen regains its old feature by fixing itself when it is deformed. Nickel improves the re-formation of the oxide layer, especially in environments that have an effect of degrading the oxide layer; however, if the nickel rate exceeds 8-10%, the stress corrosion resistance of the steel deteriorates. As the nickel ratio increases to around 30%, the stressed corrosion resistance improves again. Nickel improves the stainless steel's yield point, toughness and resistance to acids degrading oxide layer (Aran, 2004; Can, 2006; Aslan 2015).

Coatings are an engineering solution for improving the material surfaces against wear, corrosion, degradation and other surface events. Thermal spray is the general name of a group of processes in which metallic or non-metallic surface coating materials are molten or semi- molten by a heat source and applied to form a coating on a previously prepared surface. The surface coating material may be in powder, wire or rod form. The thermal spray gun produces the required heat with a flammable gas or electric arc. When heated, the coating materials become semi- molten, accelerated with the help of the gases used in the process and moved towards the surface of the base material. The particles flatten by hitting the surface, form thin plates and stick to the rough surface and to each other. When the particles sprayed hit to the surface of the base material, they cool, solidify and form a lamella structure which forms the coating. The adhesion of each particle takes place with mechanical bonding or metallurgical bonding or diffusion in some cases. Increasing speed of the particles provides a better adhesion strength and higher density. In order to ensure a good bonding between the base material and the coating, it is extremely important to roughen the surface of the base material by sandblasting and to

remove and clean the oil completely before coating. Figure 1.1 shows a schematic view of the HVOF Spray method (Esschnauer, 1985; Lugscheider, 1993; AWS, 1997; Smolka, 2002; Büyükleblebici, 2015; Kambur, 2018)



## Figure 1.1. HVOF spray system

HVOF spraying technique is the general definition for different metallic or non-metallic coating application group in the molten or semi-molten state on a substrate prepared for spray accumulation. Molten or semi-molten particles are rapidly directed to the previously prepared surface with the gases and atomized jets used. A mechanical bonding occurs at the interface with the hitting of molten or semi-molten particles to the surface. A coating structure forms with repeating hits and bonding of the directed particles. These finely adherent particles suddenly solidify on the surface of the substrate by showing a very rapid cooling regime. Thus, the spraying process is completed (Sarıkaya, 2003; Sert, 2007; Özel, 2009; Özorak, 2017).

SONG B. et al. (2017); In their study, they coated the surface of ASME P92 steel with Ni50Cr powder using HVOF technique. They carried out the coating process using two different fuels, gas and liquid. As a result of the work they carried out using gas fuel coatings; observed that more oxide is formed, the pores are spread over larger areas and have lower corrosion resistance (Song, Vd., 2017).BUYTOZ S. et al. (2013); In their study, they coated the surface of SAE 1030 steel with NiCrBSi i SiC powders by HVOF method. They found that the degree of mixing of NiCrBSi and SiC components in the powder had a great effect on the phase composition, microstructure and hardness of the coatings (Buytoz, Vd., 2013).ISLAK S. et al. (2015); In their study, they covered the surface of AISI 1050 steel using WCCo and SiC powders by HVOF method. As a result of the study they have done XRD analysis and on the surface of the sample WC, W<sub>2</sub>C, SiC, Cr<sub>5</sub>B<sub>3</sub>,  $\gamma$ -Ni and Co phases have occurred. Microstructure and micro hardness of the SiC and WCCo have determined that depends on amount of the powder (Islak, Vd., 2015).

AISI 304 and AISI 316L austenitic stainless steels are used in the flue gases in the casting industry. In this study, was to improve the surface properties of the AISI 304 and AISI 316L austenitic stainless steels by coating with the  $Cr_3C_2$  powders in the thickness of 0-100µm, 100-200µm and 200-300µm by using the HVOF spray method, one of thermal spray methods. The results obtained were examined and it was observed that the corrosion resistance increased with the coating thickness treatment in both  $Cr_3C_2$  coated materials and AISI 304 stainless steel had higher corrosion resistance than AISI 316L stainless steel.

### 2. MATERIAL AND METHODS

After the AISI 304 and AISI 316L Austenitic Stainless Steel plates were cut, their surfaces were coated with  $Cr_3C_2$  powders, whose properties were shown in Table 3.1, to have different coating thicknesses by using a HVOF Spray coating device setup in a private company in Figure 3.1. Table 3.2 shows the samples used in the experimental study. SEM micrographs and X-ray diffraction patterns of Al2O3/13wt.%TiO2 powder are shown in Figure 3.2. Powders are in different grain sizes, irregular and sharp-edged. As was evident from the XRD data, Al2O3/13wt.%TiO2 powder consists of a dense  $\alpha$ -Al2O3 and rutile-(TiO2) phases.

Table 2.1. Properties of powders used in experimental studies

| Oxide Powders                         | Powder Grain Size              | Powder<br>Morphology | Chemical Composition |
|---------------------------------------|--------------------------------|----------------------|----------------------|
| Aluminum Oxide<br>Al2O3<br>Amdry 6220 | -45 + 15μm(-325 mesh+<br>15μm) | Spherical, sintered  | Cr3C2(Ni 20Cr)       |

Table 2.2. Samples used in the experimental study

| Material  | 0-100µm        | 100-200µm      | 200-300µm      |
|-----------|----------------|----------------|----------------|
| AISI 304  | N <sub>1</sub> | N <sub>2</sub> | N <sub>3</sub> |
| AISI 316L | N <sub>4</sub> | N5             | N <sub>6</sub> |





The coated samples were sanded with coarse grained and fine sandpaper for corrosion tests and cleaned in an ultrasonic bath. The corrosion measurements were performed using the Reference 3000 Potentiostat / Galvanostat / ZRA corrosion system given in Figure 3.2. Corrosion tests were conducted after keeping the samples at room temperature ( $25 \,^{\circ}$ C) in a NaCI solution of 3.5 wt.% (pH 3) for 1 hour. A conventional three-electrode cell was used for all electrochemical measurements. Ag/AgCl electrode was used as the reference electrode and a carbon graphite was used as the counter electrode. Potentiodynamic sweep was performed with a sweep of 1mV/s sweep rate in the range of  $\pm 0.25$  V potential range according to Eocp. Anodic and cathodic Tafel regions conducting extrapolation were used in order to determine the corrosion rates and corrosion potentials. Polarization resistance values were calculated from linear regions of current-potential curves close to corrosion potential.



Figure 3.2. Corrosion testing system

# 3. RESULTS AND DISCUSSION

When examining SEM images of the samples coated with  $Cr_3C_2$  using HVOF spray method (Figure 3.1 and Figure 3.2), no gap was found between the coating interfaces of the samples and coatings were found to be successful.







**Figure 3.2**. Optical (50x) and SEM (750x) image of AISI 316L samples coated with Cr<sub>3</sub>C<sub>2</sub> using HVOF Spray Coating method

Figure 3.3 shows potential change polarization curves of the samples determined in 3.5% NaCI corrosive medium. The results of potentiodynamic polarization measurements are summarized in Table 3.1. Corrosion potential (Ekor), anodic and cathodic Tafel curves ( $\beta a$  and  $\beta c$ ), corrosion resistance (Rp), corrosion rate and corrosion current (Ikor) were found from Tafel curves. Corrosion resistance was calculated by the Stern and Geary equation given below [14].



# Figure 3.3 Tafel Polarization Curve

$$I_{kor} = \frac{\beta_a x \beta_c}{2.303 x R_p \left(\beta_a + \beta_c\right)}$$
(3.1)

Corrosion rates in coatings generally depend on the amount of porosity and microcracks that occur in the characteristics of the coatings. Porosities and microcracks significantly reduce the corrosion resistance of coatings [15].

| Samples | Samples    | Ecor  | Icor     | βa (mV) | $\beta c (mV)$ | Corrosion rate | Corrosion  |
|---------|------------|-------|----------|---------|----------------|----------------|------------|
| number  |            | (mV)  | (µAcm-2) |         |                | (mpy)          | resistance |
|         |            |       |          |         |                |                | (kΩ.cm2)   |
| N1      | AISI 304 - | -118  | 3,65     | 188,1   | 122,1          | 12,66          | 8,8080     |
|         | 100 micron |       |          |         |                |                |            |
| N2      | AISI 304 - | -200  | 3,23     | 187,8   | 100,7          | 11,2           | 8,8122     |
|         | 200 micron |       |          |         |                |                |            |
| N3      | AISI 304 - | -56,4 | 1,94     | 183,6   | 132,5          | 6,741          | 17,2254    |
|         | 300 micron |       |          |         |                |                |            |
| N4      | AISI 316 - | 12.5  | 17 1     | 2967    | 220.1          | 50.51          | 3 7517     |
|         | 100 micron | -12,5 | 17,1     | 580,7   | 239,1          | 59,51          | 5,7517     |
| N5      | AISI 316 - | -118  | 12       | 313,7   | 132            | 41,59          | 3,3618     |
|         | 200 micron |       |          |         |                |                |            |
| N6      | AISI 316 - | 43,5  | 7,17     | 521,2   | 148,9          | 24,89          | 7,0137     |
|         | 300 micron |       |          |         |                |                |            |

Table 3.1. Electrochemical results of Cr<sub>3</sub>C<sub>2</sub> coating samples

It was determined from the results of corrosion test that corrosion resistance of AISI 304 austenitic stainless steel was higher than AISI 316 L austenitic stainless steel in coated steels. In addition, it was observed that the highest corrosion resistance (200-300 $\mu$ m) belongs to the Cr<sub>3</sub>C<sub>2</sub> coated sample. Pores and micro-cracks caused by thermal stress differences of the pores and phases in thermal spray coatings allow aggressive chlorine ions to reach deeper (Zhang, 2018).

#### 4. CONCLUSION

As a result of the experiments and investigations, AISI 304 and AISI 316L austenitic stainless steel surfaces were successfully coated with the HVOF method using Cr3C2 powders. It was determined that the corrosion resistance increased with the increase of the coating thickness. In addition, it was concluded that AISI 304 austenitic stainless steels have a higher corrosion resistance than AISI 316L austenitic stainless steel after coating.

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#### REFERENCES

Aran, A., Temel, M.A., (2004). Production, Use and Standards of Stainless Steels, Sarıtaş Technical Publication, Istanbul.

Aslan O., (2015). Examining Corrosion Behaviors of Single and Double layered AISI 316L Stainless Steel Coated by Plasma Spray Coating, Afyon Kocatepe University, Afyon.

AWS Committee, (1997), Thermal Spraying: Practice, Theory, and Application, American Welding Society Inc, 12-15.

- Buytoz, S., Ulutan, M., Islak, S., Kurt, B., ve Çelik, O.N., (2013). Microstructural and Wear Characteristics of High Velocity Oxygen Fuel (HVOF) Sprayed NiCrBSi–SiC Composite Coating on SAE 1030 Steel. Arabian Journal for Science and Engineering, 38(6) 1481–1491.
- Büyükleblebici, B., (2015). Ti6Al4V Alaşımının HVOF Tekniğiyle B2O3 Ve Al2O3 Katkılı Hidroksiapatit Kaplanması Ve Yüzey Karakterizasyonu. Afyon Kocatepe University, Afyon.

Can, A.Ç., (2006). Material Science, Birsen Publishing house, Istanbul.

Doruk, M., (1982). " Corrosion and Its Prevention", METU Publications, 20-29, Ankara.

Esschnauer H., Lugscheider E., (1985). Fortschritte beim thermisvhen Spritzen, Metal Heft 3, 218-224.

- Günen, A., (2012). Nano Bor Tozu İle Yüzeyi Alaşımlandırılan Östenitik Paslanmaz Çeliğin Mekanik Özellikler Ve Korozyon Davranışının Araştırılması. Fırat University, Elazığ.
- Islak, S., Özorak, C., Küçük, Ö., Akkaş, M., ve Sezgin, C., (2015). Microstructure and Hardness Properties of high velocity oxygen fuel (HVOF) Sprayed WCCo-SiC Coatings. KUJES, 1, 1–7.

- Kambur, K., (2018). The Effect of Different Substrate Temperatures on Coating Properties in Coating of Ti6Al4V alloy with HA. Master's thesis, Manisa Celal Bayar University, Turkey.
- Lugscheider E., Jokiel P., (1993). Plasmaspritzen-Verfahren, Anwendungen, Enteicklungen, Metal Heft 3, 230-235.
- Özel, S., (2009). Alüminyum Alaşımı Ve Bronzu Yüzeyine Oksit Ve Karbür Bileşiklerinin Plazma Sprey Yöntemiyle Kaplanmasının Araştırılması, Fırat University, Elazığ.
- Özorak, E., (2017). Weldox Çeliği Yüzeyinde Hvof İle Üretilen Kaplamaların Mikroyapı ve Aşınma Özelliklerinin Araştırılması. Kastamonu Üniversity, Turkey.
- Sarıkaya, Ö., (2003). Al-12Si Malzeme Üzerine Plazma Püskürtme Tekniği ile AlSi+B4C Kaplama, Sakarya Üniversity, Turkey.
- Sert, Y., (2007). Examining Wear Properties of Textile Piece Coated with Plasma Spray Technique, Master's thesis, Sakarya University Institute of Science, Sakarya.
- Smolka K., Thermisches Spritzen, (2002). DVS-Verlag, 50-57.
- Song, B., Pala, Z., Voisey, K.T., ve Hussain, T., (2017). Gas and Liquid-Fuelled HVOF Spraying of Ni50Cr Coating: Microstructure and High Temperature Oxidation, Surface and Coatings Technology318, 224–232.
- Stern M., Geary A. L., (1957). Electro chemical Polarization I. A Theoretical Analysis of the Shape of Polarization Curves. Journal of the Electro Chemical Society 104 56-63.
- Zhang, D., & Kong, D. (2018). Microstructures and immersion corrosion behavior of laser thermal sprayed amorphous Al-Nicoatings in 3.5% NaCl solution. Journal of Alloys and Compounds, 735, 1-12. Kirik, I., Ozdemir, N., Sarsilmaz, F., (2012). Microstructure and Mechanical Behaviour of Friction Welded AISI 2205/AISI 1040 Steel Joints. MP Material Testing, Vol. 54(10), pp. 683-688.



# **Determination of the Scattering Radiation Dose from CT in PET-CT Imaging**

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Abstract: There are many imaging modalities that use ionizing radiation in health science. One of the imaging methods using ionizing radiation is PET-CT. Today, positron emission tomography is used in the determination of cancer diseases. While positron emission tomography can detect the cancer tissue physiologically, its success in determining its anatomical position is limited. For this reason, PET-CT devices were produced by combining positron emission tomography and computed tomography. In PET-CT devices, the patient is injected with the positron emitting radiopharmaceutical prior to entering the imaging system. This radiopharmaceutical is kept more in the cancerous tissue while the other tissues are kept less. The patient immediately emits radiation to the environment immediately after the injection. This radiation is affected by both the patient and those around him. When the patient enters the PET-CT imaging system, it is also affected by the radiation emitted by the x-rays from the computed tomography, along with its radiation. However, x-rays are scattered from the patient or from some materials around the patient.

The main aim of this study is to determine the amount of radiation scattered in the computerized tomography imaging. For this purpose, Alderson rando phantom was used. Thermoluminescent detectors (TLD) were placed at different distances from the patient. Chest and head-neck imaging were performed in the phantom. The calibrations and evaluations of TLD detectors were carried out at the Cekmece nuclear research center.

The radiation concentration scattered across the chest imaging environment was found to range between 3.65  $\mu$ Sv and 25.18  $\mu$ Sv. In the head-neck imaging, it was found that the radiation concentration scattered around the environment varied between 6.56  $\mu$ Sv and 28.11  $\mu$ Sv.

Keywords: PET-CT, Radiation, TLD



# Nanostructured Fullerene-Like Metal Chalcogenides' Preparation Via Microwave Energy-Based Approach

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Abstract: Metal chalcogenides (MCs) have emerged as an extremely important class of materials that can be utilized within a large span of applications from lubrication to energy storage devices. Here, a recent discovery of a universal, ultrafast (60 s), energy-efficient, and facile technique to prepare MC nanostructures via microwave (MW) energy-based heating is introduced. A suitable combination of precursors is selected for the chemical intereactions on Polypyrrole nanofibers (PPy NFs) in MW medium. PPy NFs served as the conducting material to absorb MW energy, heat the precursors, and to reveal the metal and chalcogenide constituents. The MCs were formed as nanoparticles (NPs) that eventually undergo a size-dependent, multi-stage aggregation process to yield various MC nanostructures. Most importantly, this novel MC formation process is both proven to be much faster and energy-efficient than all the other existing methods and can be universally employed to produce different kinds of MCs (e.g. MoS2, and WS2), as well.

Keywords: Carbonization, conducting polymer, metal chalcogenide, microwave energy.

## **1. INTRODUCTION**

During the past decades, intense interests have been aroused in metal chalcogenides because of their unique properties and promising applications.1-4 The first closed-cage inorganic fullerene-like nanoparticles (IFNPs), layered MoS2 and WS2, were reported around 1990's.1-3 After a decade-long effort, many other layered compounds, such as transition metal chalcogenides MX2 (M = W, Mo, Sn, Ti, Re, Nb, Ta, Hf and Zr, etc.; X = S, Se),5 are now achievable. Recently, MX2 has been investigated extensively because of their unique layered structures and excellent physical and chemical properties, which make them ideal candidates for use as lubricants,6-9 heterogeneous catalysts,10-12 solar cells,13 lithium-ion batteries,14,15 hydrogen storage elements,16,17 and high performance protective composites.18-20

There has been significant progress made in synthetic methods for MS2 production, characterization, and in investigation of its physical and chemical properties as well as various applications. Synthetic methods for MS2 production mainly consist of (1) chemical methods such as solid-gas or gas-phase reactions, 3,21 thermal decomposition, 22,23 hydrothermal or solvothermal synthesis, 24,25 and template synthesis26 and (2) other instant stimulation methods such as laser ablation, 27 arc discharge, 7, 28 and electron beam irradiation. 29, 30 Despite major progress, there are still several limitations and disadvantages associated with the existing MS2 fabrication techniques. Some of them include the presence of extreme reaction conditions (high temperature, argon protection), involvement of many toxic and hazardous gases (e.g. H2S) and complicated processes, requirement of intense facilities (e.g. furnace, laser, arc discharge, and high voltage beams), and many more.31-35 These challenges warrant the need to develop a method that can synthesize high-purity but low cost products of MS2 at industrial scale.

### 2. MATERIALS AND METHODS

In this study, the discovery of a facile and energy-efficient route for the MW-assisted synthesis of IF-MS2 NPs is reported. The technique represents a clean energy nano-manufacturing approach by using MW heating.36-38 Very recently, there has been a major surge in employing MW-based ultrafast, energy-efficient, and facile approaches to synthesize multi-component nanostructures (e.g. metal oxides, carbon nanotube, carbon fiber, conducting polymer).39-43 Here, MW heating is utilized to synthesize IF-MS2 NPs using a highly non-intuitive, MW-assisted multi-scale aggregation dynamics. Figure 1 exhibits the schematic of the as-hypothesized mechanism of the process.



Figure 1. Schematic representation of the experimental process

It starts with a uniform blend of PPy NFs and  $M(CO)_6$  (M = Mo, W) forming homogenous dark powder composites. This blend is mixed with sulfur (S) powder or S/CS<sub>2</sub> solution, and is subjected to MW irradiation (frequency 2.45 GHz, power 1250 W) for 60 seconds. PPy NFs has been synthesized via previously developed seeding polymerization method<sup>42,43</sup> and serves as the substrate owing to its low cost, relatively high electrical conductivity, tunable de-/doping characteristics, and long-term environmental stability. MW heating triggers the reaction between  $M(CO)_6$  and S or S/CS<sub>2</sub> solution. This reaction first leads to the formation of MS<sub>2</sub> NPs, as  $M(CO)_6$  provides the metal element. These NPs undergo intense heating which promotes both molecular collisions and the formation of aggregates. In this first stage of aggregation, where the NP sizes are too small (~ few nms), van der Waals (vdW) interactions govern the aggregation of particles leading to spherical growth of the aggregate. The second stage of the aggregation involves NPs with much larger diameters (~100 nm) and the difference in respective surface energies (NP-air and NP-NP) becomes dominant to lead the directional growth of the aggregate. This results in ring-like growth of MoS<sub>2</sub> aggregates (see Figure 1), along with spherical growth of MoO<sub>x</sub> aggregates (not depicted in Figure 1), which is the product of MW heated PPy NF and M(CO)<sub>6</sub> mixture.

# 3. RESULTS AND DISCUSSION

The proposed MW-based nano-manufacturing set up is first calibrated by producing MoOx NPs. Figure 2 shows the SEM images of the as-produced MoOx particles decorated on the PPy NFs by using the MW heating-assisted reaction with Mo(CO)6 precursor.



**Figure 2.** SEM images of the  $MoO_x$  NPs decorated PPy NFs at different magnifications (a, b). (Inset: EDX spectrum of the as-formed  $MoO_x$  NPs)

The  $MoO_x$  particles uniformly distribute on the PPy NF network. On a given PPy NF, the  $MoO_x$  particles fully cover the surface of NFs. Here, the PPy NF serves to conduct the MW heat to  $Mo(CO)_6$ , yielding  $MoO_x$ . The inserted EDX spectrum is used to confirm the particles are mainly composed of Mo and O, while the presence of gold (Au) is due to the sputter coating applied prior to SEM imaging.

Using the same conditions and process,  $MoS_2$  particles can be easily produced by only adding S powder to the initial mixture. Figure 3 shows the SEM images of  $MoS_2$  particles on the "bed" of PPy NF. The  $MoS_2$  particles are aggregated together on PPy NF. The EDX result shown in Figure 3 indicates that particles are primarily composed of Mo and S.



Figure 3. SEM images of PPy NF and MoS<sub>2</sub> NPs. Inset: EDS of MoS<sub>2</sub> NPs

Advanced TEM has been widely utilized as a very powerful instrument for the analysis of NP synthesis in recent studies to determine the crystal structure and for surface reconstruction in both particles and thin films.<sup>45-47</sup> The MoS<sub>2</sub> particles' low and high magnification TEM images show that the particle size is ~200-300 nm (Figure 4). Typical HRTEM image of the same particles shows that the spacing of adjacent lattice is determined to be ~0.26 nm, which is in good agreement with the interplanar spacing of the MoS<sub>2</sub>'s (200) planes.<sup>48,49</sup>



**Figure 4.** (a) and (b) HRTEM images of  $MoS_2$  particles on PPy NFs, (d) and (e) are line profiles of the framed area in (c), (f) the schematic of  $MoS_2$  structure

A closer look at these NPs' TEM images (Figure 4 (b) and (c)) also reveals that the particles are hexagonal or heptagonal in shape and possess a compact multiwalled structure (> 25 walls) with a prominent oxide NP core. The line profile of the framed area (box 1) indicates that the spacing distance of the interlayer is ~0.62 nm (6.172 nm of 10 layers) [see Figure 4(d)], which is very close to the separation of the (002) planes of MoS<sub>2</sub> (c/2 lattice spacing). The hexagonal atomic arrangement shown Figure 4 (f) and the FFT pattern indicate that the basal plane of the synthesized thin film is (001), i.e. the c-axis of MoS<sub>2</sub> materials is perpendicular to the thin film. Each S center is pyramidal and connected to three Mo centers. The lattice constant is a = b = 0.315 nm. Along the wall, the distance between Mo is  $\frac{\sqrt{3}}{2}a = 0.273$  nm which is very close to the measured value of 0.261 nm [see Figure 4(e)]. The above mentioned results prove that the MW-assisted nano-manufacturing can easily provide the IF-like MoS<sub>2</sub> in a single step. To confirm its robustness, this method is also employed to prepare other MCs (e.g., IF-WS<sub>2</sub> from W(CO)<sub>6</sub> and S source). The size of both IF-WS<sub>2</sub> and IF-MoS<sub>2</sub> MCs is similar, but the shape of IF-WS<sub>2</sub> NPs is more spherical. The size of metal oxide core is much smaller than the one in IF-MoS<sub>2</sub> NPs. The multiwalled structure is not well compacted and well-organized, and the number of layers can be more than 40. The interlayer spacing is around 0.613 nm, and the distance between layers of W in one plane is around 0.22 nm; both of these values are smaller than that of IF-MoS<sub>2</sub> NPs'.

To the best of knowledge, in all the existing studies, the synthesis of IF-like  $MS_2$  is performed as a two-step process, including the synthesis of NPs being followed by an annealing process applied in an inert atmosphere inside a

conventional tube furnace.<sup>4, 50</sup> While in this novel approach, the temperature generated by MW heating can reach above 1000 °C for the direct synthesis of  $MoS_2$ .<sup>51</sup> The reaction is similar to the MOCVD which involves the reaction between organic gas and metal particles at very high temperature (>700 °C).<sup>50</sup> The schematic experimental procedure is shown in Figure 1. The particles of M(CO)<sub>6</sub> and S contact well with each other on the PPy NF network which provides the channel of conduction and pathway of heating. When MW energy is applied, the reactions occur and the amorphous nuclei are formed. Tangential MS<sub>2</sub> layers are being gradually formed from the edges. As shown in Figure 5, there are many different shapes and intermediates of MoS<sub>2</sub>, such as needle-like (N), onion-like (O), and irregular (S) shaped particles.<sup>50</sup> Such segments of multi-layers represent the different stages of MoS<sub>2</sub>, i.e. growth, assembly and formation, as discussed in Figure 1.



Figure 5. HRTEM image of MoS<sub>2</sub> particles with needle-like (N), onion-like (O), and irregular shaped (S) particles

Working principle of the same mechanism can be proven in  $WS_2$  growth, as well. Figure 6 clearly shows the intermediates of  $WS_2$  in the process of assembly.



Figure 6. HRTEM images of WS<sub>2</sub> particles with (a-c) growth and (d-f) assembly formations

Very thin layers (3-5 layers) are assembled from the edges of the amorphous particles and form irregular boundaries, as shown in Figure 6 (a-c). The crystallization process is shown to start at the outer surface of the round NP and propagate inwards. High temperature or thermal stress may initiate bending or faceting of the crystals. The strong interlayer covalent

bonding causes the crystal (layers) to close by itself and the structure is expected to be stable, as observed in Figure 6 (df). Finally, closed-cage IF-like  $WS_2$  NPs are formed. It should be noted that the particles in Figure 6 (a) are all in growth and assembly stages, while the particles in Figure 6 (d) exhibit the IF morphology. This indicates that these particles undergo uniform MW heating and concurrent morphological change steps.

Recently, MCs' NPs, tubes, and sheets were generated through a synthetic pathway that employs  $CS_2$  as the source of S instead of  $H_2S$ .<sup>4</sup> In this study,  $CS_2$  was also selected as co-precursor to obtain nanostructured MCs. The TEM images of IF-MoS<sub>2</sub>/WS<sub>2</sub> NPs synthesized from Mo(CO)<sub>6</sub> or W(CO)<sub>6</sub> with S/CS<sub>2</sub> solution indicate that both MC products are not well faceted, compared with the ones obtained from pure S powders. The number of layer is also less than those produced by S powders, as well. These cage-like materials possess different morphologies such as perfect spheres, semispherical particles or structures in which the crystalline directions were evident and resulted in polyhedral shapes. For IF-MoS<sub>2</sub>, the structure is loose and the metal oxide core is much larger than those obtained from the reaction with S powders. The results indicate that the use of S/CS<sub>2</sub> solution for the generation of MS<sub>2</sub> is very different from the solely use of S powder. Firstly, the S powders are homogeneously dispersed in CS<sub>2</sub> solvent, which can improve the uniformity of interactions between the metal and S. Secondly, CS<sub>2</sub> vapor may serve as additional S source to obtain this MCs. Thirdly, using S/CS<sub>2</sub> solution may provide a potential way to synthesize MCs with few layers or even with a single layer.

Central to this single-step formation of the MW-assisted MC nanostructures is the two-step aggregation process. The first stage of aggregation involves the *as-formed*  $MoS_2$  and  $MoO_x$  NPs with dimensions of only few nms. These NPs are at a thermally excited state; they vibrate rapidly as a consequence two of these NPs come to close enough proximity of each other to allow van der Waals (vdW) interactions to become influential triggering the aggregation of these two NPs. This aggregation displays no directional preference; as a result, the nanocluster grows in a spherical form. Things change when these nanoclusters have grown sufficiently large (~100 nm). As shown in Figure 7, when two typical spherical nanoclusters, each of which are 100 nm in diameter, come in close enough proximity, vdW effects dictate their possible aggregation. However, when a third nanocluster attempts to join this newly formed aggregate, two arrangements are possible, as illustrated in Figure 7.



Figure 7. Schematic representation of the two possible aggregation mechanisms of large (~100 nm) nanoclusters

As for the state A, two contact zones (each of area  $\delta$ ) are being created and the consequent change in the surface energy is  $\Delta U = 2\delta(\gamma_{pp}-\gamma_{pa})$ ; on the other hand for the state B, only one contact zone (of area  $\delta$ ) is formed and the consequent change in the surface energy is  $\Delta U = \delta(\gamma_{pp}-\gamma_{pa})$ . Here,  $\gamma_{pp}$  and  $\gamma_{pa}$  are the particle-particle and particle-air surface tension values, respectively. When  $\gamma_{pp} < \gamma_{pa}$ , state A occurs, since this leads to a larger energy decrease (as compared to state B) due to aggregation – this typically occurs for MoO<sub>x</sub>. On the other hand, when  $\gamma_{pp} > \gamma_{pa}$ , state B occurs, since this leads to a less energy increase (as compared to state A) due to aggregation – this typically occurs for MoO<sub>x</sub>. On the other hand, when  $\gamma_{pp} > \gamma_{pa}$ , state B occurs, since this leads to a less energy increase (as compared to state A) due to aggregation – this typically occurs for MoO<sub>x</sub> seems to grow as a bulk cluster, whereas MoS<sub>2</sub> grows in the form of a ring.

#### 4. CONCLUSION

To conclude, in this study, the discovery of a single-step, universal MW-assisted technique to produce different kinds of MC nanostructures is reported. The process is ultrafast, extremely energy efficient and is based on three key steps including; (a) MW-heating assisted reaction yielding MC NPs, (b) thermal vibration of these NPs to trigger the

aggregation and formation of larger sized nanoclusters, and (c) preferential growth of these nanoclusters dictated by the relative preference of surface interactions forming different nanostructures. The material characterization test results indicate that morphological specifications of these nanostructures strongly depend on the nature of the MCs and as well as the reactants used to obtain these MCs. It is anticipated that the discovery of this new technique to prepare MCs will catalyze substantial development in more widespread uses of MCs in more energy-efficient and easily-accessible approaches.

#### REFERENCES

- 1. Tenne, R., Margulis, L., Genut, M. & Hodes, G. (1992). Polyhedral and cylindrical structures of tungsten disulfide. Nature, 360: 444–446.
- 2. Margulis, L., Salitra, G., Tenne, R. & Talianker, M. (1993). Nested fullerene-like structures. Nature, 365: 113-114.
- Feldman, Y., Wasserman, E., Srolovitz, D. J. & Tenne, R. (1995). High-rate, gas-phase growth of MoS2 nested inorganic fullerenes and nanotubes. Science, 267: 222–225.
- 4. Tenne, R. & Redlich, M. (2010). Recent progress in the research of inorganic fullerene-like nanoparticles and inorganic nanotubes. Chem. Soc. Rev., 39: 1423–1434.
- 5. Shi, Y. F., Wan, Y. & Zhao, D. Y. (2011). Ordered mesoporous non-oxide materials. Chem. Soc. Rev., 40: 3854–3887.
- 6. Rapoport, L. et al. (1997). Hollow nanoparticles of WS2 as potential solid-state lubricants. Nature, 387: 791-793.
- 7. Chhowalla, M. & Amaratunga, G. A. (2000). Thin films of fullerene-like MoS2 nanoparticles with ultra-low friction and wear. Nature, 407: 164–167.
- Huang, H. D., Tu, J. P., Zou, T. Z., Zhang, L. L. & He, D. N. (2005). Friction and wear properties of IF-MoS2 as additive in paraffin oil. Tribol. Lett., 20: 247–250.
- 9. Adini, A. R., Redlich, M. & Tenne, R. (2011). Medical application of inorganic fullerene-like nanoparticles. J. Mater. Chem., 21: 15121–15131.
- Chinaelli, R. R., Berhault, G. & Torres, B. (2009). Unsupported transition metal sulfide catalysts: 100 years of science and application. Catal. Today, 147: 275–286.
- 11. Afanasiev, P., Rawas, L. & Vrint, M. (2002). Synthesis of dispersed Mo sulfides in the reactive fluxes containing liquid sulfur and alkali metal carbonates. Mater. Chem. Phys., 73: 295–300.
- 12. Mdleleni, M. M., Hyeon, T. & Suslick, K. S. (1998). Sonochemical synthesis of nanostructured molybdenum sulfide. J. Am. Chem. Soc., 120: 6189–6190.
- Levy, M. et al. (2010). Synthesis of Inorganic Fullerene-like Nanostructures by Concentrated Solar and Artificial Light. Isr. J. Chem., 50: 417–425.
- Wang, Z. et al. (2013). CTAB-assisted synthesis of single-layer MoS2-graphene composites as anode materials of Li-ion batteries. J. Mater. Chem. A, 1: 2202–2210.
- 15. Radisavljevic, B., Radenovic, A., Brivio, J., Giacometti, V. & Kis, A. (2011). Single-layer MoS2 transistors. Nat. Nanotechnol., 6: 147–150.
- 16. Chen, J., Li, S. L., Tao Z. L. & Gao, F. (2003). Low-temperature synthesis of titanium disulfide nanotubes. Chem. Commun., 8: 980–981.
- 17. Wang, D. Z. et al. (2013). Distorted MoS2 nanostructures: An efficient catalyst for the electrochemical hydrogen evolution reaction. Electrochem. Commun., 34: 219–222.
- 18. Zhu, Y. Q. et al. (2005). Shock-absorbing and failure mechanisms of WS2 and MoS2 nanoparticles with fullerene-like structures under shock wave pressure. J. Am. Chem. Soc., 127: 16263–16272.
- 19. Zhu, Y. Q. et al. (2003). Shock-wave resistance of WS2 nanotubes. J. Am. Chem. Soc., 125: 1329–1333.
- Cook, J., Rhyans, S., Roncase, L., Hobson, G. & Luhrs, C. C. (2014). Microstructural study of IF-WS2 failure modes. Inorganics, 2: 377–395.
- 21. Rothschild, A., Sloan J. & Tenne, R. (2000). Growth of WS2 Nanotubes Phases. J. Am. Chem. Soc., 122: 5169–5179.
- 22. Nath, M., Mukhopadhyay K. & Rao, C. N. R. (2002). Mo1-xWxS2 nanotubes and related structures. Chem. Phys. Lett., 35: 163-168.
- 23. Nath, M., Govindaraj, A. & Rao, C. N. R. (2001). Simple Synthesis of MoS2 and WS2 Nanotubes. Adv. Mater., 13: 283-286.
- Li, Y. D., Li, X. L., He, R. R., Zhu, J. & Deng. Z. X. (2002). Artificial lamellar mesostructures to WS2 nanotubes. J. Am. Chem. Soc., 124: 1411–1416.
- 25. Berntsen, N. et al. (2003). A solvothermal route to high-surface-area nanostructured MoS2. Chem. Mater., 15: 4498–4502.

- Zelenski, C. M. & Dorhout, P. K. (1998). Template synthesis of near-monodisperse microscale nanofibers and nanotubules of MoS2. J. Am. Chem. Soc., 120: 734–742.
- 27. Parilla, P. A. et al. (1999). The first true inorganic fullerenes? Nature, 397: 114.
- Hu, J. J., Bultman, J. E. & Zabinski, J. S. (2004). Inorganic fullerene-like nanoparticles produced by arc discharge in water with potential lubricating ability. Tribol. Lett., 17: 543–546.
- 29. Yacaman, M. J. et al. (1996). Studies of MoS2 structures produced by electron irradiation. Appl. Phys. Lett., 69: 1065–1067.
- Golberg, D., Bando, Y., Stephan, O. & Kurashima, K. (1998). Octahedral boron nitride fullerenes formed by electron beam irradiation. Appl. Phys. Lett., 73: 2441–2443.
- 31. Li, X. L. & Li, Y. D. (2003). Formation of MoS2 inorganic fullerenes (IFs) by the reaction of MoO3 nanobelts and S. Chem. Eur. J., 9: 2726–2731.
- Yang, H. B. et al. (2006). Synthesis of inorganic fullerene-like WS2 nanoparticles and their lubricating performance. Nanotechnology, 17: 1512–1519.
- 33. Leonard-Deepak, F., Castro-Guerrero, C. F., Mejia-Rosales, S. & Jose-Yacaman, M. (2011). Structural transformation of tungsten oxide nanourchins into IF–WS2 nanoparticles: an aberration corrected STEM study. Nanoscale, 3: 5076–5082.
- 34. Tian, Y. et al. (2005). A facile route to synthesis of MoS2 nanorods. Mater. Lett., 59: 3452–3455.
- 35. Wiesel, I. et al. (2009). Synthesis of WS2 and MoS2 fullerene-like nanoparticles from solid precursors. Nano. Res., 2: 416-424.
- 36. Zhu, Y. J. & Chen, F., (2014). Microwave-assisted preparation of inorganic nanostructures in liquid phase. Chem. Rev., 114: 6462.
- 37. Schwenke, A. M., Hoeppener, S., Schubert, U. S., (2015). Synthesis and modification of carbon nanomaterials utilizing microwave heating. Adv. Mater., 27: 4113.
- 38. Zhang, X. Y. & Liu, Z. (2012). Recent advances in microwave initiated synthesis of nanocarbon materials. Nanoscale, 4: 707–714.
- Zhang, X. Y. & Manohar, S. K. (2006). Microwave synthesis of nanocarbons from conducting polymers. Chem. Commun., 23: 2477–2479.
- 40. Liu, Z. et al. (2014). An ultrafast microwave approach towards multicomponent and multi-dimensional nanomaterials. RSC Adv., 4: 9308–9313.
- 41. Liu, Z. et al. (2014). Ultrafast Cr(VI) removal from polluted water by microwave synthesized iron oxide submicron wires. Chem. Commun., 50: 8036–8039.
- Zhang, X. Y., Goux, W. J. & Manohar, S. K. (2004). Synthesis of polyaniline nanofibers by "nanofiber seeding". J. Am. Chem. Soc., 126: 4502–4503.
- 43. Zhang, X. Y. & Manohar, S. K. (2004). Bulk synthesis of polypyrrole nanofibers by a seeding approach. J. Am. Chem. Soc., 126: 12714–12715.
- 44. Tarcha, P. J., Salvati, L. & Johnson, R. W. (2001). Polypyrrole latex: surface analysis by XPS, Surf. Sci. Spectra., 8: 323–328.
- 45. Kim, Y. J., Tao, R. Z., Klie, R. F. & Seidman, D. N. (2013). Direct atomic-scale imaging of hydrogen and oxygen interstitials in pure niobium using atom-probe tomography and aberration-corrected scanning transmission electron microscopy. ACS Nano, 7: 732–739.
- 46. Zhang, K. et al. (2014). Water-free Titania–Bronze thin films with superfast lithium-ion transport. Adv. Mater., 26: 7365–7370.
- 47. Xin, H. L. et al. (2014). Revealing the atomic restructuring of Pt-Co nanoparticles. Nano. Lett., 14: 3203-3207.
- Luo, W., Hu, X. L., Sun, Y. M. & Huang, Y. H. (2011). Electrospinning of carbon-coated MoO2 nanofibers with enhanced lithiumstorage properties. Phys. Chem. Phys., 13: 16735–16740.
- 49. Liu, X. L., Ji, W. X., Liang, J. Y., Peng, L. M. & Hou, W. H. (2014). MoO2@carbon hollow microspheres with tunable interiors and improved lithium-ion battery anode properties. Phys. Chem. Chem. Phys., 16: 20570–20577.
- 50. Zink, N. et al. (2008). In situ heating tem study of onion-like WS2 and MoS2 nanostructures obtained via MOCVD. Chem. Mater., 20: 65–71.
- 51. Liu, Z., Wang, J. L., Kushvaha, V., Poyraz, S., Tippur, H., Park, S. Y., Kim, M., Liu, Y., Bar, J., Chen, H., Zhang, X. Y., (2011). Poptube approach for ultrafast carbon nanotube growth. Chem. Commun., 47: 9912.


# Effects Of Process Parameters In The Machining of AA 7075 Alloy Reinforced with B4C Composite by Powder-Mixed Electrical Discharge Machining

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Abstract: In this study, machinability of boron carbide particle reinforced aluminum metal matrix composites (B4Cp / AA 7075) has been investigated by using powder-mixed electrical discharge machining (PMEDM). For this reason, AA 7075 reinforced with various rates of B4C (10% and 20%) with different sintering time (90 minutes, 120 minutes and 150 minutes) has been produced via powder metallurgy method. In order to obtain improved material removal rate (MRR) and decreased surface roughness, graphite powder with mean particle size of smaller than 50 nanometers were mixed in kerosene dielectric fluid and experiments were carried on using graphite electrode having 6 mm diameter. Samples were machined with different currents (2A, 4A and 8A) and material removal rates (MRR) values were calculated. After powder-mixed electrical discharge machining (PMEDM) process, machined surfaces were examined by scanning electron microscopy (SEM). Obtained results indicate that current and percentage of reinforced particle in composite have mostly significant influence on MRR. It was observed from results that MRR mostly increases with increasing of current values, on the other hand MRR decreases with percentage of reinforced particle in composite. In some cases, similar results were not observed due to the presence of low wettability and agglomeration problem in composite samples.

Keywords: Metal matrix composite, powder-mixed EDM, material removal rate, nano size graphite powder, SEM



# Turkey of Thorium Reserves and Thorium Based Reactor Design

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**Abstract:** The purpose of this study, Turkey is as important thorium reserves of the owners, according to international agreements, indigenous, peaceful and as clean energy source by allowing to take place in Turkey's energy system would be enable the development of technology and manpower can use us as an indigenous source of energy as well as external dependence of our energy It is a model for increasing our safety. At this point, having one of the world's richest reserves of thorium is that Turkey is no longer required to come to the stage of establishment of thorium research and production center. Thorium is defined as a nuclear fuel that is more abundant in the world than uranium and longevity, less toxic, less waste volume and the half-life of its waste is shorter than uranium waste. Energy need is one of the main problems of today as the reason of developing technology. As a new solution to this problem, the usability of the Thorium mine in nuclear power plants is an important step for our country, which is rich in world and thorium reserves. In this study, the usability, economic aspect, advantages and efficiency of thorium in nuclear power plants are investigated. Then of Turkey reserves and thorium-fueled nuclear power plant in Turkey could be established were tried to be determined by certain criteria. All of the results of the world, and increasingly it is seen that Turkey's energy need for further increases. However, to use their own resources to the shortage of rich thorium resources are considered new generation of reactor fuel in Turkey has emerged importance that should certainly evaluation.

Keywords: Thorium, Turkey, ADS, Reactor Design, Nuclear Power

## **1.INTRODUCTION**

Thorium (Th). The atomic number 90 is a radioactive element used as a source of atomic energy, atomic weight of about 232 g / mol, melting at 11.7 g / mL, melting at 1755 [deg.] C, not deteriorating in the air, as a source of atomic energy.[6]

Thorium is not capable of self-division. Therefore, it cannot be used directly as a nuclear fuel. The isotope 232Th (thorium-232) must be converted to 233U, which is a fission capable isotope by swallowing a neutron. As a result of the reaction of 232Th with low energy neutrons (neutron ingestion), first 233Th is formed which has less stability.

The half-life of 233Th, which is 23 minutes, is converted to 233Pa (Protactinium), with a half-life of 27 days, spreading a beta particle (b). 233Pa is transformed into 233U (a half-life of 163 thousand years) which can be divided by a beta and a gamma particle (g). Thus, it is used in combination with a fissile material such as 232Th, 235U or 239Pu (plutonium-239).[14]

Thorium is considered to be the cleanest fuel of nuclear power plants, as less plutonium and other trans-uranium elements are produced in the thorium fuel cycle than uranium. In the future, it is thought that nuclear reactors will be used instead of uranium in order to cause less harm to the environment.

Studies on the use of thorium as nuclear fuel are still ongoing. However, there is no commercially available nuclear reactor working with thorium today.[14]

### 2.MATERIALS AND METHODS

As a result of research conducted by MTA towards the end of 1959, in a 15 km2 area between Kızılcaören, Karkın and Okçu villages to the north - west of Sivrihisar district of Eskişehir, the complex built beds containing rare earth elements, barite and fluorite as well as thorium It was found.[15]



Figure 1. Thorium Reserve In Turkey

Thorium is found in nature about three times more than uranium. According to the 2006 data, it is possible to say that the total thorium reserve known in the world is 2.5 million tons and contains an average of 6-7% thorium oxide.[15]

| Table 1. | Toryum | Reserves | In | World[1] |  |
|----------|--------|----------|----|----------|--|
|----------|--------|----------|----|----------|--|

| COUNTRY            | RESERVE(Tonne<br>THO2) | %<br>PERCENT |  |
|--------------------|------------------------|--------------|--|
| AUSTRALIA          | 452000                 | 18           |  |
| USA                | 400000                 | 16           |  |
| TURKEY             | 380000                 | 15           |  |
| INDIA              | 319000                 | 13           |  |
| VENEZUELA          | 300000                 | 12           |  |
| BRAZIL             | 221000                 | 9            |  |
| NORWAY             | 132000                 | 5            |  |
| EGYPT              | 100000                 | 4            |  |
| RUSSIA             | 75000                  | 3            |  |
| OTHER<br>COUNTRIES | 149000                 | 5            |  |
| TOTAL              | 2528000                | 100          |  |



Chart 1. Thorium reserve percentages of countries[1]

Today, there is no commercial-scale nuclear power plant operating with thorium, as a result of which the consumption of thorium as energy raw material is almost non-existent.[6]

The economics of a thorium-based fuel cycle can only be possible with a nuclear program with a number of plants. For thorium-based power generation, there is a need to establish facilities that require high investment and operating costs.

Since all of these facilities are not economical in today's conditions, commercial scale technologies have not been developed in the world yet.[6] For this reason, the sale of thorium in our country as a product or mine is not today.

However, research and development studies should be continued in order to separate the thorium ore that is present in our country from rare earth elements and to obtain it as a by-product.[5]

Thorium itself is not a nuclear fuel, but fissile substances such as U-235 or Pu-239 are needed to be used as fuel. When used in combination with U-235 or Pu-239, it can save uranium need. Thorium use in open cycles is expected to save around 20-30% uranium. Separation and manufacturing technologies are needed for closed loop. Decomposition is a sensitive technology owned by some of the major countries and involves the risk of nuclear weapons. Today, the low cost of uranium fuel shows the use of uranium as the most rational way to produce nuclear power in the near future. However, parallel to the technological developments in the world, research and development studies on thorium based fuel cycle should be continued in our country.

Thorium reactor has safer aspects than conventional reactors. In conventional reactors, water is used for cooling and lithium fluoride salt is used in thorium reactor. This salt is not flammable.

The radioactive material is in the molten salt and the nuclear reaction takes place in this solution; no fuel rods. The fuel cools while passing through the heat exchanger containing molten salt and the heated salt is then used in the operation of turbines and in the production of electricity.

Since water is not used in cooling, it is not possible to vaporize and decompose the water due to vapor pressure to form a hazardous gas such as hydrogen.Nuclear waste can react again. therefore, the amount of nuclear waste is less. In conventional reactors, fuel rods are affected by radiation and they crack and leak.[5]

Explosions in conventional reactor accidents are caused by hot water vapor and hydrogen gas. This is what is in the fukushima reactor.

There is no danger in thorium reactor. The problem to be solved in the thorium reactor is that the liquid lithium fluoride is highly corrosive. For this purpose, wear-resistant nickel-molybdenum alloy protection container is used. But even this alloy is damaged.

USA, CHINA, INDIA The European Union is doing research in this direction. Turkey has joined the international project on innovative nuclear reactors and fuel cycle in 2001. In order to minimize the dangers created by the consumption society to the global reactors arising from the need for more energy, it is reasonable to choose this type of reactor in the way of development.

However, it is necessary to overcome difficult problems such as the development of thorium reactor technology, conversion of existing plants and storage of nuclear waste from them.

### **3. RESULT**

Considering the advantages of thorium; Security, natural security, Stable cooler, no fission pressure increase, it's easier to control, slow heating, proliferation resistance ... etc. are the features. [5]

Thorium in terms of economy and productivity; Thorium abundance, lack of natural resources, reactor efficiency, thermodynamic efficiency, no enrichment and fuel element manufacture, low fuel cost, LFTRs (Liquid Fluorinated Thorium Reactor) are cleaner, required less fissile fuel, no downtime for refueling, no high pressure pot, excellent heat transfer, smaller, lower pressure air casing, efficient mining. [5]

Looking at the negative aspects of thorium, it still needs to be developed a lot, starting fuel, freezing salts, Beryllium toxicity, loss of delayed neutrons, waste management, unclear usage costs, metal accumulation, limited graphite life, limited plutonium solubility, reprocessing risk of reprocessing, insoluble due to protactinium separation, business model, development of the energy cycle ... etc. situations need to be improved.[5]

Considering the toryum based reactor design works in the world, there is no reactor working with thorium yet. But they continue to work on nuclear power plant working with india(60% thorium in 2020, 40% uranium and plutonium) and chinese (100% thorium in 2030) thorium.

In our country, it is necessary to start the necessary studies based on the 3.5 and 4th generation reactor types. Because the increasing population ratio and this increase in the need for energy need to solve the problems that may be encountered in our country in the coming years, urgent decisions should be taken and concrete steps should be taken. These reactors of the new generation have no harm to the environment. The risk of explosion is low. Even in terms of reserves of thorium reactor to be used in Turkey is ranked 3rd in the world.

#### **4.REFERENCES**

- [1] Temurçin K.,2003, Nükleer Enerji ve Tartışmalar Işığında Türkiye'de Nükleer Enerji Gerçeği, Coğrafi Bilimler Dergisi, 1(2), 25-39.
- [2] Taner A.C.,2006, Yeni Nesil Nükleer Güç Reaktörleri, Fizik Mühendisleri Odası Yayınları, Faydalı Bilgiler.
- [3] Taner A.C,2016, Global Karbonsuz Toryum Yakıtlı Nükleer Güç Santralleri Elektrik Üretimi için Çin ve Hindistan'da Yürütülen Araştırma Geliştirme ARGE Faaliyetleri, Fizik Mühendisleri Odası Yayınları, Faydalı Bilgiler.
- [4] ERGÜN, S., & POLAT,2013, M. A. NÜKLEER ENERJİ VE TÜRKİYE'YE YANSIMALARI. İnönü University International Journal of Social Sciences, 34.
- [5] URL-1,http://nukleerakademi.org/nukleer-enerjinin-5-avantaji/
- [6] URL-2, https://www.enerjiportali.com/nukleer-enerjide-toryumun-yeri-nedir/
- [7] URL-3, https://sifiratikturkiye.net/nukleer-enerjinin-fayda-ve-zararlari/
- [8] URL-4,https://www.enerji.gov.tr/tr-TR/Sayfalar/Nukleer-Enerji
- [9]YARMAN, T. (2011), Geçmişte ve Bugün Nükleer Enerji Tartışması, Okan Üniversitesi Yayınları:14, 1. Basım, İstanbul.
- [10]AKKOYUNLU, A. (2006), "Türkiye'de Enerji Kaynakları ve Çevreye Etkileri", I. Ulusal Türkiye'de Enerji ve Kalkınma Sempozyumu, 26 Nisan, İstanbul, s.131-145.

[11]URL-5,www.nei.org/

[12]EİAŞ, 2010 Elektrik Üretim Sektör Raporu:5-8

[13] URL-6, www.trntp.org/turkiyede-nukleer-enerji/73-alt-yap-faaliyetleri.html

[14]URL-7,https://www.mta.gov.tr/v3.0/sayfalar/bilgi-merkezi/maden-serisi/Uranyum-Toryum.pdf,2017

[15]URL-8,http://www.taek.gov.tr/tr/2016-06-09-00-43-55/162-nukleer-yakit-cevrimi/1071-toryum.html



## Frequency Dependent Dielectric Properties of Bi1.75Pb0.25Sr2Ca2Cu3-xSnxO10+δ Structure

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Abstract: In this study, Bi1.75Pb0.25Sr2Ca2Cu3-xSnxO10+ $\delta$  glass ceramic structure; (x = 0.0, 0.3, 0.5) Depending on Cu3-x - Snx displacement, frequency-dependent (100Hz-2MHz), capacitance-conductivity measurements were taken at 75K and 125K temperature values. Dielectric constant real part ( $\epsilon$ '), virtual dielectric constant ( $\epsilon$ "), dielectric loss (tan $\delta$ ) and ac electrical conductivity ( $\sigma$ ac) frequency dependence were analyzed. Dielectric properties of samples were found to be dependent on frequency and temperature values. Sn doping had a negative capacitive effect and a decrease in capacitive effect was observed with the addition of Sn doping rate. The negative dielectric constant and the virtual negative dielectric constant were calculated from the negative capacitance values. Dielectric properties were shown at low frequency values and dielectric properties could not be observed at high frequency values. This situation is considered to pass the samples to the conductor state at high frequency values.

Keywords: Glass ceramic, Dielectric.



# Current-Voltage Characteristics of Au/Coronene/n-GaP/In Schottky Barrier Diodes

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Abstract: Coronene thin film on n-GaP substrate is deposited by using spin coating method. Gold (Au) and indium (In) metals are also prepared as Schottky contact and ohmic contact by using thermal evaporation, respectively. It has been ultimately produced Au/Coronene/n-GaP/In Schottky diode. The effect of the Coronene organic layer on rectifying junction parameters of Au/Coronene/n-GaP/Au Schottky diodes has been investigated using current-voltage (I-V) measurements at room temperature.

The barrier height and ideality factor values are found to be 1.016 eV and 2.64 from I-Vcharacteristics, respectively. These results are gained by the thermionic emission theory at room temperature. Two important parameters such as series resistance ( $R_s$ ) and shunt resistance ( $R_{sh}$ ) are also evaluated from the *I*–*V* characteristics. Furthermore, Cheung method is used to evaluate the Schottky barrier height and series resistance. Results are compared with literature. The obtained results support that the electronic properties of the GaP-based Schottky diode can be controlled by the interlayer organic layer (Coronene).

Keywords: Coronene, Schottky diode, barrier height, ideality factor, series resistance



# An Investigation on Effect of Homogenization Heat Treatment on the Microstructure and Mechanical Properties of As-Cast ZM21 Magnesium Alloy

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Abstract: In this study, it is aimed to investigate the effect of homogenization annealing applied to ZM21 (Mg-2.1Zn-1.10Mn) magnesium alloy on microstructure, hardness and mechanical properties. For this purpose, ZM21 magnesium alloy was melted in the induction furnace under a protective atmosphere and cast into a steel mold of Y form. Samples were homogenized for 16 hours at 400 ° C and then cooled in water. Microstructure studies such as optical, SEM, EDX as well as micro hardness (Vickers) and tensile tests (according to TS EN ISO 6892-1 standard, at room temperature with a strain rate  $10^{-3}s^{-1}$ ) were performed both as-cast and homogenized ZM21 Mg alloys. Fracture surface morphologies were also examined. Microstructure results showed that average grain size of as-cast and homogenized ZM21 alloy were measured 236.5 µm and 200.5 µm, respectively. i.e. the grains were reduced by approximately 15% by the homogenization process. MgZn secondary phase of microstructure is replaced by smaller  $\alpha$ -Mg and dispersed more homogeneously in structure. Hardness values of the as-cast and homogenized ZM21 alloys were 37.76 HV and 50.25 HV respectively. Due to the thinning of the grain structure might cause increasing in the hardness value of the homogenized ZM21 alloy. Mechanical test results demonstrate that tensile strength of 248.25 and 253.65 MPA, yield strength of 89.4 and 107.8 MPA, elongation values of 10.5% and 7.6% were obtained for as-cast and homogenized ZM21 samples, respectively. The increase in mechanical test results such as tensile strength, yield strength and hardness values by homogenization might be related to the reduction of internal stresses in the cast structure as well as the refinement of grains observed in the microstructure after homogenization heat treatment. According to the Hall-Petch equation, small grain size positively affects the mechanical strength.

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Keywords: ZM21, Magnesium alloy, As-cast, Homogenization, Microstructure, Mechanical Properties.



# Design and Implementation of Memory Controller for Josephson-CMOS Hybrid Memories

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Abstract: Rapid Single Flux Quantum (RSFQ) digital circuits which are capable of high-speed operation with low-power consumption have been developed as an alternative to CMOS integrated circuits. Unfortunately, because of the low integration density and low driving capability of SFQ circuits, realizing large-scale memories by using only SFQ circuits is tough. Josephson-CMOS hybrid memory hybridizing high-speed, low power SFQ circuits and high-density CMOS memories is presented as a solution to the large-scale memory problem in RSFQ digital systems. In this study, memory control unit which is working with 10 GHz clock is designed for Josephson-CMOS hybrid memory systems. The memory controller, which controls the data flow between the superconductor processing unit and the CMOS memory and changes depending on the capacity of the memory, is designed to be scalable using superconducting digital technology. In the first stage of the study, units of reading data from memory and writing data into memory were designed separately to include 4-bit data and 2-bit address signals. In the second stage of the study, by combining these units form a single circuit design called control unit was constituted which a total of 8-bit data and 2-bit address signal including 4-bit writing data and 4-bit reading data. By making analog and digital simulations of designed circuits, their workableness in 10 GHz had been tested.

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**Keywords:** Josephson-CMOS Hybrid Memory, Rapid Single Flux Quantum(RSFQ), Superconducting Electronics, Memory Control Unit



## **ORAL PRESENTATION**

### Adsorption of Remazol Yellow Onto Eggshell And Calcined Eggshell

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**Abstract:** In the present study, waste eggshell (ES) and calcined eggshell (CES) were chosen as adsorbent and Remazol Yellow as a model of organic pollutant. Calcined eggshell was obtained with the calcination of eggshell at 800 °C for 4 hours. The adsorbents were characterized with FTIR (Fourier Transform Infrared Spectroscopy) analysis technique. Adsorption studies were carried out in batch reactor at 50 ppm dye concentration, 0.1-0.5 g/L adsorbent dosage and 25-45 °C temperature. Dye removal increased with increasing of the amount of adsorbents. Increase of temperature was caused to increase of dye removal percent. Dye removal percent was obtained as 83 % and 88 % for ES and CES at 0.1 g adsorbent dosage and 45 °C, respectively. At the same experiment condition, maximum adsorption capacity was determined as 39 mg/g and 61 mg/g for ES and CES, respectively. Shortly, the CES adsorbed Remazol Yellow dye more than ES. The presence of the characteristic peaks of adsorbents and Remazol Yellow's adsorption on adsorbents were determined by FTIR analysis.

Keywords: Remazol Yellow, adsorption, eggshell, calcined eggshell

#### **1. INTRODUCTION**

Organic dyes are commonly used in many industrial areas such as textile, leather, medicine, food processing, dying and cosmetics. Most of them are toxic, carcinogenic, and mutagenic and a large amount of dyes in untreated wastewater are discharged into the ecological systems. This problem results in adverse effects on human and animal health. Therefore, it is important to remove them from wastewater (Kyzas and Kostoglou, 2014; Wen et al., 2017; Akti and Okur, 2018; Khalid et al., 2016; Al-Ghouti and Salih 2018). Physical, chemical and biological methods have been studied in order to removal of dyes from wastewater. Adsorption is a common and frequently used process for the cleansing of natural air and water through physical or chemical bonding between the adsorbate and adsorbent entities followed by final removal of the adsorbed material, thus causing the removal of pollutants in the form of adsorbates. The phenomenon of adsorption can be monitored through various physical and chemical methods. Moreover, this method of removing pollutants from wastewater is effective and low cost compared to other techniques. One of the most effective methods used for dye removal is adsorption (Guru and Dash, 2014). Besides, different sorbents such as activated carbon, zeolites, clay, fly ash, natural and biological materials, chitin, chitosan, graphene, polypyrrole and polyaniline composites have been used in the adsorption process (Gómez-Treviño et al., 2013; Aref et al., 2017; Wen et al., 2017; Akti and Okur, 2018). Though the most widely used adsorbent is commercial activated carbon, the cost and the difficulty of regeneration however, limit the application of activated carbon in some processes (Aref et al., 2017; Al-Ghouti and Salih 2018). So it is necessary to find more efficient and cheaper alternate adsorbent (Khalid et al., 2016; Al-Ghouti and Salih 2018). The eggshell, which is a low cost adsorbent, attracts attention because it has a removal efficiency often ranging from 94 to 98 %. Literature studies confirm that eggshells are very effective in removing heavy metals, dyestuffs and other contaminants. Approximately 85-95% of the eggshell is made up of calcium carbonate and is confirmed as an adsorbent in which the adsorption takes place mainly by the exchange reaction. Because eggshells are an agricultural waste; it is environmentally friendly and economically feasible. Therefore, studying and analyzing the structure of the eggshell is important to understand its properties as an adsorbent (Khalid et al., 2016; Al-Ghouti and Salih 2018).

The Remazol Yellow is an organic azo dye and a limited number of literature studies have been conducted with Remazol Yellow adsorption (Trujillo-Reyes et al., 2012; Gómez-Treviño, et al., 2013; Khalid et al., 2016). On the other hand, adsorption studies of Remazol Yellow on eggshell and calcined eggshells are not included in the literature. Trujillo-Reyes et al., 2012 has been studied adsorption of Remazol Yellow on different composite materials such as Fe-Ni, Fe-Cu, C/ Fe-Ni and C/Fe-Cu. Maximum adsorption capacity of 157.8 mg/g at 30 h with Fe-Ni nanoscale oxides material. Torres-Pérez et al., 2008 was obtained Remazol Yellow adsorption capacity as 3.61 and 12.72 for zeolitic material and

carbonaceous material/HCl materials, respectively. In the study performed by Gómez-Treviño et al., 2013, adsorption capacity of iron modified montmorillonite to Remazol Yellow observed as 8.08 mg/g. Also in a study used Al-SBA-15 material, the amount of Remazol Yellow adsorbed determined as 670 mg/g. In the study of Cestari et al., 2007, Remazol Yellow was adsorbed 30.26 mg/g on modified mesoporous silica.

### 2. MATERIAL AND METHODS

The eggshell samples were collected from household eggshell waste. Firstly, eggshell samples were rinsed several times with deionized water and then the samples were allowed to air at 105 °C for 24 hours and the dried eggshells were crushed. The calcined eggshell was obtained by calcination of the eggshell at 800 ° C for 4 hours. FTIR analyses of adsorbents were carried out by Bruker Vertex 70/70v instrument a resolution of 4 cm<sup>-1</sup> and averaged over 16 scans in the range of 400–4000 cm<sup>-1</sup> wavelength. Before analyses, 1 mg of samples was mixed with 99 mg KBr and then samples which are powder form were transferred to DRIFT cell. Remazol Yellow (C.I. Reactive Yellow 160) was chosen as a model organic pollutant (Fig. 1).



Figure 1. Molecular structure of Remazol Yellow (C.I. Reactive Yellow 160)

Adsorption experiments were carried out in batch reactor at temperature of 25, 35 and 45 °C. Two different adsorbents dosage (0.1 and 0.5 g/L) was used and dispersed into 100 mL of aqueous solution of Remazol Yellow with an initial concentration of 50 ppm. During adsorption experiments, 4 mL suspension was withdrawn from the reactor at predetermined time intervals and filtered using 0.45  $\mu$ m naylon filters (Milipore). The Remazol Yellow concentrations were measured using an UV–VIS spectrophotometer (Genesys 10S UV–VIS, Thermoscientific) at 429 nm wavelength. The dye degradation % was calculated by the following equation:

Degradation % =  $(C_o-C)/C_o*100$ 

The adsorption capacity of adsorbents  $q_{max}$  (mg/g) at time t (min) was determined according to the following equation:

#### $q_{max} = [(C_o - C) * V]/m$

where,  $C_o$  and C are the initial and final concentration of Remazol Yellow (mg/L), V is the solution volume (mL) and m is the weight of adsorbent (g).

### 3. RESULTS AND DISCUSSION

The effect of adsorbent dosage on the adsorption of Remazol Yellow was investigated at adsorbent dosage of 0.1 and 0.5 g/L (Fig. 2). It is clear that adsorption increased dramatically with the increasing adsorbent dosage which can be attributed to an increase in the accessible surface sites (Laabd et al., 2016; Akti and Okur, 2018). A five-fold increase in the amount of adsorbent resulted in an increase in dye removal about 1.5 times. In the amount of adsorbent of 0.1 g / L, while the eggshell had a rapid adsorption in the first 60 minutes and a 50% dye removal, after this period the adsorption rate slowed down. Dye removal reached to 60 % within 60 minutes by the increasing of adsorbent amount. With a calcined eggshell, a faster adsorption rate (within 30 minutes) and a dye removal of 57% were obtained compared to the eggshell. This value increased to 86 % with increasing the amount of adsorbent.



Figure 2. Effect of adsorbent dosage on dye degradation onto (a) eggshell and (b) calcined eggshell

Effect of temperature on the adsorption of Remazol Yellow is given in Fig.3. Increase of temperature was caused to increasing of dye removal. This is attributed to a result of an increase in the mobility of the dye molecules with an increase in the temperature. More number of molecules may acquire sufficient energy to undergo an interaction with active sites on the adsorbent surface (Shirsath et al., 2015). Dye removal percent was increased up to 83 % and % 88 with the increasing of temperature from 25 °C to 45 °C, respectively.

Adsorption capacity of adsorbents having different adsorbent dosage at 25 °C is given in Fig. 4 and results are tabulated in Table 1. By increasing the temperature from 25 °C to 45 °C, the adsorption capacity of the eggshell increased from 30.5 to 5 mg/g to 38.5 mg/g. Similarly, it increased from 47.5 mg/g to 60.9 mg/g for the calcined eggshell. In low adsorbent dosage, it was observed that higher adsorbent capacity.



Figure 3. Effect of temperature on dye degradation onto (a) eggshell and (b) calcined eggshell



Figure 4. Adsorption capacity of adsorbents at different adsorbent dosages and 25 °C.

Table 1. Dye adsorption capacity of adsorbents for 0.1 g/L adsorbent dosage.

| Adsorbent         | Adsorption capacity (mg/g) |       |       |  |
|-------------------|----------------------------|-------|-------|--|
|                   | 25 °C                      | 35 °C | 45 °C |  |
| Eggshell          | 30.5                       | 34.3  | 38.5  |  |
| Calcined Eggshell | 47.5                       | 52.6  | 60.9  |  |

#### FTIR spectrums of adsorbents and dye adsorbed adsorbents

The FTIR spectrums of original adsorbents and dye adsorbed adsorbents were plotted on Fig. 5. The peaks at 713, 875, 1433, 1797 and 2513 cm<sup>-1</sup> are characteristics peaks of eggshell which are associated with the vibrations of the carbonate groups. The broad band was observed between 3200 and 3700 cm<sup>-1</sup> corresponds to the O-H groups originating from water molecule. This band was disappeared after calcination treatment of eggshell at 800°C, because water molecule was removed from the structure of eggshell and this band was became sharp at 3700 cm<sup>-1</sup> in the calcined eggshell. And also the observed peak at 500–580 cm<sup>-1</sup> was assigned to Ca–O (Elkady et al., 2011; Tangboriboon et al., 2012). On the other hand, the peaks at 3290 and 1651 cm<sup>-1</sup> showed presence of amines in the eggshell membrane particle (Tsai et al., 2006). The peak intensity at 875 cm<sup>-1</sup> (C=C stretching from dye) in the calcined eggshell was decreased with dye adsorbed, while the peak intensity at 1797 cm<sup>-1</sup> (C=O stretching) was increased. On the other hand, the intensity of appeared peak at 3645 cm<sup>-1</sup> was determined very low; the peak at 3670 cm<sup>-1</sup> was shifted to 3697 cm<sup>-1</sup> and pulled down. This is probably due to the O-H groups within the dye structure (Elkady et al., 2011; Tangboriboon et al., 2012).



**Figure 5.** FTIR spectrums of (a) eggshell (b) calcined eggshell (c) dye adsorbed eggshell (d) dye adsorbed calcined eggshell (0.1 g/L adsorbent dosage, 25°C)

### 4. CONCLUSION

Remazol Yellow removal increased with increasing of the amount of adsorbents. Increase of temperature caused to increase of dye removal percent. Dye removal percent was obtained as 83 % and 88 % for ES and CES at 0.1 g adsorbent dosage and 45 °C, respectively. The maximum adsorption capacity for eggshell and calcined egg shell was determined as

39 mg/g and 61 mg/g, respectively. In short, the calcined eggshell adsorbed more dye than the eggshell and FTIR analysis results supported this situation.

#### REFERENCES

- Akti F & Okur M (2018). The Removal of Acid Violet 90 from Aqueous Solutions Using PANI and PANI/Clinoptilolite Composites: Isotherm and Kinetics, Journal of Polymers and the Environment 26: 4233-4242.
- Al-Ghouti M A & Nusrath R S (2018). Application of eggshell wastes for boron remediation from water, Journal of Molecular Liquids 256: 599-610.
- Aref L, Navarchian A H & Dadkhah D (2017). Adsorption of Crystal Violet Dye from Aqueous Solution by Poly(Acrylamide-co-Maleic Acid)/Montmorillonite Nanocomposite, Journal of Polymer and Environmet (2017) 25:628-639.
- Cestari A R, Vieira E F S, Vieira G S & Almeida L E (2007). Aggregation and adsorption of reactive dyes in the presence of an anionic surfactant on mesoporous aminopropyl silica Journal of Colloid and Interface Science 309: 402-411.
- Elkady M F, Ibrahim A M & Abd El-Latif M M (2011). Assessment of the adsorption kinetics, equilibrium and thermodynamic for the potential removal of reactive red dye using eggshell biocomposite beads, Desalination 278: 412–423.
- Gómez-Treviño A P D, Martínez-Miranda V& Solache-Ríos M (2013). Removal of remazol yellow from aqueous solutions by unmodified and stabilized iron modified clay, Applied Clay Science 80–81: 219-225.
- Guru P S & Dash S (2014).Sorption on eggshell waste—A review on ultrastructure, biomineralization and other applications, Advances in Colloid and Interface Science 209: 49–67.
- Kyzas G Z & Kostoglou M (2014). Green Adsorbents for Wastewaters: A Critical Review, Materials 7: 333-364.
- Khalid A, Fozia R, Cleo T G V M T P, Abdur R, Ana L S & Claudio A (2016). Aluminum doped mesoporous silica SBA-15 for the removal of remazol yellow dye from water, Microporous and Mesoporous Materials 236: 167-175.
- Laabd M, Ait Ahsaine H, El Jaouhari A, Bakiz B, Bazzaoui M, Ezahri M, Albourine A & Benlhachemi A (2016). Congo Red removal by PANi/Bi2WO6 nanocomposites: kinetic, equilibrium and thermodynamic studies, Journal of Environental Chemical Engineering 4:3096–3105.
- Ozbay N & Yargic A S (2015). Factorial experimental design for Remazol Yellow dye sorption using apple pulp/apple pulp carbontitanium dioxide co-sorbent, Journal of Cleaner Production, 100: 333-343.
- Shirsath S R, Patil A P, Bhanvase B A & Sonawane S H (2015). Ultrasonically prepared poly(acrylamide)-kaolin composite hydrogel for removal of crystal violet dye from wastewater, Journal of Environmental Chemical Engineering 3: 1152–1162.
- Tangboriboon N, Kunanuruksapong R & Sirivat A (2012). Preparation and properties of calcium oxide from eggshells via calcination, Materials Science-Poland, 30(4): 313-322.
- Torres-Pérez J, Solache-Ríos M & Colín-Cruz A (2008). Sorption and desorption of dye remazol yellow onto a Mexican surfactantmodified clinoptilolite-rich tuff and a carbonaceous material from pirólisis of sewage sludge, Water Air Soil Pollution 187: 303-313.
- Tsai W T, Yang J M, Lai C W, Cheng Y H, Lin C C & Yeh C W (2006). Characterization and adsorption properties of eggshells and eggshell membrane, Bioresource Technology 97: 488–493.
- Trujillo-Reyes J, Sánchez-Mendieta, V, Solache-Ríos M J & Colín-Cruz A (2012). Removal of remazol yellow from aqueous solution using Fe–Cu and Fe–Ni nanoscale oxides and their carbonaceous composites, Environmental Technology 33: 545-554.
- Weiwen H, XuehuaY, Qiong H, Jinming K, Lianzhi L, & Xueji Z (2017). Methyl Orange removal by a novel PEI-AuNPs-hemin nanocomposite, Journal of Environmental Sciences 53: 278-283.



# A Numerical Study for Different Humidity Ratios of Intake Air in A Cng Engine About the Effects on The Performance and Emission Values

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Abstract: In recent years, as a result of the decrease in fuel reserves in the world and increasing costs, scientists have tried to find alternatives to gasoline and diesel fuels. The most important of these alternative fuels is undoubtedly natural gas fuel. Because of its high global reserve and economic fuel, natural gas has become attractive. In addition, it has become very important because it meets the emission standards that are constantly shrinking due to being a green fuel. This study involves the conversion of a four-cylinder turbocharged four-stroke diesel engine, which is available in the Istanbul Technical University Mechanical Engineering Faculty Laboratory, to a completely natural gas-fueled gas engine that works with Otto-cycle principle. Diesel engine was modeled numerically with the help of a CFD (AVL Fire) program and the results were compared with experimental results and validation study of the model was done. Then, the amount of natural gas fuel corresponding to the bond energy of diesel fuel as energy under diesel engine operating conditions was calculated. At different compression ratios (10, 11, 12, 13, 14, 15 and 16), engine conversion was made with the acceptance of dry intake air and the engine being considered as natural suction. In the analyzes carried out at different compression ratios, the situation with the highest performance values in certain reference ignition advances (20 CA, 25 CA and 30 CA BTDC) was determined and the knock tendencies were checked with a written Python code. According to the results, at the 16 compression ratio values, both high performance values and no knock problems were observed. After this stage, the specific moisture content of the intake air was increased in different proportions (1%, 3%, 5%, 7%, 10%, 15%, 20%, 25% and 30%) and the most suitable results in terms of emissions were examined. According to the results, increasing the compression ratio rised the thermal efficiency, improved the burning efficiency and triggered the tendency to knock. The increment in specific humidity was highly influential in NOx emissions as it reduced the incylinder regional temperatures. However, falling temperatures reduced performance values. Since the increment in humidity ratio upper 25% significantly worsens the performance values, that limit should not be exceeded for the ideal specific humidity ratio. When both performance and emission values were considered together, 20% of the specific humidity rate was optimal. There was a 12.5% depreciation of the power value, while the NOx emissions improved by about 92.7%. In PM emissions, it showed about 60.5% lower results.

Keywords: CNG, Spark Ignition, Humidity Ratio, Emission, Numerical Study

## **1. INTRODUCTION**

Nowadays, shrinking emission standards have led scientists to do research on alternative fuels. Internal combustion engines have been generally considered with gasoline and diesel fuels until recently. However, the studies have shifted to different fuels due to the decrease in their reserves on the world and air pollution. One of these different fuels is natural gas fuel. Natural gas is an energy source that can be burned easily both in diesel cycle and otto cycle. In addition, the abundance of reserves on the world increases the interest in natural gas. The studies in the literature are based on the conversion of existing internal combustion engines to natural gas engines after some operations. For example; The compression ratio can be reduced while converting a diesel engine to the natural gas engine. This is because the compression ratio must be high in order to realize the combustion process in diesel engines. The compression ratio should be reduced if necessary to avoid a knock when converting. The opposite is the case for gasoline engines. The octane number of the natural gas fuel is higher than that of gasoline, which means that the knock resistance of the natural gas is also higher. Therefore, the compression ratio should be increased slightly in the conversion of gasoline engines.

The effect of natural gas fuel on the different injection advances and the formation of a mixture with air in different spray patterns was investigated and the effect on the performance and emission values were examined both experimentally and numerically. (Sevik Jr, 2017). In the study, it was stated that the spraying process after the intake valve closes caused the flame rates of the mixture to increase and the decrease in the formation time of the mixture as a result of the stratification of the flow led to decreases in the thermal efficiency. It was emphasized that increasing the advances of spraying led to higher thermal efficiency and it was said that the effect of the spraying process in the direction of the tumble movement affected the combustion positively.

The effects of the intake air moisture content on the engine emission values were investigated experimentally in a fourcylinder engine operated with compressed natural gas fuel. (Lee, 2018). NOx and PM emission measurements were obtained by sending the air to the engine by adjusting the various relative humidity levels and ensuring the operation of the engine at various power ranges. According to the results of the study, the increase of the intake air humidity caused a 91% decrease in NOx emissions and an increase of 7.4 mg in PM emissions.

The effects of different fuels and fuel mixtures on performance and emission values in a spark ignition engine were investigated numerically and the numerical results were confirmed by experimental results. (Majmudar, 2003). In study, the effects of ethanol/gasoline, methanol/gasoline and butanol/gasoline fuel mixtures and methane and gasoline fuels on their performance and emission values were investigated. It was stated that the air excess ratio in all fuel types gave the highest power between 0.833 and 1 values. It was emphasized that the most suitable value of the ignition timing was 20° CA BTDC and the increase of compression rate increased the thermal efficiency.

In this study, conversion of the diesel test engine, which is present in the engine laboratory of İstanbul Technical University Mechanical Engineering Department, to a natural gas engine operated by otto cycle. General characteristics of the test engine; 4-cylinder, 4-stroke, piston diameter 105 mm, piston stroke 115 mm, turbo-charged, with a compression ratio of 16.2:1, and a direct injection compression ignition engine with a motor volume of 3.908 liters. Firstly, a numerical model was created in a CFD program (AVL Fire) for the engine. The numerical results and experimental results were compared and validated. While the diesel engine was converted into a natural gas engine, the total energy contained in the fuel injected under diesel fuel conditions was considered as constant and the amount of natural gas fuel corresponding to this energy was calculated. In addition, the presence of turbo under the conditions of the diesel engine was not considered in the natural gas operating condition and was considered as a natural suction engine. In these conditions, performance values were calculated by reference ignition advances at various compression ratios. When examining these conditions, the intake air humidity was considered to be zero. Also, a Python code was checked to determine whether a knock was occurred in these cases. Since these conditions were done with dry intake air, NOx emissions were very high. The specific values of the amount of moisture present in the intake air were changed in various ratios and the optimum conditions in terms of emissions were investigated. In all analyzes, the presence of a knockout was controlled by a Python code created with the help of an empirical equation proposed by Douaud and Eyzat.

$$J = \frac{1}{Da} \left(\frac{MON}{100}\right)^{-3.4107} (10.2p)^{1.7} \exp\left(\frac{-E_g}{T_{mix}}\right) \frac{d\alpha}{6n}$$
(1)

In this equation, Da and Eg are constant values of 30.75 and 42000 respectively. MON is the motor octane number and the value for natural gas fuel is 130. In the above equation, if the J value is greater than one, there is a knock. If one and lower are, it means no knock.

#### 2. MATERIAL AND METHODS

#### **Engine Specifications and Models**

The study was carried out in two stages. In the first stage, a turbocharged 4-cylinder intercooler diesel engine located in the Engine Laboratory of Istanbul Technical University, which had experimental results, was modeled. And the modeled motor was compared with experimental results and validation study was performed. In the second stage, this diesel engine was transformed into a otto cycle engine burning with a completely natural gas fuel. The characteristics of the modeled diesel engine are shown in Table 1.

After creating the geometric model of the motor, the simulation parameters must be entered according to the values in the test conditions. Initial conditions are shown in Table 2. After the solutions were obtained in different cell numbers, the solution was provided independently of the cell number. In addition, the accuracy of the model was compared with the

experimental results and a validation study was conducted. Different cell sizes and corresponding cell numbers are shown in Table 3.

The higher the number of cells increases the time of analysis unnecessarily. The cell size and the corresponding cell number at which the results did not change were obtained as 0.00085 m and about 50000, respectively. The generated combustion chamber geometry is shown in Figure 1.

It was seen that the results in different cell numbers did not change after about 50000 cell numbers and all analyzes were analyzed in this cell number so that the analyzes did not take much time.

 Table 1. Engine Specifications

| Cylinder number                          | 4      |
|--|--------|
| Bore (mm)                                | 104    |
| Stroke (mm)                              | 115    |
| Connecting rod length (mm)               | 182    |
| Nozzle hole number                       | 4      |
| Spray angle                              | 160    |
| Spray cone angle                         | 6      |
| Nozzle hole diameter (mm)                | 0.16   |
| <b>Compression ratio</b>                 | 16.2:1 |
| Total cylinder volume (cm <sup>3</sup> ) | 3908   |

#### Table 2. Initial Conditions

| Intake pressure (bar)                 | 1.546    |
|---------------------------------------|----------|
| Intake temperature (K)                | 303      |
| Swirl ratio                           | 2        |
| IVC (CA BTDC)                         | 154      |
| EVO (CA ATDC)                         | 130      |
| Turbulence kinetic energy $(m^2/s^2)$ | 10       |
| Turbulence model                      | k-zeta-f |
| Injection timing (CA BTDC)            | 12       |
| Fuel temperature (K)                  | 330.15   |

#### Table 3. Cell Size for Analyzes

| Mean Cell Size | <b>Total Cell Number</b> |
|----------------|--------------------------|
| 0,00250 m      | 15000                    |
| 0,00150 m      | 25000                    |
| 0,00100 m      | 35000                    |
| 0,00085 m      | 50000                    |
| 0,00075 m      | 65000                    |
| 0,0005 m       | 100000                   |



Figure 1. Combustion Chamber Geometry

#### **Mathematical Model and Boundary Conditions**

After the geometric model is created, it is necessary to make the necessary mathematical assumptions for the simulation. The k-zeta-f four equality model proposed by Hanjelic was used as a turbulence model. The ECFM-3Z model, which a combustion model that is suitable for diffusion combustion and pre-mixed combustion, which is commonly used in the literature, was used as combustion model. While Zeldovich model was used for NOx emissions, Kinetic model was used for PM emissions. Models used for analysis and boundary conditions are shown in the Table 4.

Table 4. Boundary Conditions and Models

| Cylinder head temperature (K) | 550.15            |
|-------------------------------|-------------------|
| Piston wall temperature (K)   | 575.15            |
| Liner temperature (K)         | 475.15            |
| Turbulence model              | k-zeta f          |
| Evaporation model             | Dukowicz          |
| Spray break-up model          | KHRT              |
| Combustion model              | ECFM-3Z           |
| Nox model                     | Extended Zeldovic |
| SOOT model                    | Kinetic           |

The AVL Fire program analyzes conservation equations in three dimensions by finite volume method by accepting compressible flow. Conservation equations are conservation of mass, conservation of momentum (Navier-Stokes) and conservation of energy. These equations are shown below.

Mass continuity equation:

$$\frac{\partial \vec{\rho}}{\partial t} + \frac{\partial}{\partial x_j} \left( \vec{\rho} \vec{U}_j \right) = 0 \tag{2}$$

Momentum equation:

$$\vec{\rho} \frac{D\overrightarrow{U_{1}}}{Dt} = \vec{\rho} \frac{\overrightarrow{U_{1}}}{\partial t} + \vec{\rho} \overrightarrow{U_{j}} \frac{\partial \overrightarrow{U_{1}}}{\partial x_{j}} = \vec{\rho} g_{i} - \frac{\partial \vec{p}}{\partial x_{i}} + \frac{\partial}{\partial x_{j}} \left[ \mu \left( \frac{\partial \overrightarrow{U_{1}}}{\partial x_{j}} + \frac{\partial \overrightarrow{U_{j}}}{\partial x_{i}} - \frac{2}{3} \frac{\partial \overrightarrow{U_{k}}}{\partial x_{k}} \delta_{ij} \right) \right]$$
(3)

Energy equation:

$$\vec{\rho}\frac{\vec{DH}}{Dt} = \vec{\rho}\left(\frac{\vec{DH}}{\partial t} + \vec{U}_{j}\frac{\partial\vec{H}}{\partial x_{j}}\right) = \vec{\rho}\dot{q}_{g} + \frac{\partial\vec{p}}{\partial t} + \frac{\partial}{\partial x_{i}}\left(\vec{\tau}_{ij}\vec{U}_{j}\right) + \frac{\partial}{\partial x_{j}}\left(\lambda\frac{\partial\vec{T}}{\partial x_{j}}\right)$$
(4)

Concentration equation:

$$\vec{\rho} \frac{D\vec{C}}{Dt} = \vec{\rho} \frac{\partial \vec{C}}{\partial t} + \vec{\rho} \vec{U}_{j} \frac{\partial \vec{C}}{\partial x_{j}} = \vec{\rho} \vec{r} + \frac{\partial}{\partial x_{j}} \left( D \frac{\partial \vec{C}}{\partial x_{j}} \right)$$
(5)

#### **Model Validation**

The experimental results were compared with the numerical result. While validating the experimental study with numerical model, the differences between the pressure values and performance and emission values were compared in some reference crank angles ( $12^{\circ}CA$  BTDC, TDC and  $9^{\circ}$  CA ATDC). The pressures between the model and the experimental result at different crank angles are shown in Table 5. The results were confirmed with a little error. The pressure graphs between the model and the experimental result are shown in Figure 2. The performance and emission values obtained from numerical model were compared with experimental results. These comparisons are shown in Table 6 and Table 7.



**Figure 2.** Mean Pressure Curve between Model and Experimental Result **Table 5.** Comparison of Mean Pressures in-cylinder for Some Crank Angles

| Mean Pressure (bar)     |       |       |       |  |  |  |
|-------------------------|-------|-------|-------|--|--|--|
| 8 CA BTDC TDC 9 CA ATDC |       |       |       |  |  |  |
| Model                   | 53,46 | 92,38 | 111,9 |  |  |  |
| Experimental            | 53,2  | 84,8  | 108,4 |  |  |  |
| Rel. Error (%)          | 0.5   | 8.9   | 3.2   |  |  |  |

Table 6. Comparison of Performance Values for Model and Experimental Data

| Performance Values                    |       |       |        |  |  |  |  |
|---------------------------------------|-------|-------|--------|--|--|--|--|
| BSFC (g/kW.h) Power (kW) Torque (N.m) |       |       |        |  |  |  |  |
| <b>Model</b> 199,6                    |       | 56,32 | 358,44 |  |  |  |  |
| <b>Experimental</b> 201,16            |       | 56,87 | 360,31 |  |  |  |  |
| Rel. Error (%)                        | 0.775 | 0.97  | 0.52   |  |  |  |  |

Table 7. Comparison of NOx emissions for Some Crank Angles

| Nox Emission          |        |          |  |  |  |
|-----------------------|--------|----------|--|--|--|
| NOx (g/h) NOx (g/kW.h |        |          |  |  |  |
| Model                 | 540,83 | 9,603    |  |  |  |
| Experimental          | 537    | 9,442588 |  |  |  |
| Rel. Error (%)        | 0.71   | 1.7      |  |  |  |

The performance values obtained from the numerical model are shown in Table 8.

Table 8. Performance Values Obtained from Model

| Power (kW)                   | 56.32     |
|------------------------------|-----------|
| Torque (N.m)                 | 358.44    |
| Load (%)                     | 100       |
| Engine speed (rpm)           | 1500      |
| BSFC (g/kW.h)                | 199.6     |
| Injected fuel per cycle (mg) | 14.5      |
| Fuel Type                    | Diesel-D1 |

#### 3. RESULTS AND DISCUSSION

While the diesel engine was converted into a natural gas engine, the total energy contained in the fuel injected under diesel fuel conditions was considered as constant and the amount of natural gas fuel corresponding to this energy was calculated.

$$\dot{m}_{CH_4} = \frac{\dot{m}_{Diesel}\dot{Q}_{LHV,Diesel}}{Q_{LHV,CH_4}}$$
(6)

In addition, the presence of turbo under the conditions of the diesel engine was not considered in the natural gas operating condition and was considered as a natural suction engine. In these conditions, performance values were calculated by reference ignition advances at various compression ratios. 10, 11, 12, 13, 14, 15 and 16 values for compression ratios and  $20^{\circ}$ ,  $25^{\circ}$  and  $30^{\circ}$  CA BTDC (Before Top Dead Center) for reference ignition advances were selected. Performance values (Power, Torque and BSFC) were calculated for 21 cases (7x3). It is shown in the Table 9. When examining these conditions, the intake air humidity was considered to be zero. In addition, in these cases, whether or not a knock was checked by writing a Python code with the help of an empirical expression suggested by Douaud and Eyzat. The performance values are shown in Figure 3 and Figure 4.

When the conditions were examined, it was observed that the performance values at the 16 compression ratio were highest and there were no knock problems. Since the rise in ignition advance at increased compression ratios resulted in a decrease in performance values, at the 16 compression ratio, 13°, 15° and 17° CA BTDC ignition advances were also examined. The cases are shown in Table 10, Figure 5 and Figure 6 are drawn to the performance values of these cases.

The highest performance values for all compression ratios and ignition advances were achieved at 16 compression ratios and 15° CA BTDC of ignition timing. As these conditions were done with dry intake air, NOx emissions were very high. The optimum condition obtained according to the performance values was then optimized according to the emission values. The specific value of the amount of humidity present in the intake air was set at 1%, 3%, 5%, 7%, 10%, 15%, 20%, 25% and 30%. And then, both performance and emission values were obtained again at 15° CA BTDC ignition advance and 16 compression ratios. The analyzes are shown in Table 11. Performance and emission values of the cases are seen in the Figure 7, Figure 8, Figure 9 and Figure 10. In this study, 33 cases were investigated. The conditions examined were the results of the natural gas engine at only one operating point.

Excessive increase in the compression ratio was not preferable as the mixture ignited before the ignition timing and caused to increase friction in the in-cylinder particles. Increasing the compression ratio boosted thermal efficiency and improved combustion efficiency. It increased the performance values, but, boosted the NOx emissions, too. The cases of the different compression ratios in the study were made with the acceptance of mass of dry intake air. In the analyzes carried out by increasing the humidity content of the intake air, slight decreases were observed in performance values up to moist intake air with a ratio of about 25%.

Table 9. Case Matrix for Different Compression Ratio and Ignition Timing

|                 | CR      |         |         |         |         |         |         |
|-----------------|---------|---------|---------|---------|---------|---------|---------|
| Ignition Timing | 10      | 11      | 12      | 13      | 14      | 15      | 16      |
| 20° CA BTDC     | Case 1  | Case 2  | Case 3  | Case 4  | Case 5  | Case 6  | Case 7  |
| 25° CA BTDC     | Case 8  | Case 9  | Case 10 | Case 11 | Case 12 | Case 13 | Case 14 |
| 30° CA BTDC     | Case 15 | Case 16 | Case 17 | Case 18 | Case 19 | Case 20 | Case 21 |



Figure 3. Compression Ratio – Power Graph



## Figure 4. Compression Ratio – BSFC Graph

**Table 10.** Case Matrix for Different Ignition Timings

|    | Ignition Timing |             |             |             |  |
|----|-----------------|-------------|-------------|-------------|--|
| CR | 20° CA BTDC     | 17º CA BTDC | 15° CA BTDC | 13º CA BTDC |  |
| 16 | Case 7          | Case 22     | Case 23     | Case 24     |  |

Table 11. Case Matrix for Different Intake Air Humidity Ratios

|    | Specific Humidity Ratio |         |         |         |         |         |         |         |         |         |
|----|-------------------------|---------|---------|---------|---------|---------|---------|---------|---------|---------|
| CR | 0%                      | 1%      | 3%      | 5%      | 7%      | 10%     | 15%     | 20%     | 25%     | 30%     |
| 16 | Case 23                 | Case 25 | Case 26 | Case 27 | Case 28 | Case 29 | Case 30 | Case 31 | Case 32 | Case 33 |



Figure 5. Ignition Timing – Power Graph



**Figure 6.** Ignition Timing – BSFC Graph



Figure 7. Humidity Ratio – Power Graph



Figure 8. Humidity Ratio – BSFC Graph



Figure 9. Humidity Ratio – NOx Emission Graph



Figure 10. Humidity Ratio – SOOT Emission Graph

There was a sharp decrease in performance values after this amount of moisture. At 20% humidity, power decreased by 12.5% while NOx emissions decreased by 92.7%. Although this value is good in terms of emissions, it is a big loss in terms of performance values. The reason for this loss is that the natural gas air mixture ratio is poorer than the cytokiometric limit. The basic assumption in the analyzes was that the value of the diesel fuel bond energy was equal corresponding to the bond energy of the natural gas fuel. In Otto engines, the cytokiometric limits are preferred for the mixture. In the study, excess air ratio was measured as 1.36 and this value was preferred in all analyzes. By increasing the intake air humidity, the in-cylinder regional temperatures decreased and NOx emissions were reduced. PM emissions reached to the lowest value at 25% of specific humidity ratio. When we looked at the graph of PM emissions, it was seen that the regimen decreased up to a certain amount of moisture and then increased. The main reason for this situation was that when the humidity ratio increased to a certain value, combustion process in-cylinder moved to the expansion stroke and the combustion of unburned carbon particles occurred in the last stage of combustion. The prolongation of the late combustion phase reduced PM emissions. However, after 25% of the humidity ratio, sharp decreases in performance values caused PM emissions to increase. The deterioration of both the last burning phase and the phase where the main combustion took place affected the values of performance and PM emissions poorly.

#### 4. CONCLUSION

In the natural gas engine compared to the diesel engine, the in-cylinder regional temperatures were higher due to the fact that the pre-prepared air-fuel mixture entered into to cylinder was more richer and the combustion progressed more rapidly thanks to a spark plug. It is known that poor mixtures in-cylinder cause lower combustion temperatures. Furthermore, the combustion temperatures are lower due to the incomplete combustion in the richer mixtures. Excess air ratio of 2.13 in diesel engine was calculated as 1.36 in natural gas engine. It was quite natural to see a higher combustion temperature on the natural gas engine. Therefore, it was seen that there was a need for more specific humidity in reducing NOx emissions compared to diesel engine. The NOx emission resulting from the combustion of the diesel engine that had the intake air with humidity ratio of 5% was equivalent to the NOx emission resulting from the natural gas engine burning with the intake air with a moisture content of 20%. However, considering PM emissions, it was seen that more environmental results gived in compared to the diesel engine without high performance loses. In order to eliminate this disadvantage, the natural gas engine must be started by increasing the intake air humidity content and with a richer mixture of air fuel mixture. In addition, the excess air ratio and humidity rate must be considered together.

In conclusion;

- A validation study was conducted by comparing the numerical model of the diesel engine with the experimental results.
- The generated numerical model was converted to otto cycle engine at different compression ratios and the highest performance values were revealed.
- Increasing the compression ratio had a positive effect on thermal efficiency, but increased the tendency to knock.
- Ignition advances were shown to cause different effects on different compression rates. In terms of performance values, a low ignition advances were more favorable at higher compression ratios, while higher ignition advances were found to be preferable at low compression rates. However, a high reduction of the ignition advance at high compression rates resulted in loss of performance.
- The case had the highest performance was evaluated in terms of emissions by being examined different specific humidity ratios.
- The increase in specific humidity rates caused slight decreases in performance values, while quite desirable resulted in terms of emissions. Approximately 93% improvements in NOx emissions were observed.
- It is emphasized that the excess air ratio and the humidity rate of intake air must be considered together when comparing the diesel engine with the differences in the working principles of the natural gas engine.

#### REFERENCES

- Aydın M (2016). Çift yakıtlı sıkıştırmalı ateşlemeli bir motorda yanmanın üç boyutlu modellenmesi [Three dimensional modelling of a dual fuel compression ignition engine]. İstanbul Teknik Üniversitesi Fen Bilimleri Entitüsü, Yüksek Lisans Tezi, İstanbul.
- Deniz O (2008). İçten Yanmalı Motorlar [Internal Combustion Engine]. Yıldız Teknik Üniversitesi Makine Fakültesi Ders Notları, İstanbul.
- Ekin F (2017). Bir gemi dizel motorunda dizayn ve işletme parametrelerinin performans ve emisyon değerleri üzerindeki etkilerinin sayısal olarak incelenmesi [A numerical investigation on the effects of design and operation parameters on the performance and emissions of a marine diesel engine]. İstanbul Teknik Üniversitesi Fen Bilimleri Entitüsü, Yüksek Lisans Tezi, İstanbul.
- Saraçoğlu H (2018). Sıkıştırmalı ateşlemeli motorlarda tasarım ve işletme koşullarına yönelik parametrelerin performans ve emisyona etkilerinin istatistiksel yöntemlerle incelenmesi [Investigation of the effects of engine design and operating parameters on performance and emissions of compression ignition engines by statistical]. İstanbul Teknik Üniversitesi Fen Bilimleri Entitüsü, Doktora Tezi, İstanbul.
- Soruşbay C (2016). Egzoz Gazı Emisyonları [Exhaust Gases Emissions]. İstanbul Teknik Üniversitesi Makine Fakültesi Ders Notları, İstanbul.
- Şahin T (2019). Biyodizel ve bioetanol karışımının tek silindirli bir dizel motorda kullanımının motor performansına ve emisyonlara etkileri [Effects of using biodiesel and bioethanol mixture in single cylinder diesel engine to engine performance and emissions]. Necmettin Erbakan Üniversitesi Fen Bilimleri Entitüsü, Yüksek Lisans Tezi, Konya.
- Zeldovich, Y.B., Sadovnikov, P.Y. & Frank-Kamenetskii, D.A. (1947). Oxidation of Nitrogen in Combustion. Academey of Sciences of USSR, Institute of Chemical Physics, Moscow-Leningrad.



# Microwave Energy-Assisted Approach to Simultaneous Carbon Nanotube and Metal Oxide Nanowire Growth On Nanostructured Conducting Polymer Surface for Energy Storage Applications

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**Abstract:** Carbonized conducting polymer, e.g. polypyrole (PPy), nanostructures (nCCPs) decorated with simultaneously grown carbon nanotubes (CNTs) and metal oxide nanowires (MONWs) on their surface are prepared with a great potential as a building material for advanced engineering applications, such as energy storage. A well-established, in-situ polymerization/coating method and a simple and straightforward microwave (MW) energy-assisted carbonization approach, i.e. PopTube, are systematically combined to prepare this novel nanomaterial. Through this simple, facile, yet highly efficient, affordable and easily scalable combined method, it becomes possible to prepare such nCCPs with unique morphological (SEM/TEM), compositional (EDX), spectroscopic (XRD, Raman), and promising electrochemical (CV) features, all of which are strongly supported by both indicated various material characterization test results and the relevant literature data. Thus, it is believed that the as-obtained CNT and MONW decorated nCCPs (CNT-MONW/nCCP) via the above mentioned combined approach would soon become a material of preference for advanced energy storage applications.

Keywords: Carbonization, carbon nanotube, conducting polymer, metal oxide nanowire, microwave energy.

## **1. INTRODUCTION**

As a result of their uniquely blended morphological, thermal, spectroscopic and electrochemical features, nanostructured hybrid materials (nHMs), containing different types of active species with relevant functional groups, have attracted intense research interest from almost every research institute in various fields of science and industry worldwide.

Since its discovery at the beginning of 1990s, CNTs, in either single or multi-walled forms, have been observed to have the biggest share as one of the most common building blocks in affordable and high performance value added hybrid material production for sensory, energy storage, filtration, catalysis, magnetic resonance imaging (MRI) and electromagnetic interference (EMI) shielding applications due to their extraordinary thermal, electrical, mechanical and electrochemical properties. For this purpose, different approaches, mainly including arc discharge, pulsed laser vaporization, chemical vapor deposition, laser ablation and MW radiation-based synthesis have been utilized for productive, cost effective and pure CNT preparation at industrial scale. Among them, the MW energy-assisted approach comes one step forward before other conventional options due to its facile, rapid, tunable, affordable, clean and environmentally friendly working principle that is based on selective volumetric (from inside out) heating of the target material with certain polarity and MW absorption property. Besides this, accelerated process times with often relatively higher yield and as-obtained products with a higher degree of functionalization can be given as the other major advantages of MW energy-assisted approach against other methods, as well.

In order to both enhance and diversify the above mentioned inherent properties and to optimize the use of CNTs in nHMs by means of coating, filling or attaching via spontaneous self-assembly, a wide range of inorganic materials, e.g. metals or MOs made up of CdS, CdSe, Fe2O3, ZnO, MnO2, TiO2, RuO2 etc., have been commonly utilized. Among those, Fe2O3's high theoretical specific capacity, environmental friendly nature, low cost and high abundance can be attributed as the other reasons behind the attention paid for it for this study, as well. However, its low electrical conductivity, which significantly limits the application performance, and poor structural cycling stability, which is negatively affected by volumetric expansion in electrochemical energy storage applications remains as two main drawbacks that needs to be tackled. Thus, either in situ (nanoscale MO production in different morphologies, e.g. nano-sheet, nanowire, nano-rod or hollow nano-sphere) or ex situ (MOs simultaneous use with carbonaceous nanostructures, e.g. CNTs, graphene or reduced

graphene oxide (rGO)) solutions have been proposed. Here, the in situ solution depends on MOs preparation in advantageous nanoscale architecture since this provides a large amount of accessible material surface area in which the electrolyte molecules can easily and rapidly penetrate and interact with the active material during electrochemical applications. On the other hand, the ex situ solution relies on the employment of MOs together with nano-carbon materials, which enable high intrinsic electrical conductivity and excellent ductility that can effectively compensate both downsides of Fe2O3 during repetitive electrochemical processes such as charge-discharge.

As one of the most well-known and well-studied members of CP family owing to its advantageous properties such as facile and low-cost synthesis at ambient conditions with high yield, relatively high electrical conductivity, long-term environmental stability, and high electrochemical performance, PPy is chosen as the active host material to prepare the targeted nHM along with the previously mentioned CNTs and Fe2O3 NWs. To date, PPy has been both extensively synthesized in various nanostructured morphologies, e.g. from 0D nano-granules to 3D rambutan-like nano-spheres, and elaborately investigated in relevant studies and applications. Moreover, MW energy-assisted approach has been effectively utilized to prepare the carbonized derivatives of such nanostructured PPy in various studies, as well. Thus, the targeted nHM's preparation in 3D form via MW energy-assisted approach by the utilization of nCCP (PPy) as the host and the simultaneously grown CNTs and MONWs (Fe2O3) as the surface decoration would result in combination of both the advantageous nanostructured material design and unique electrochemical properties of such compounds in one structure. Also, a promising solution is envisioned to be provided for CNTs' common tendency to easily form agglomerates (via the intrinsic Van der Waals interactions towards each other) and exhibit poor dispersion in most of both the organic and aqueous solvents, as a result of the as-proposed approach.

With that, in current study, a well-established, in-situ polymerization/coating method and a simple and straightforward MW energy-assisted carbonization approach, i.e. PopTube, were systematically combined to prepare this novel nHM. The first method provides a uniform nanostructured PPy coating, which is essential to absorb MW energy and transfer the generated heat to the precursors, for CNT and MONW surface decorations growth. Additionally, the method has many advantages of being simple, facile yet highly efficient, affordable, easily scalable and applicable at ambient conditions. On the other hand, the MW energy-assisted approach can be considered more useful than the previously mentioned conventional approaches by the following aspects of; (i) being in need of simple experimental conditions and instrumental setup, (ii) being universal in terms of working principle that is template-free and capable of growing various nanostructured materials, (iii) being versatile by allowing to simultaneously work with different materials, (iv) being controllable in terms of the targeted functional nanostructured materials' growth uniformity and their properties, and most importantly (v) being ultrafast to provide 3D nHMs.

## 2. MATERIALS AND METHODS

#### Materials

Following chemicals and reagents, including ammonium peroxydisulfate (APS,  $(NH_4)_2S_2O_8$ , 98% min.), ferrocene (99%), acetone, and pyrrole (98%), were all purchased from Alfa Aesar and used for the synthesis reactions. Iron pentacarbonyl (Fe(CO)<sub>5</sub>, 99.5% and 99%-Fe) was purchased from Strem Chemicals. All chemicals and reagents were used as received without any further purification unless otherwise specified.

#### In-situ synthesis and coating of PPy on fine ferrocene particles (PPy/ferrocene)

Fine ferrocene particles were obtained by screening ferrocene through a metal sieve. Next, as shown in Figure 1, these particles were used as substrates to coat conducting PPy on their surfaces via an in-situ oxidative polymerization reaction at ambient conditions. Here, 10 g of these particles was dispersed with in a beaker containing 90 mL 1 M aq. HCl under vigorous magnetic stirring. After 10 min dispersion time, 1 mL pyrrole monomer was added into this medium. The solution was kept stirred for another 10 min to allow the homogenous distribution of monomers. The reaction medium was then moved into an ice bath; in which it was cooled down to 5 °C. The oxidant solution was prepared by initially dissolving 1.15 g of APS in 10 mL 1 M aq. HCl, and then cooling the solution down to 5 °C, and finally adding it dropwise into the previous mixture under magnetic stirring. Once the oxidant had been completely added, the reaction was left to proceed for 4 h. The resulting black suspension was firstly filtered and washed with excess 1 M aq. HCl and acetone to remove any impurities, and eventually, it was dried overnight under vacuum at ambient conditions to yield a fine black PPy/ferrocene powder.



Figure 1. Schematic illustration of the combined nHM synthesis approach

### Preparation of CNT and MONW decorated cPPy (CNT & MONW@cPPy) via MW energy-assisted approach

In order to obtain the targeted nHM samples; 50 mg of PPy/ferrocene powder was placed in a glass vial and soaked with 0.1 mL of Fe(CO)<sub>5</sub> until its surface was completely covered. After the partial evaporation of the liquid phase, the sample was placed into a standard kitchen MW oven (Panasonic Inverter, NN-SN936B) chamber for the quick heating process. During 30 s of MW irradiation, vigorous sparking, glowing and even flames were observed on the sample surface. Upon the rapid heat gain through MW-PPy interactions; (i) complete PPy carbonization was occurred, while (ii) ferrocene and Fe(CO)<sub>5</sub> got rapidly decomposed and simultaneously formed a gaseous phase, which lead to the (iii) formation/decoration of CNTs and MONWs, respectively. Finally, after the vial was taken out and cooled down to ambient temperature, nHM samples were gently collected from the vial bottom with a steel spatula and stored for further characterizations.

## Characterization of the CNT & MONW@cPPy samples

Different characterization techniques were used in order to identify the as-obtained nHM samples' properties. Their morphological features were analyzed by JEOL JSM-7000F scanning electron microscope (SEM) equipped with an energy dispersive X-Ray (EDX) detector. The in-depth morphological analysis of nHM samples was performed on a JEOL 2100F transmission electron microscope (TEM) that was operated at 200 kV. Additionally, crystalline nature of the as-formed MONWs was determined by X-ray diffractometry (XRD) analysis that was performed on a Rigaku powder XRD instrument, while the as-grown CNTs' spectroscopic analysis was conducted on a Raman instrument. Also, electrochemical performance of the as-obtained nHM samples was evaluated by cycling voltammetry (CV). The tests were performed on a CHI-601D workstation equipped with a conventional three-electrode cell system. The as-prepared WE (polished graphite with ~0.5 mg nHM application on its surface via isopropyl alcohol-based colloidal carbon paste) was used with a Pt wire as the counter (CE) and Ag/AgCl as the reference electrode (RE), respectively. 6 M aq. KOH solution was used as the electrolyte to perform CV tests in sweeping voltage mode at 5 mV/s scan rate and between -0.2 - +0.5 V potential window.

## 3. RESULTS AND DISCUSSION

The as-obtained nHM samples' SEM images are shown in Figures 2A and 2B.



Figure 2. SEM images of the as-grown; (A) CNTs and (B) Fe<sub>2</sub>O<sub>3</sub> NWs on nHM sample

Based on the results, a few microns long CNTs with ~200 nm in hollow center diameter were homogenously grown on nHM surface. Similarly, noodle-like Fe2O3 NWs with more than 10  $\mu$ m in length and ~0.6  $\mu$ m in diameter were densely grown on nHM surface. These results are in good agreement with the ones from the previous literature. Expectedly, the as-obtained nHMs can be effectively used for electrochemical energy storage applications with respect to their ultra-high carbonized active surface area that is homogenously decorated with CNTs and MONWs.

The in-depth morphological and elemental features of nHM samples' surface decorations were further characterized by both TEM microscopy and EDX analysis. Collected results from these analyses are shown in Figures 3 and 4.



Figure 3. TEM image of the; (A) catalyst Fe NPs encapsulated within the as-grown multi-walled CNTs, (B) the EDX diffractogram of the indicated Fe NP in Figure 3A, (C) HR-TEM image of the marked area in Figure 3A, and (D) the EDX diffractogram of the indicated Fe NP in Figure 3C.

The TEM image in Figure 3A provides more detailed information about the as-grown CNTs by indicating their hollow center that encapsulates catalyst Fe NPs. This is a typical structure that is obtained upon a tip-growth process, which will be explained later on. As it can be seen from the high resolution TEM image in Figure 3C, a single ~25 nm × 5 nm Fe catalyst NP is encapsulated within the as-grown multi-walled CNTs' walls that was made up of thin graphene layers. The EDX diffractograms of both the as-grown CNTs and the encapsulated catalyst iron NPs are shown in Figures 3B and 3D. They provide certain evidences about the elemental composition of both nanostructures. It should be noted that two sharp peaks with Cu indicators in these diffractograms, can be attributed to the copper grid sample holder that is used for TEM imaging process. Moreover, the as-grown MONWs were observed to be composed of tiny core-shell NP structures, as shown in Figure 4. These NPs are made up of Fe2O3 cores that are encapsulated within graphitized carbon shells. This phenomenon can be simply explained with the disintegration of microns long, noodle-like MONWs into its constituents, most probably by the ultrasonication-based sample preparation process applied prior to TEM imaging. This result is also agrees well with the ones from the relevant previous studies, in which the NWs obtained from Fe(CO)5 do not have continuous solid structure. Instead, they exhibit a pearl necklace-like morphology that is composed of adjacent NPs encapsulated within graphitized carbon layers. Based on the measurements taken from this figure, it was observed that these NPs' diameter size distribution ranges between 40-160 nm.



Figure 4. TEM image of the core-shell NP constituents of the as-grown MONWs

Further morphological and elemental analysis results of the as-grown MONWs are shown in Figure 5. It can be observed from the summarized results of the indicated spectra that such NWs are made up of Fe (67.5 and 58 wt%, respectively) and O (32.5 and 42 wt%, respectively) elements.



Figure 5. The as-grown MONWs' EDX analysis results taken from the relevant SEM image

In order to both confirm the as-grown CNTs and MONWs presence and identify these nanostructures' crystalline and spectroscopic nature, Raman and XRD analyses were performed, respectively and their results are shown in Figure 6. Here, the as-grown CNTs are observed to be in radial breathing mode (200-300 cm-1) with characteristic D (1317 cm-1) and G (1587 cm-1) bands located at wave numbers that were previously discussed in relevant studies (Figure 6A). It can be also observed in Figure 6B that characteristic diffraction peaks appeared on the as-grown MONWs' spectrum coincides with the ones from standard Fe2O3 hematite (JCPDS: 33-0664).



Figure 6. (A) Raman spectrum, (B) XRD diffractogram of the as-grown CNTs and MONWs, respectively

Last but not the least; the as-produced nHMs' electrochemical properties were analyzed via CV in a comparative manner and the relevant results are shown in Figure 7.



**Figure 7.** CV voltammograms of cPPy NPs, CNT@cPPy NPs and CNT+MONW@cPPy NPs in 6 M aq. KOH electrolyte at 5 mV/s scan rate.

Based on the analyzed samples' quasi-rectangular voltammogram shapes, the electric double-layer carbon (EDLC)-based electrochemical characteristic is clearly indicated. Here, since majority of the samples are carbonized, their electrochemical capacitance behavior depends on forming a double layer at their interface with the electrolyte. In terms of shape and symmetry, all different sample-based electrodes' voltammograms exhibit similarity, but when it comes to their sizes, i.e. current density, approximately two and four folds of difference can be observed in CNT@cPPy and CNT+MONW@cPPy samples' voltammograms compared to cPPy sample's voltammogram, respectively. This is a clear proof of a similar electrochemical process with much higher capacitive performance difference between these three electrodes. The main reason behind this phenomenon is the presence of MONW compound and its significant pseudo-capacitive property contribution that is based on storing charges on both its interface with the electrolyte, like EDLCs, and within their structures via a faradic reaction with electrosorption, redox reaction and intercalation steps.

#### 4. CONCLUSION

The particular aims of this study can be listed as follows; (i) to conduct well-organized experimental procedures, preferably with previous literature data support, (ii) to develop novel nanostructured materials with a promising potential for advanced engineering applications, (iii) to characterize and verify the structure and chemistry of the as-obtained materials to understand their properties in details, and (iv) to reveal the effective mechanisms involved in the synthesis and production of the as-obtained nHMs as a route to optimize their application performances. Thus, along with these aims, preparation of the simultaneously grown CNT and MONW decorated nCCPs was accomplished throughout this study. Two well-established approaches, namely in-situ polymerization/coating of CPs on a precursor material and MW energy-assisted PopTube, were systematically combined in order to achieve nHMs with promising features, which are strongly supported by both various material characterization test results and the previous literature data. To conclude, it is envisaged that the as-obtained novel nHMs will soon become a material of preference for various advanced applications such as energy harvesting/storage, magnetic resonance imaging (MRI), and water treatment.

#### REFERENCES

- De Adhikari A, Oraon R, Tiwari S K, Saren P, Maity C K, Lee J H, Kim N H & Nayak G C (2018). Zn-doped SnO2 Nano-urchinenriched 3D Carbonaceous Framework for Supercapacitor Application. New Journal of Chemistry, 42: 955-963.
- Hou Y, Cheng Y, Hobson T & Liu J (2010). Design and Synthesis of Hierarchical MnO2 Nanospheres/Carbon Nanotubes/Conducting Polymer Ternary Composite for High Performance Electrochemical Electrodes. Nano Letters, 10: 2727-2733.
- Lakshmi A, Gracelin D L, Vigneshwari M, Karpagavinayagam P, Veeraputhiran V & Vedhi C (2015). Microwave Synthesis and Characterization of Multiwalled Carbon Nanotubes (MWCNT) and Metal Oxide Doped MWCNT. Journal of Nanoscience and Technology, 1: 19-22.
- Lokhande V C, Lokhande A C, Lokhande C D, Kim J H & Ji T (2016). Supercapacitive Composite Metal Oxide Electrodes Formed with Carbon, Metal Oxides and Conducting Polymers. Journal of Alloys and Compounds, 682: 381-403.
- Lota G, Fic K & Frackowiak E (2011). Carbon Nanotubes and Their Composites in Electrochemical Applications. Energy and Environmental Science, 4: 1592-1605.
- Luo Y, Wang K, Luo S, Zhao F, Wu H, Jiang K, Li Q, Fan S & Wang J (2018). Three-Dimensional Carbon Nanotube/Transition-Metal Oxide Sponges as Composite Electrodes with Enhanced Electrochemical Performance. ACS Applied Nano Materials, 1: 2997-3005.
- Mazloumi M, Shadmehr S, Rangom Y, Nazar L F & Tang X (2013). Fabrication of Three-Dimensional Carbon Nanotube and Metal Oxide Hybrid Mesoporous Architectures. ACS Nano, 7: 4281-4288.
- Motshekga S C, Pillai S K, Ray S S, Jalama K & Krause R W M (2012). Recent Trends in the Microwave-Assisted Synthesis of Metal Oxide Nanoparticles Supported on Carbon Nanotubes and Their Applications. Journal of Nanomaterials, 2012: 1-15.
- Poyraz S, Cook J, Liu Z, Zhang L, Nautiyal A, Hohmann B, Klamt S & Zhang X Y (2018). Microwave Energy-based Manufacturing of Hollow Carbon Nanospheres Decorated with Carbon Nanotubes or Metal Oxide Nanowires. Journal of Materials Science, 53: 12178-12189.
- Wang Y W, Du Y, Ping X, Qiang R & Han X (2017). Recent Advances in Conjugated Polymer Based Microwave Absorbing Materials. Polymers, 9: 29-56.
- Yu G, Xie X, Pan L, Bao Z & Cui Y (2012). Hybrid Nanostructured Materials for High-Performance Electrochemical Capacitors. Nano Energy, 2: 213-234.
- Zhao Y, Li J, Wu C & Guan L (2011). A General Strategy for Synthesis of Metal Oxide Nanoparticles Attached on Carbon Nanomaterials. Nanoscale Research Letters, 6: 71-76.
- Zheng W, Zhang P, Chen J, Tian W B, Zhang Y M & Sun Z M (2019). Microwave-assisted Synthesis of Three-dimensional Mxene Derived Metal Oxide/Carbon Nanotube/Iron Hybrids for Enhanced Lithium-ions Storage. Journal of Electroanalytical Chemistry, 835: 205-211.



## A Numerical Study About the Effects of Spray Angle and Injector Nozzle Diameter on the Performance and Emission Values in A Diesel Engine

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Abstract: Today, internal combustion engines are used in many areas such as increasing industrialization and transportation. Emissions gotten out from the exhaust gases of these engines to the atmosphere have extremely damaging effects on living things. These damages caused by air pollution to living things were not taken into consideration until recently. Increasing global warming, imbalance in climates and serious disturbances caused by air pollution on living creatures have received many international and national mandatory emission standards. Scientists have done many studies on internal combustion engines in particular. Gasoline and diesel engines have a great importance in terms of performance values. However, the emissions of these engines to the environment are also very high. In the literature, there are many studies, both numerical and experimental, to reduce the emission values of these engines without experiencing performance losses. The ratio between the surface area and the volume of the fuel droplets sprayed on diesel engines plays a major role in fuel combustion. Furthermore, these fuel particles must be combined with the air in the cylinder at a suitable spray angle. Therefore, the spray angle and the nozzle nozzle diameter are extremely important in order to form a suitable air fuel mixture. In this study, a 4-cylinder, turbo-charged, four-stroke diesel engine located in the Istanbul Technical University Motors Laboratory was modeled numerically with the help of the AVL Fire CFD program. The modeled diesel engine was compared with experimental results and validation study was performed. Then, the performance and emission values of the engine at different spray angles and injector nozzle hole diameters were examined. The original spray angle of the engine was  $160^{\circ}$  degrees and the injector hole diameter was 0.16 mm. In the numerical study, 120°, 140° and 160° degrees spray angles and 0.15, 0.16 and 0.17 mm injector nozzle hole diameters were examined. These parameters were investigated in binary combination and performance and emission values were compared with experimental values. According to the results obtained, the rise in spray angle had a positive effect on the performance values and increased the NOx emissions. Ideal results were observed for SOOT emissions. Decreasing the diameter of the injector holes resulted in reductions in fuel particle diameters. This circumstance increased the performance values but increased NOx emission values due to increased in-cylinder regional temperatures, too. SOOT emission values decreased due to increasing in-cylinder temperatures. When both parameters were considered together, the optimum condition for both performance and emission values according to the test conditions was obtained for 160° degree spray angle and 0.17 mm injector nozzle hole diameter. The power loss was only 0.07%, while the NOx emission decreased by about 9%. There was a 24% increase in SOOT emissions. However, despite this increase of SOOT emissions, it can be said that this situation is more ideal due to the positive results in the NOx emissions and the narrowing NOx emission limits despite the slight decrease in the performance values.

Keywords: Diesel Engine, Spray Angle, Injector Nozzle Diameter, Emission, Numerical Study

#### **1. INTRODUCTION**

Air pollution is the main cause of the natural disasters and the health threatening diseases. One of the main reasons causing air pollution is undoubtedly internal combustion engines. For this reason, many studies have been done in literature about internal combustion engines. The researches were experimentally and numerically diversified to keep both performance values and emission values of the engines at optimum levels. The high compression efficiency of diesel engines caused high thermal efficiency. Both being more economical and having high torque values have made diesel engines indispensable especially in the trade sector. Therefore, studies on diesel engines are very high.

Performance and emission values of diesel engine at different swirl ratios and spray angles were investigated. (Ekin et al., 2018). In the study, the effects of spray angle and swirl ratio on engine performance and emission values are

investigated numerically. The AVL FIRE CFD software is used to obtain numerical results. The numerical model generated is compared with the experimental results and validated. The original engine's injector spray angle is 148 degrees and the swirl ratio is 2.37. Numerical results were obtained by examining the effect of different spray angles and swirl ratios in this study. The values of 140°, 148° and 153° as the spray angle and the values of 2, 2.37 and 2.5 of the swirl ratio were investigated. 9 different cases were created by obtaining the binary combinations of all these values. The case in which the experimental conditions are modeled is called "Reference Case". By comparing the other 8 cases both the Reference Case and among themselves, comments are made on the most optimum conditions in terms of both emission values and performance values. Whereas highest values of NOx, SOOT and HC emissions are determined at 140° of spray angle and 2 of swirl ratio, lowest values are determined 153° and 2.5, respectively. Lowest specific fuel consumption per unit power in terms of performance values is obtained at 153° of spray angle, 2 of swirl ratio. Power and torque values are found to be very close to each other at all spray angle and 2.5 swirl ratio value give better results compared to the reference motor.

The effect of different operating and design parameters of a ship diesel engine on the performance and emission values was investigated numerically. (Ekin, 2017). In the study, the optimum working points of the engine were obtained by examining the conditions which provide more optimum results than the existing parameters.

In a diesel engine, the effect of the swirl ratio on the emission and performance values of the combustion process was investigated numerically. (Wei et al., 2013). In the study, a certain combustion chamber geometry was modeled using an CFD program (AVL FIRE) and turbulence kinetic energy, in-cylinder temperature and in-cylinder air-fuel regional mixtures were monitored in different swirl ratios (0.2, 0.4, 0.8, 1.2, 1.7, 2.2, 2.7 and 3.2). In the study, the best air-fuel mixture was observed at 0.2 swirl ratio. The lowest NOx emissions were observed at 0.8 swirl ratio, while the highest NOx emissions were obtained at 2.7 swirl ratio.

The effect of spray angle on the diesel engine has a great effect on the emission and performance values. Therefore, it is possible to find many studies about spray angle in the literature. In the study, numerical results were obtained about the emission values by monitoring the in-cylinder temperature and air-fuel regional mix distributions. (Wei et al., 2014). In the study, for the specific values of the spray angle (146°, 150° and 154°), analyzes of the changes of NOx and SOOT emissions were made. As a result of the analyzes, it was seen that the best air-fuel mixture ratio was at 146° spray angle. The lowest value of NOx emissions was observed at 154°, while the lowest value of SOOT emissions was observed at 146°.

In a diesel engine with single cylinder, numerical results were obtained on performance and emission values by the examining the change of both the combustion chamber and the swirl ratio. (Gafoor and Gupta, 2015). 80%, 75%, 70%, 64%, 60%, 55%, and 50% values of the diameter ratio and 0.5, 1.5, 2.5, 3.5 and 4.5 values of the swirl ratio were changed at regular intervals and a total of 35 conditions were examined. As a result of the study, it was found that there were 2 most suitable conditions in terms of emissions. These cases were 70% of diameter ratio and 0.5 of swirl ratio and 55% of diameter ratio and 2.5 of swirl ratio. The numerical results were shown to be at optimum level in terms of performance values and emission values.

In this study, a 4-cylinder, turbo-charged, four-stroke diesel engine located in the Istanbul Technical University Motors Laboratory was modeled numerically with the help of the AVL Fire CFD program. General characteristics of the test engine; 4-cylinder, 4-stroke, piston diameter 105 mm, piston stroke 115 mm, turbo-charged, with a compression ratio of 16.2:1, and a direct injection compression ignition engine with a motor volume of 3.908 liters. The modeled diesel engine was compared with experimental results and validation study was performed. Then, the performance and emission values of the engine at different spray angles and injector nozzle hole diameters were examined. The original spray angle of the engine was 160° degrees and the injector hole diameter was 0.16 mm. In the numerical study, 120°, 140° and 160° degrees spray angles and 0.15, 0.16 and 0.17 mm injector nozzle hole diameters were examined. These parameters were investigated in binary combination and performance and emission values were compared with experimental values. Firstly, a numerical model was created in a CFD program (AVL Fire) for the engine. The numerical results and experimental results were compared and validated

### 2. MATERIAL AND METHODS

### **Engine Specifications and Models**

The study was carried out in two stages. In the first stage, a turbocharged 4-cylinder intercooler diesel engine located in the Engine Laboratory of Istanbul Technical University, which had experimental results, was modeled. And the modeled motor was compared with experimental results and validation study was performed. The characteristics of the modeled diesel engine are shown in Table 1.

After creating the geometric model of the motor, the simulation parameters must be entered according to the values in the test conditions. Initial conditions are shown in Table 2. After the solutions were obtained in different cell numbers, the solution was provided independently of the cell number. In addition, the accuracy of the model was compared with the experimental results and a validation study was conducted. Different cell sizes and corresponding cell numbers are shown in Table 3.

The higher the number of cells increases the time of analysis unnecessarily. The cell size and the corresponding cell number at which the results did not change were obtained as 0.00085 m and about 50000, respectively. The generated combustion chamber geometry is shown in Figure 1.

It was seen that the results in different cell numbers did not change after about 50000 cell numbers and all analyzes were analyzed in this cell number so that the analyzes did not take much time.

 Table 1. Engine Specifications

| Cylinder number                          | 4      |
|--|--------|
| Bore (mm)                                | 104    |
| Stroke (mm)                              | 115    |
| Connecting rod length (mm)               | 182    |
| Nozzle hole number                       | 4      |
| Spray angle                              | 160    |
| Spray cone angle                         | 6      |
| Nozzle hole diameter (mm)                | 0.16   |
| <b>Compression ratio</b>                 | 16.2:1 |
| Total cylinder volume (cm <sup>3</sup> ) | 3908   |

#### Table 2. Initial Conditions

| Intake pressure (bar)                 | 1.546    |
|---------------------------------------|----------|
| Intake temperature (K)                | 303      |
| Swirl ratio                           | 2        |
| IVC (CA BTDC)                         | 154      |
| EVO (CA ATDC)                         | 130      |
| Turbulence kinetic energy $(m^2/s^2)$ | 10       |
| Turbulence model                      | k-zeta-f |
| Injection timing (CA BTDC)            | 12       |
| Fuel temperature (K)                  | 330.15   |

#### Table 3. Cell Size for Analyzes

| Mean Cell Size | <b>Total Cell Number</b> |  |
|----------------|--------------------------|--|
| 0,00250 m      | 15000                    |  |
| 0,00150 m      | 25000                    |  |
| 0,00100 m      | 35000                    |  |
| 0,00085 m      | 50000                    |  |
| 0,00075 m      | 65000                    |  |
| 0,0005 m       | 100000                   |  |





#### **Mathematical Model and Boundary Conditions**

After the geometric model is created, it is necessary to make the necessary mathematical assumptions for the simulation. The k-zeta-f four equality model proposed by Hanjelic was used as a turbulence model. The ECFM-3Z model, which a combustion model that is suitable for diffusion combustion and pre-mixed combustion, which is commonly used in the literature, was used as combustion model. While Zeldovich model was used for NOx emissions, Kinetic model was used for PM emissions. Models used for analysis and boundary conditions are shown in the Table 4.

Table 4. Boundary Conditions and Models

| Cylinder head temperature (K) | 550.15            |
|-------------------------------|-------------------|
| Piston wall temperature (K)   | 575.15            |
| Liner temperature (K)         | 475.15            |
| Turbulence model              | k-zeta f          |
| Evaporation model             | Dukowicz          |
| Spray break-up model          | KHRT              |
| Combustion model              | ECFM-3Z           |
| Nox model                     | Extended Zeldovic |
| SOOT model                    | Kinetic           |

The AVL Fire program analyzes conservation equations in three dimensions by finite volume method by accepting compressible flow. Conservation equations are conservation of mass, conservation of momentum (Navier-Stokes) and conservation of energy. These equations are shown below.

Mass continuity equation:

$$\frac{\partial \vec{\rho}}{\partial t} + \frac{\partial}{\partial x_j} \left( \vec{\rho} \vec{U}_j \right) = 0 \tag{1}$$

Momentum equation:

$$\vec{\rho} \frac{D\overline{U_{i}}}{Dt} = \vec{\rho} \frac{\overline{U_{i}}}{\partial t} + \vec{\rho} \overline{U_{j}} \frac{\partial \overline{U_{i}}}{\partial x_{j}} = \vec{\rho} g_{i} - \frac{\partial \vec{p}}{\partial x_{i}} + \frac{\partial}{\partial x_{j}} \left[ \mu \left( \frac{\partial \overline{U_{i}}}{\partial x_{j}} + \frac{\partial \overline{U_{j}}}{\partial x_{i}} - \frac{2}{3} \frac{\partial \overline{U_{k}}}{\partial x_{k}} \delta_{ij} \right) \right]$$
(2)
Energy equation:

$$\vec{\rho}\frac{\vec{DH}}{Dt} = \vec{\rho}\left(\frac{\vec{DH}}{\partial t} + \vec{U}_{j}\frac{\partial\vec{H}}{\partial x_{j}}\right) = \vec{\rho}\dot{q}_{g} + \frac{\partial\vec{p}}{\partial t} + \frac{\partial}{\partial x_{i}}\left(\vec{\tau}_{ij}\vec{U}_{j}\right) + \frac{\partial}{\partial x_{j}}\left(\lambda\frac{\partial\vec{T}}{\partial x_{j}}\right)$$
(3)

Concentration equation:

$$\vec{\rho} \frac{D\vec{C}}{Dt} = \vec{\rho} \frac{\partial\vec{C}}{\partial t} + \vec{\rho} \overline{U_j} \frac{\partial\vec{C}}{\partial x_j} = \vec{\rho} \vec{r} + \frac{\partial}{\partial x_j} \left( D \frac{\partial\vec{C}}{\partial x_j} \right)$$
(4)

#### **Model Validation**

The experimental results were compared with the numerical result. While validating the experimental study with numerical model, the differences between the pressure values and performance and emission values were compared in some reference crank angles (12°CA BTDC, TDC and 9° CA ATDC). The pressures between the model and the experimental result at different crank angles are shown in Table 5. The results were confirmed with a little error. The pressure graphs between the model and the experimental result are shown in Figure 2. The performance and emission values obtained from numerical model were compared with experimental results. These comparisons are shown in Table 6 and Table 7.



**Figure 2.** Mean Pressure Curve between Model and Experimental Result **Table 5.** Comparison of Mean Pressures in-cylinder for Some Crank Angles

| Mean Pressure (bar)     |       |       |       |  |  |  |  |
|-------------------------|-------|-------|-------|--|--|--|--|
| 8 CA BTDC TDC 9 CA ATDC |       |       |       |  |  |  |  |
| Model                   | 53,46 | 92,38 | 111,9 |  |  |  |  |
| Experimental            | 53,2  | 84,8  | 108,4 |  |  |  |  |
| Rel. Error (%)          | 0.5   | 8.9   | 3.2   |  |  |  |  |

Table 6. Comparison of Performance Values for Model and Experimental Data

| Performance Values                   |       |       |        |  |  |  |  |
|--------------------------------------|-------|-------|--------|--|--|--|--|
| BSFC (g/kW.h) Power (kW) Torque (N.n |       |       |        |  |  |  |  |
| Model                                | 199,6 | 56,32 | 358,44 |  |  |  |  |
| <b>Experimental</b> 201,16           |       | 56,87 | 360,31 |  |  |  |  |
| Rel. Error (%)                       | 0.775 | 0.97  | 0.52   |  |  |  |  |

Table 7. Comparison of NOx emissions for Some Crank Angles

| Nox Emission          |        |          |  |  |  |  |
|-----------------------|--------|----------|--|--|--|--|
| NOx (g/h) NOx (g/kW.h |        |          |  |  |  |  |
| Model                 | 540,83 | 9,603    |  |  |  |  |
| Experimental          | 537    | 9,442588 |  |  |  |  |
| Rel. Error (%)        | 0.71   | 1.7      |  |  |  |  |

The performance values obtained from the numerical model are shown in Table 8.

**Table 8.** Performance Values Obtained from Model

| Power (kW)                   | 56.32     |
|------------------------------|-----------|
| Torque (N.m)                 | 358.44    |
| Load (%)                     | 100       |
| Engine speed (rpm)           | 1500      |
| BSFC (g/kW.h)                | 199.6     |
| Injected fuel per cycle (mg) | 14.5      |
| Fuel Type                    | Diesel-D1 |

# 3. RESULTS AND DISCUSSION

In the study, the effects of different spray angles  $(120^{\circ}, 140^{\circ} \text{ and } 160^{\circ})$  and injector hole diameters (0.15, 0.16 and 0.17 mm) on the performance and emission values were examined numerically. The conditions in the study are shown in Table 9. After the results of all conditions were obtained in the form of a binary combination, it was tried to determine the more suitable condition by comparing with the numerical results in the current motor conditions. The results were evaluated by showing on the graphs. In the graphs, while injector nozzle hole diameters were fixed, change in performance and emission values were examined of spray angles. In addition, while the fuel spray angles were fixed, the differences in performance and emission values were examined as a result of the change of the nozzle hole diameters.

Table 9. Case Matrix for Different Compression Ratio and Ignition Timing

| Cases |      |      |      |  |  |
|-------|------|------|------|--|--|
|       | 0,15 | 0,16 | 0,17 |  |  |
| 120   | A1R1 | A1R2 | A1R3 |  |  |
| 140   | A2R1 | A2R2 | A2R3 |  |  |
| 160   | A3R1 | A3R2 | A3R3 |  |  |

When the power, torque and BSFC values were examined, it was observed that the performance values increased by increasing the spray angle for fixed nozzle hole diameter. Accordingly, the break specific fuel consumption value decreased. This situation is seen in Figure 3, Figure 4, Figure 5, Figure 6, Figure 7 and Figure 8.



Figure 3. Spray Angle – Power Graph



Figure 4. Injector Nozzle Diameter – Power Graph



Figure 5. Spray Angle – Torque Graph

Higher spray angles are effective in the homogenous combination of air and fuel. Lower injector nozzle hole diameters have allowed the fuel to break down into smaller droplets and increased combustion efficiency. In addition, it caused the ignition delay to decrease as it caused the fuel to evaporate in a shorter time.



Figure 6. Injector Nozzle Diameter - Torque Graph



Figure 7. Spray Angle – BSFC Graph



Figure 8. Injector Nozzle Diameter – BSFC Graph

NOx emissions increased due to the increase in regional temperatures with increasing spray angle. At the constant spray angle, the increase in injector nozzle hole diameters resulted in a decrease in NOx emissions. This is shown in Figure 9 and Figure 10. NOx emissions are caused by the combustion of oxygen with nitrogen in the air after 1800 K temperature.



Figure 9. Spray Angle – NOx Emission Graph



Figure 10. Injector Nozzle Diameter - NOx Emission Graph



Figure 11. Spray Angle – SOOT Emission Graph

As the combustion process in-cylinder takes place in a very short time, the nitrogen oxide cannot be broken down again into nitrogen and oxygen. Increasing the spray angle and decreasing the injector hole diameter led to a more homogeneous air fuel mixture so that the resulting in increased regional temperatures.



Figure 12. Injector Nozzle Diameter - SOOT Emission Graph

There is an opposite relationship between PM emissions and NOx emissions. One is increasing while the other is a decreasing characteristic. SOOT emissions are known as unburned carbon particles. SOOT emissions occur especially during diffusion combustion phase. It is an emission type that should be considered in diesel engines and direct injection combustion strategies. Increased spray angle or reduction of injector nozzle hole diameters reduced SOOT emissions.

This is shown in Figure 11 and Figure 12. Reduction of combustion quality or increased fuel ratio leads to an increase in SOOT emissions in the in-cylinder combustion process.

#### **4. CONCLUSION**

The spray angle and injector hole diameters are known to be extremely important in the homogeneous formation of the air and fuel mixture. In this study, the effect of these parameters was investigated. The test engine has a spray angle of  $160^{\circ}$  and an injector nozzle hole diameter of 0.16 mm (A3R2). 9 different conditions were examined at different spray angles and injector nozzle hole diameters with the aim of revealing more suitable emission and performance values. According to the numerical results, it was found that the condition of the spray angle of  $160^{\circ}$  and the diameter of the injector nozzle hole of 0.17 mm (A3R3) were more preferable in terms of both performance values and emission values when compared with the conditions of the current diesel engine. It was seen that it was possible to improve the NOx emissions by 9% without loss of performance values.

In conclusion;

- A validation study was conducted by comparing the numerical model of the diesel engine with the experimental results.
- Increasing the spray angle and reducing the nozzle hole diameter rased the performance values. It led to an increase in NOx emissions while decreasing SOOT emissions.
- Changes in the diameter of the injector nozzle were not very effective at high spray angles.
- There was a 9% improvement in NOx emissions and 0.07% decrease in performance values when compared to the current situation of the engine.

#### REFERENCES

AVL. (2013) AVL FIRE® VERSION 2013 Spray Module Edition, Februaray.

AVL. (2013) AVL FIRE® VERSION 2013 Combustion Module Edition, February.

AVL. (2013) AVL FIRE® VERSION 2013 Emission Module Edition, February.

- Aydın M (2016). Çift yakıtlı sıkıştırmalı ateşlemeli bir motorda yanmanın üç boyutlu modellenmesi [Three dimensional modelling of a dual fuel compression ignition engine]. İstanbul Teknik Üniversitesi Fen Bilimleri Entitüsü, Yüksek Lisans Tezi, İstanbul.
- Deniz O (2008). İçten Yanmalı Motorlar [Internal Combustion Engine]. Yıldız Teknik Üniversitesi Makine Fakültesi Ders Notları, İstanbul.
- Ekin F (2017). Bir gemi dizel motorunda dizayn ve işletme parametrelerinin performans ve emisyon değerleri üzerindeki etkilerinin sayısal olarak incelenmesi [A numerical investigation on the effects of design and operation parameters on the performance and emissions of a marine diesel engine]. İstanbul Teknik Üniversitesi Fen Bilimleri Entitüsü, Yüksek Lisans Tezi, İstanbul.
- Ekin, F., Kafalı, M. & Ozsoysal, O. A. (2018). A Numerical Study of the Effects of Spray Angle and Swirl Ratio on Combustion and Emission of Diesel Engines. International Congress on Engineering and Life Science (ICELIS), Kastamonu-Turkey.
- Gafoor, C. A., & Gupta, R. (2015). Numerical investigation of piston bowl geometry and swirl ratio on emission from diesel engines. Energy Conversion and Management, 101, 541-551.
- Kaario, O., Larmi, M., & Tanner, F. X. (2002). Non-evaporating liquid spray simulations with the ETAB and WAVE droplet breakup models. Zaragoza, 9(11).
- Saraçoğlu H (2018). Sıkıştırmalı ateşlemeli motorlarda tasarım ve işletme koşullarına yönelik parametrelerin performans ve emisyona etkilerinin istatistiksel yöntemlerle incelenmesi [Investigation of the effects of engine design and operating parameters on performance and emissions of compression ignition engines by statistical]. İstanbul Teknik Üniversitesi Fen Bilimleri Entitüsü, Doktora Tezi, İstanbul.
- Soruşbay C (2016). Egzoz Gazı Emisyonları [Exhaust Gases Emissions]. İstanbul Teknik Üniversitesi Makine Fakültesi Ders Notları, İstanbul.
- Sorusbay, C. (1998). Developments in Internal Combustion Engine Technology. In International Conference on Energy and Automotive Technologies, Istanbul, October 17.

- Şahin T (2019). Biyodizel ve bioetanol karışımının tek silindirli bir dizel motorda kullanımının motor performansına ve emisyonlara etkileri [Effects of using biodiesel and bioethanol mixture in single cylinder diesel engine to engine performance and emissions]. Necmettin Erbakan Üniversitesi Fen Bilimleri Entitüsü, Yüksek Lisans Tezi, Konya.
- Wei, S., Ji, K., Leng, X., Wang, F., & Liu, X. (2014). Numerical simulation on effects of spray angle in a swirl chamber combustion system of DI (direct injection) diesel engines. Energy, 75, 289-294.
- Wei, S., Wang, F., Leng, X., Liu, X., & Ji, K. (2013). Numerical analysis on the effect of swirl ratios on swirl chamber combustion system of DI diesel engines. Energy Conversion and Management, 75, 184-190.
- Wu, J., Wang, H. M., Zhu, L. L., & Hua, Y. (2014). Simulation Investigation about Combustion and Emission Characteristics of n-Butanol/Diesel Fuel Mixture on Diesel Engine. In Applied Mechanics and Materials (Vol. 541, pp. 763-768). Trans Tech Publications.
- Xiao, M., & Feng, C. L. (2014). Simulation research on combustion process in a natural gas-diesel dual-fuel marine engine. In Applied Mechanics and Materials (Vol. 525, pp. 227-231). Trans Tech Publications.
- Zeldovich, Y.B., Sadovnikov, P.Y. & Frank-Kamenetskii, D.A. (1947). Oxidation of Nitrogen in Combustion. Academey of Sciences of USSR, Institute of Chemical Physics, Moscow-Leningrad.
- Zhang, Z. R., Wang, Q., He, Z. X., & Dai, L. M. (2014). Numerical Study on Effect of Injection Rates on Combustion and Emission Performance. In Advanced Materials Research (Vol. 860, pp. 1357-1361). Trans Tech Publications.
- Zhang, X. D., Yuan, Y. N., & Du, J. Y. (2015). Simulation Research of the Effect of Compression Ratios on Combustion and Emission for Methanol/Diesel Dual Fuel Engine. In Applied Mechanics and Materials (Vol. 709, pp. 78-82). Trans Tech Publications.



# A Case Study on the Achievement of Students Related to Context-Based Physics Questions

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Abstract: In the context-based teaching processes related outcomes are presented to students through an appropriate context. In this way, students are asked questions about where and why they will use the information in daily life, and motivation of the students are increased. In the context of context-based learning, it is important to use context-based questions for the assessment of courses. Through these questions, students are encouraged to meaningful learning, and are removed from memorization. This cases provides the students with the opportunity to measure the knowledge seen in the daily lives. With the context-based questions, students are also directed to the use of metacognitive skills. In this study, it is aimed to examine the success of teacher candidates for the context based physics questions.

The study was conducted with 30 mathematics teacher candidates who take physics courses. The participants were asked two basic context-based questions. The analysis of the responses was made according to the praxeological approach within the anthropological theory of the didactic. Praxeological approach contains four components that types of tasks and techniques as a part of practical block, technology and theory as a part of theoretical block. In this study, analysis was qualified on the practical and theoretical blocks within a general meaning. Almost all students have failed to make context-based questions. Only a few students answered the 1st the question correctly. Although the students were failed, the equations used by them for the practical block were true. They have no explanation for the theoretical block. That is, the students only learned kinematic equation related with the questions but they did not question the theoretical knowledge of kinematic equations. The results of the research show that the students were memorized kinematic equations only to solve questions. The reason for this failure may be that students are not sufficiently confronted with context-based questions and theoretical block is not sufficiently made.

Keywords: Physics teaching, context based question, anthropological theory, praxeological approach.

## **1. INTRODUCTION**

One of the main objectives of science education is to enable individuals to solve the problems they may face in their daily lives. For this purpose, it is aimed to gain the necessary scientific attitudes and skills to individuals with science education (Captain, 1999; Akgün, 2001). In learning environments, it is very important to create problem situations that students may face in their daily lives. Because the problems they may face in their daily lives are more interesting for students (Boujaoude, 2000). Our education system updates itself to provide more effective training to students. While updating the curriculum, it is aimed to educate students as science literate individuals. Therefore, 'Context-Based Learning' approach has been included in the programs (MEB, 2007; 2013).

Information obtained from context-based approach is transferred to daily life. In this way, individuals are allowed to configure the information they have learned in their minds. In the context-based approach, giving examples from daily life draws the attention of the students who lost their interest in classes (Sjöström & Talanquer, 2014). In this approach, it is aimed to realize permanent learning by structuring the information that individuals have learned in their minds. The use of contexts in the lessons and the processing of the courses according to the context-based approach increase the students' motivation to the lessons and contribute to the understanding of the concepts (Christensson and Sjöström, 2014; Çekiç Toroslu, 2011; Demircioğlu, 2008; İlhan, 2010). Thus, students will have the opportunity to associate the knowledge they have learned in the lessons with their experiences in their daily lives.

In context-based courses, context-based questions should be used during evaluation. In traditionally defined questions, students may not be able to relate to daily life (Benckert, 1997). The questions directed to the students are related to the

daily life of the students to be successful (Rennie and Parker, 1996; Benckert, 1997). Using context-based questions distracts students from memorization and directs reasoning (Elmas and Eryılmaz, 2015; Sak, 2018). Teachers play an active role in the courses designed according to the context-based approach. For this reason, teachers should also have the ability to prepare context-based questions. However, it is determined that teachers do not have enough knowledge about context-based questions (Kurnaz, 2013). It is thought that this is due to the fact that teachers have not met sufficiently with context-based questions. In this study, it was aimed to examine the success of teacher candidates in the context based physics questions.

### 2. METHOD

The survey model was used in this study. The survey model is the type of research that is intended to be described as it is in the past or the present situation (Karasar, 2007). The study was conducted with the participation of 30 pre-service teachers. The data was gathered from an achievement test, and they were analyzed according to the praxeological analysis approach.

### Data collection tool

The achievement test consisting of 2 context based questions was applied to teacher candidates. There are some points to be considered when preparing context-based questions. In the preparation of context-based questions, care should be taken to ensure that it is qualified, understandable and applicable (Ahmad & Pollitt, 2007; Tekbuyık & Akdeniz, 2010; Kurnaz, 2013). The questions in the context-based achievement test, which are prepared with these in mind, are as follows:

### Question 1

The police recruited you as a consultant for a robbery investigation. It is claimed that the bank robbers, who were alleged to be a thief, went to the roof of the building with a lift to escape the security guards and threw the money bag to the friends waiting on the roof of the building. To reach the roof of the side building, the defense lawyer claims that the defendant has to throw the money bag in the horizontal direction with a minimum speed of 10 m/s. The tests, however, reveal that the defendant can dispose of the bag at a maximum speed of 5 m/s. The prosecutor reached the information that the bank building was 250 m high, the side building was 100 m high and the distance between them was 20 m. Do you think the prosecutor can refute the defense lawyer's claim?

### Question 2

You're going uphill with your car. Suddenly, you saw a car coming to the intersection at speed, and you guessed it wasn't going to stop at the red light. You have created a 100 m long straight line of brake traces. A police officer who sees the incident is penalized for passing the red light. You are also penalized for exceeding the speed limit of 36 km / h. When you came home, you read the relevant topic in your physics book and you estimated that the kinetic friction coefficient between the tire and the road was 0.60 and the static friction coefficient was 0.80. When you look at the car's license, you saw that it weighed 2050 N. Will you appeal the traffic ticket?

### Data Analysis

In the analysis of the data obtained within the scope of the study, the praxeological analysis approach, which is part of the anthropological theory of didactics, was used. The praxeological analysis approach can be used to model the theoretical structure of mathematical and other human activities (Winslow, 2006). This approach consists of two basic components: practical and theoretical. In the analysis of the answers, the teacher candidates' use of kinematic equations were evaluated in the practical block. In addition, teacher candidates were asked to determine the laws that validate the equations used in the theoretical block. The responses expected to be given in the practical and theoretical block during these analyzes are presented in Table 1.

Table 1: The responses expected to be given in the practical and theoretical blocks

|            | Practical Block          |                       | Theoretical Block                   |
|------------|--------------------------|-----------------------|-------------------------------------|
| Question 1 | $h = \frac{1}{2}at^2$    | $x = v \cdot t$       | Law of gravitation<br>Law of motion |
| Question 2 | $W = F_s \cdot \Delta x$ | $K = \frac{1}{2}mv^2$ | Work-energy theorem                 |

When Table 1 is examined, the equations that the teacher candidates should use in the practical block of question 1 are  ${}^{\prime}h = \frac{1}{2}at^{2\prime}$  and  ${}^{\prime}x = v \cdot t'$ . In the theoretical block of question 1, they must associate the equations used in the practical block with the gravitational and movement laws. In the practical block of question 2, it is expected from the teacher candidates to use the  ${}^{\prime}W = F_s \cdot \Delta x'$  and  ${}^{\prime}K = \frac{1}{2}mv^{2\prime}$  equations. In the theoretical block of question 2, it is required to associate the equations used with the work-energy theorem.

## **3. FINDINGS**

The answers to the context-based achievement test questions by teacher candidates are presented in Table 2.

|                   | Quality                  | Question 1 | Question 2 |
|-------------------|--------------------------|------------|------------|
|                   | Quanty                   | (f)        | (f)        |
|                   | True                     | 3          | -          |
|                   | Partly True              | 9          | 8          |
| Practical block   | False                    | 6          | 5          |
|                   | Unanswered or unexpected | 6          | 11         |
|                   | answer                   | 0          | 11         |
|                   | True                     | -          | -          |
|                   | Partly True              | -          | -          |
| Theoretical block | False                    | -          | -          |
|                   | Unanswered or unexpected | 24         | 24         |
|                   | answer                   | 24         | 24         |

Table 2: Classification of responses\*

\*Since 6 students did not answer any questions, they were excluded from the analysis.

When Table 2 is examined, it is seen that pre-service teachers give answers using the kinematic equations in practical block of 1st question. Among the answers included in the analysis, it was determined that 3 participants were correct, 9 participants were partially correct, 6 participants were wrong and 6 participants left blank or gave meaningless answers. In the theoretical block of the 1st question, it was seen that all of the teacher candidates did not specify the laws that validate the kinematic equations.

In the practical block of question 2, teacher candidates decide using the work of friction force and kinetic energy equations. When the answers were analyzed, it was determined that no prospective teacher could give the correct answer. It was determined that 8 participants were partially correct, 5 participants were wrong and 11 participants left blank or gave meaningless answers. In the theoretical block of question 2, teacher candidates did not specify the theory which confirms why all the pre-service teachers equalized their equations.

### **Discussion, Conclusion and Recommendations**

In this study, the success of teacher candidates for context based physics questions is examined. The findings were analyzed according to the praxeological analysis approach and examined in detail. Almost all teacher candidates have failed to make context-based questions. Only a few students answered the 1st question correctly. Even though teacher candidates fail, the equations used for the practical block are correct. They have no explanation for the theoretical block. This shows that teacher candidates only memorize kinematic equations to solve the questions.

Context-based problems require a relationship as well as operational solutions (Tekb1y1k & Akdeniz, 2010). While teacher candidates solved context-based questions, they could not establish theoretical block relations with practical blocks. The reason for the failure of teacher trainees to solve context-based questions is thought to be due to the fact that they have

not met sufficiently these types of questions. In the studies conducted, it has been determined that teachers have deficiencies about context based problems (Kurnaz, 2013).

In this study, it was concluded that teacher candidates did not succeed in solving the context based problems. In order to overcome this situation, it is thought that it would be useful to make teacher candidates more interaction with context based questions. In addition, it is recommended that the teachers who will take active roles in the training of future generations to prepare context-based questions and to conduct measurement-evaluation by using these questions.

#### REFERENCES

- Ahmed, A. & Pollitt, A. (2007), Improving The Quality Of Contextualized Questions: An Experimental Investigation Of Focus, Assessment in Education, 14(2), 201-232.
- Benckert, S. (1997), Conversation and context in physics education, *Swedish Council fort he Renewal of Higher Education. Project Report* 161/97.
- Boujaoude S. (2000). What Might Happen If "What might happen if...?: Students Use the Futures Wheel to Analyze Science-Related Social Issues," *The Science Teacher*, 67 (4), pp 45-47.
- Christensson, C., & Sjöström, J. (2014), Chemistry İn Context: Analysis Of Thematic Chemistry Videos Available Online, *Chemistry Education Research and Practice*, 15(1), 59-69.
- Çekiç Toroslu, S. (2011). Yaşam Temelli Öğrenme Yaklaşımı ile Desteklenen 7E Öğrenme Modelinin Öğrencilerin Enerji Konusundaki Başarı, Kavram Yanılgısı ve Bilimsel Süreç Becerilerine Etkisi. Yayınlanmamış Doktora Tezi, Gazi Üniversitesi, Eğitim Bilimleri Enstitüsü, Ankara.
- Demircioğlu, H. (2008). Sınıf Öğretmeni Adaylarına Yönelik Maddenin Halleri Konusuyla İlgili Bağlam Temelli Materyal Geliştirilmesi ve Etkililiğinin Araştırılması. Yayınlanmamış Doktora Tezi, Karadeniz Teknik Üniversitesi, Fen Bilimleri Enstitüsü, Trabzon.
- Elmas R., Eryılmaz A. (2015), How to Write Good Quality Contextual Science Questions: Criteria and Myths, *Kuramsal Eğitimbilim Dergisi*, 8(4), 564-580.
- İlhan, N. (2010). Kimyasal Denge Konusunun Öğrenilmesinde Yaşam Temelli (Context Based) Öğretim Yaklaşımının Etkisi. Yayınlanmamış Doktora Tezi, Atatürk Üniversitesi, Fen Bilimleri Enstitüsü, Erzurum.
- Karasar, N. (2007) Bilimsel Araştırma Yöntemleri, Nobel Yayın, Ankara.
- Kurnaz M. A. (2013), Fizik Öğretmenlerinin Bağlam Temelli Fizik Problemleriyle İlgili Algılamalarının İncelenmesi, *Kastamonu Eğitim Dergisi*, 21(1), 375-390.
- MEB. (2007). Ortaöğretim Fizik Dersi 9. Sınıf Öğretim Programı. Talim Terbiye Kurulu Başkanlığı, Ankara.
- MEB. (2013). Ortaöğretim Fizik Dersi (9, 10, 11 ve 12. sınıflar) Öğretim Programı. Talim Terbiye Kurulu Başkanlığı, Ankara.
- Rennie, L. J. & Parker, L. H. (1996), Placing physics problems in real-life context: students' reactions and performance, Australian Science Teachers Journal, 42(1), 55–59.
- Sak M. (2018), Ortaokul Öğrencilerinin Işık Konusundaki Bağlam Temelli Sorular İle Geleneksel Soruları Cevaplama Düzeylerinin Karşılaştırılması, Yayınlanmamış Yüksek Lisan Tezi, Kocaeli, Fen Bilimleri Enstitüsü.
- Sjöström, J., Talanquer, V. (2014), Humanizing chemistry education: from simple contextualization to multifaceted problematization, *Journal of Chemical Education*, 91(8), 1125-1131.
- Tekbıyık A., Akdeniz A. R.,(2010). Bağlam Temelli ve Geleneksel Fizik Problemlerinin Karşılaştırılması Üzerine Bir İnceleme, Necatibey Eğitim Fakültesi Elektronik Fen ve Matematik Eğitimi Dergisi, 2010, 4(1), 123-140.
- Winslow (2006), Research and development of university level teaching: the interaction of didactical and mathematical organisations.
   In: M. Bosch (ed.) European Research in Mathematics Education IV. Proceedings of the Fourth Congress of the European Society for Research in Mathematics Education, 2006, Barcelona: Universitat Ramon Llull, 1821-1830.



# Investigation of Scientific Attitudes of Science Teacher Candidates in terms of Some Variables

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**Abstract:** The aim of this study is to determine the scientific attitudes of prospective science teachers. The study was carried out with the participation of a total of 55 science teacher candidates studying in the final year in the 2018-2019. A Likert-type scale was used as the data collection tool. The attitude scale used consists of 6 factors that were 'the structure of scientific laws and theories', 'the nature of science and its approach to events'' 'demonstrating scientific behavior', 'the nature and purpose of science', 'the place and importance of science in society' and 'willingness to do scientific studies'. In the study, the interactions of science teacher candidates with 'science, scientific documentaries, book and magazine readings' were analyzed.

Data were analyzed using descriptive statistics. The Cronbach's alpha reliability coefficient was found to be 0.72. It was determined that science teacher candidates have responded to the structure and purpose of the science as undecided and agree with other sub-dimensions. There were significant differences in favor of science teacher candidates who gave positive answers to some questions such as 'Do you read documentaries about scientific research?', 'Do you read books such as books and magazines about scientific research?' and have you joined 'any scientific activity / training / conference, etc.?'. Meanwhile, candidates who have followed documentaries about scientific subjects and researches and read publications such as books and journals and participated in scientific events, training and conferences have been found to have positive attitudes towards science. The study was concluded that the scientific attitudes of the science teacher candidates, scientific documentaries should be more involved in order to increase their interest in science. In addition, the participation of science teacher candidates in scientific activities and trainings may contribute to their attitudes. Based on all these cases, it is recommended that the subject be kept up-to-date in courses and extracurricular learning environments for prospective teachers.

Keywords: Science teacher candidates, attitude, science

### 1. INTRODUCTION

Science is a necessary issue for the advancement of societies. In order for countries to develop, the work of scientists is important. People need to have scientific attitudes as well as knowledge about a subject (Karasar, 2007). Having scientific attitude makes it easier to apply problem solving and research techniques (Ergin & Özgürol, 2011). According to Başaran (1978), scientific attitude enables individuals to be successful and contributes to their development. The foundations of scientific thinking are laid with the scientific attitude of individuals (Yılmaz, 2007). The planning of instruction for the development of scientific attitudes of individuals during education increases the effectiveness (Demirbaş & Yağbasan, 2005). Students who have a positive attitude towards science also have positive attitudes towards their schools, teachers and lessons (Mıhladız & Duran, 2010).

In the courses, students are not expected to memorize scientific information, but they are expected to use the information they have learned to solve the problems they face in daily life. In the curriculum of science education, it is aimed to educate individuals who develop knowledge, think critically and entrepreneurs (MEB, 2018). At the same time, it is aimed to give students high level mental skills. In this way, students will be able to approach the events as scientists and carry out the first steps of scientific learning (Regis, Albertazzi and Roletto, 1996). Students with scientific attitudes have an increase in attitudes towards science (Freedman, 1997; Schibeci, 1983). This situation affects the students' success in science. Students may seek help from their teachers to develop their attitudes (Irwin, 1997). Even if teachers are successful in their fields, having negative attitudes in any subject is thought to cause difficulties in directing students (Önen Öztürk,

2016). In the studies carried out, it has been determined that having negative attitudes towards the teaching process makes the perception of information difficult (Mumba, 2005). It is stated that students who have scientific attitudes will increase their interest towards science and their success (Liaghatdar et al. 2011). The fact that the teachers who give education to the students in schools have scientific attitudes is very important in the transfer of information to future generations. The aim of this study is to determine the scientific attitudes of prospective science teachers. In this study, the interactions of the prospective teachers' scientific attitudes with "their interest in science", "scientific documentaries", "reading books and magazines", "to participate in an activity related to the subject" and "science studies and life stories of scientists" were analyzed. Sub-problems of the study:

1. What is the scientific attitude of the prospective science teachers?

2- Are there any relationship between the scientific attitudes of science teacher candidates and the sub-dimensions of the scale?

### 2. METHOD

In this study which is aimed to determine the scientific attitudes of science teacher candidates, survey model was used. In the survey model, any case that exists in the past or is currently being investigated is to be examined as it is (Karasar, 2007). The study was conducted with a total of 55 pre-service science teachers. 'Scientific Attitude Scale' which was adapted to Turkish by Demirbaş and Yağbasan (2006) was used as a data collection tool to determine prospective teachers' scientific attitudes. 5-point Likert-type ratings were used in the scale. The scale consists of forty items, 20 of which are positive and twenty of which are negative. The scale consists of 6 factors. These are "the structure of scientific laws and theories", "the nature of science and its approach to events" "the presentation of scientific studies". The highest score that can be taken from the scale is 200 and the lowest score is 40. The range of width of the scientific attitude scale was calculated by the formula 'sequence width / number of groups to be performed' (Tekin, 1996). The arithmetic mean intervals used in the evaluation of the findings are as: absolutely I agree (4.21-5.00), I agree (3.41-4.20), undecided (2.61-3.40), I do not agree (1.81-2.60), I strongly disagree (1.00-1.80). The Cronbach's alpha reliability coefficient was found to be 0.72. Demographic data of the participants are in Table 1.

| Individual Features   | Features             | f  | %     |
|---|----------------------|----|-------|
| Candar  | Female               | 44 | 80    |
| Gender  | Male                 | 11 | 20    |
| De vou fellow degumentaries en scientific tenigs/research?        | Yes                  | 44 | 80    |
| Do you follow documentaries on scientific topics/research?        | No                   | 11 | 20    |
| Do you read books such as books, journals about scientific        | Yes                  | 40 | 72,73 |
| topics/research?  | No                   | 15 | 27,27 |
| Are you interested in science and the life stories of scientists? | It interests me      | 47 | 85,45 |
| Are you interested in science and the fire stories of scientists? | I do not interest me | 8  | 14,54 |
| Have you participated in any scientific activity (training /      | I joined             | 38 | 69,09 |
| conference etc.)?   | I did not join       | 17 | 30,91 |

 Table 1. Demographic characteristics of prospective teachers

### 3. FINDINGS AND DISCUSSION

In order to find the answer to the first sub-problem, the total scores obtained from the science attitude scale and the arithmetic mean, standard deviation and minimum and maximum values of the 6 factors are given in Table 2.

 Table 2. Scientific attitude scale descriptive statistics results

| Scientific Attitude Scale                        | Ν  | $\overline{X}$ | SS   | Min  | Max  |
|--|----|----------------|------|------|------|
| The structure of scientific laws and theories    | 55 | 3,41           | 0,37 | 2,83 | 4,33 |
| The nature of science and its approach to events | 55 | 3,68           | 0,44 | 2,00 | 4,50 |
| The presentation of scientific behavior          | 55 | 4,01           | 0,43 | 3,00 | 5,00 |
| The nature and purpose of science                | 55 | 3,18           | 0,27 | 2,50 | 3,83 |

|  |    |      | 1 .  | • . • | 1 1 70 |
|--|----|------|------|-------|--------|
| Total Points                                   | 55 | 3,66 | 0,24 | 3,28  | 4,23   |
| Willingness to do scientific studies           | 55 | 3,79 | 0,44 | 2,90  | 4,80   |
| The importance and place of science in society | 55 | 3,79 | 0,44 | 2,83  | 4,67   |
|  |    |      |      |       |        |

When Table 2 is examined, it can be said that science teachers' scientific attitudes are close to positive level. The independent t test results of 'gender' variables for science teachers candidates are presented in Table 3.

 Table 3. Independent t-test results for gender variable

| Choices | $\overline{\mathbf{X}}$   | Ν  | SS   | t  | р   |
|---------|---|--|--|--|---|
| Yes     | 3,39  | 44   | 0,36   | 1 038  | 0.304   |
| No      | 3,51  | 11   | 0,40   | -1,050   | 0,304   |
| Yes     | 3,71  | 44   | 0,39   | 0.007  | 0 3 2 3   |
| No      | 3,56  | 11   | 0,62   | 0,997  | 0,525   |
| Yes     | 4,00  | 44   | 0,44   | 0.740  | 0.457   |
| No      | 4,11  | 11   | 0,41   | -0,749   | 0,437   |
| Yes     | 3,19  | 44   | 0,28   | 0 372  | 0.712   |
| No      | 3,15  | 11   | 0,25   | 0,572  | 0,712   |
| Yes     | 3,79  | 44   | 0,44   | 0.076  | 0.040   |
| No      | 3,80  | 11   | 0,47   | -0,070   | 0,940   |
| Yes     | 3,78  | 44   | 0,46   | 0.409  | 0.685   |
| No      | 3,84  | 11   | 0,36   | -0,408   | 0,085   |
| Yes     | 3,66  | 44   | 0,24   | 0.212  | 0.756   |
| No      | 3,68  | 11   | 0,24   | -0,512   | 0,730   |
|         | Choices<br>Yes<br>No<br>Yes<br>No<br>Yes<br>No<br>Yes<br>No<br>Yes<br>No<br>Yes<br>No | Choices         X           Yes         3,39           No         3,51           Yes         3,71           No         3,56           Yes         4,00           No         4,11           Yes         3,19           No         3,15           Yes         3,79           No         3,80           Yes         3,78           No         3,84           Yes         3,66           No         3,68 | $\begin{array}{c c c c c c c c c c c c c c c c c c c $ | $\begin{array}{c c c c c c c c c c c c c c c c c c c $ | $\begin{array}{c c c c c c c } \hline \textbf{Choices} & \overline{\textbf{X}} & \textbf{N} & \textbf{ss} & \textbf{t} \\ \hline \textbf{Yes} & 3,39 & 44 & 0,36 \\ \hline \textbf{No} & 3,51 & 11 & 0,40 \\ \hline \textbf{Yes} & 3,71 & 44 & 0,39 \\ \hline \textbf{Yes} & 3,76 & 11 & 0,62 \\ \hline \textbf{Yes} & 4,00 & 44 & 0,44 \\ \hline \textbf{No} & 4,11 & 11 & 0,41 \\ \hline \textbf{Yes} & 3,19 & 44 & 0,28 \\ \hline \textbf{No} & 3,15 & 11 & 0,25 \\ \hline \textbf{Yes} & 3,79 & 44 & 0,44 \\ \hline \textbf{No} & 3,80 & 11 & 0,47 \\ \hline \textbf{Yes} & 3,78 & 44 & 0,44 \\ \hline \textbf{No} & 3,84 & 11 & 0,36 \\ \hline \textbf{Yes} & 3,66 & 44 & 0,24 \\ \hline \textbf{No} & 3,68 & 11 & 0,24 \\ \hline \end{array}$ |

<sup>\*</sup>p<0,05

When the results of t-test in Table 3 are examined, it is determined that there is no significant difference between the gender variable and sub-dimensions of the scale and total scores. The independent t test results of the 'documentary monitoring' variable for science teacher candidates are presented in Table 4.

 Table 4. Independent t-test results for 'documentary monitoring' variable

| Sub-dimensions of Scale                       | Choices | x    | Ν  | SS   | t      | р     |
|---|---------|------|----|------|--------|-------|
| The structure of scientific laws and theories | Yes     | 3,44 | 44 | 0,38 | 0,945  | 0,349 |
|   | No      | 3,32 | 11 | 0,30 |        |       |
| The nature of science and its approach to     | Yes     | 3,71 | 44 | 0,44 | 1,127  | 0,265 |
| events  | No      | 3,54 | 11 | 0,43 |        |       |
| The presentation of scientific behavior       | Yes     | 4,08 | 44 | 0,44 | 2,170  | 0,034 |
|   | No      | 3,77 | 11 | 0,32 |        |       |
| The nature and purpose of science             | Yes     | 3,18 | 44 | 0,28 | -0,041 | 0,967 |
|   | No      | 3,18 | 11 | 0,22 |        |       |
| The importance and place of science in        | Yes     | 3,84 | 44 | 0,46 | 1,476  | 0,146 |
| society                                       | No      | 3,62 | 11 | 0,30 |        |       |
| Willingness to do scientific studies          | Yes     | 3,85 | 44 | 0,44 | 1,756  | 0,085 |
|   | No      | 3,59 | 11 | 0,38 |        |       |
| Total Points                                  | Yes     | 3,70 | 44 | 0,25 | 2,365  | 0,022 |
|   | No      | 3,51 | 11 | 0,13 |        |       |

\*p<0,05

When the results of the t-test in Table 4 are examined, it can be seen that the total scores of the scale and 'exhibiting scientific behavior' sub-dimension show a significant difference according to the scientific documentary monitoring by the teacher candidates. It was found that the attitudes, on 'exhibiting scientific behavior', of the prospective teachers who followed the documentary were more favorable than those who did not watch documentaries. At the same time, it was found that the scientific attitudes of the prospective teachers who followed the documentary were more positive teachers who followed the documentary were more positive than those who did not.

The independent t test results of the 'reading a book, journal' variable for science teacher candidates are presented in Table 5.

Table 5. Independent t-test results for 'reading a book, journal' variable

| Sub-dimensions of Scale                        | Choices | x    | Ν  | SS   | t      | р     |
|--|---------|------|----|------|--------|-------|
| The structure of scientific laws and theories  | Yes     | 3,83 | 40 | 0,36 | -      | 0.240 |
|  | No      | 3,49 | 15 | 0,39 | 0,946  | 0,349 |
| The nature of science and its approach to      | Yes     | 3,71 | 40 | 0,38 | 0.811  | 0.421 |
| events   | No      | 3,60 | 15 | 0,57 | 0,811  | 0,421 |
| The presentation of scientific behavior        | Yes     | 4,07 | 40 | 0,42 | 1 / 87 | 0.1/3 |
|  | No      | 3,88 | 15 | 0,44 | 1,407  | 0,143 |
| The nature and purpose of science              | Yes     | 3,18 | 40 | 0,29 | 0 202  | 0.8/1 |
|  | No      | 3,17 | 15 | 0,22 | 0,202  | 0,841 |
| The importance and place of science in society | Yes     | 3,86 | 40 | 0,47 | 1 816  | 0.075 |
|  | No      | 3,62 | 15 | 0,28 | 1,010  | 0,075 |
| Willingness to do scientific studies           | Yes     | 3,88 | 40 | 0,45 | 2 168  | 0.017 |
|  | No      | 3,57 | 15 | 0,34 | 2,408  | 0,017 |
| Total Points                                   | Yes     | 3,70 | 40 | 0,26 | 2 062  | 0.044 |
|  | No      | 3,55 | 15 | 0,15 | 2,002  | 0,044 |

\*p<0,05

When the t test results in Table 5 were analyzed, it can be seen that the total scores of the scale and 'willingness to do scientific studies' sub-dimension show a significant difference according to the reading a book or a journal by the teacher candidates. It was determined that the scientific attitudes of the prospective teachers who read books or journals were more positive than those who did not. The independent t test results of the 'science studies and life stories of scientists' variable for science teacher candidates are presented in Table 6.

Table 6. Independent t-test results for 'science studies and life stories of scientists' variable

| Sub-dimensions of Scale                        | Choices              | X    | Ν  | SS   | t     | р     |
|--|----------------------|------|----|------|-------|-------|
| The structure of scientific laws and           | It interests me      | 3,41 | 47 | 0,39 | 0.134 | 0.804 |
| theories                                       | I do not interest me | 3,40 | 8  | 0,22 | 0,154 | 0,094 |
| The nature of science and its                  | It interests me      | 3,68 | 47 | 0,45 | 0.084 | 0.034 |
| approach to events                             | I do not interest me | 3,67 | 8  | 0,43 | 0,084 | 0,934 |
| The presentation of scientific                 | It interests me      | 4,05 | 47 | 0,46 | 0.042 | 0.251 |
| behavior                                       | I do not interest me | 3,85 | 8  | 0,19 | 0,045 | 0,231 |
| The nature and purpose of science It interests |                      | 3,17 | 47 | 0,27 | 0.602 | 0 573 |
|  | I do not interest me | 3,23 | 8  | 0,28 | 0,092 | 0,575 |
| The importance and place of                    | It interests me      | 3,85 | 47 | 0,42 | 0.604 | 0.027 |
| science in society                             | I do not interest me | 3,48 | 8  | 0,41 | 0,004 | 0,027 |
| Willingness to do scientific studies           | It interests me      | 3,83 | 47 | 0,44 | 0 722 | 0.206 |
|  | I do not interest me | 3,61 | 8  | 0,46 | 0,723 | 0,200 |
| Total Points                                   | It interests me      | 3,68 | 47 | 0,24 | 0.803 | 0.148 |
|  | I do not interest me | 3,55 | 8  | 0,24 | 0,895 | 0,148 |

#### \*p<0,05

When the t test results in Table 6 were analyzed, it can be seen that the 'the importance and place of science in society' sub-dimension show a significant difference according to the attract attention by the teacher candidates on "science studies and life stories of scientists".

The independent t test results of the 'participating in an activity related to the subject' variable for science teacher candidates are presented in Table 7.

**Table 7.** Independent t-test results for 'participating in an activity related to the subject' variable

| Sub-dimensions of Scale                       | Choices   | X    | Ν  | SS   | t      | р     |
|---|-----------|------|----|------|--------|-------|
| The structure of scientific laws and theories | I joined  | 3,40 | 38 | 0,38 | 0 388  | 0 600 |
|   | I did not | 3,44 | 17 | 0,35 | -0,588 | 0,099 |
| The nature of science and its approach to     | I joined  | 3,62 | 38 | 0,46 | 1 425  | 0 160 |
| events  | I did not | 3,80 | 17 | 0,37 | -1,423 | 0,100 |
|   | I joined  | 3,94 | 38 | 0,47 |        |       |

| The presentation of scientific behavior | I did not | 4,19 | 17 | 0,28 | -1,974  | 0,054 |
|---|-----------|------|----|------|---------|-------|
| The nature and purpose of science       | I joined  | 3,20 | 38 | 0,28 | 0.760   | 0.451 |
|   | I did not | 3,14 | 17 | 0,24 | 0,700   | 0,431 |
| The importance and place of science in  | I joined  | 3,71 | 38 | 0,42 | 2 1 8 1 | 0.034 |
| society                                 | I did not | 3,98 | 17 | 0,44 | -2,101  | 0,034 |
| Willingness to do scientific studies    | I joined  | 3,74 | 38 | 0,41 | 1 202   | 0 108 |
|   | I did not | 3,91 | 17 | 0,49 | -1,505  | 0,196 |
| Total Points                            | I joined  | 3,62 | 38 | 0,24 | 2.007   | 0.041 |
|   | I did not | 3,76 | 17 | 0,21 | -2,097  | 0,041 |

### \*p<0,05

When the t test results in Table 7 were analyzed, it can be seen that the total scores of the scale and 'the importance and place of science in society' sub-dimension show a significant difference according to the attract attention by the teacher candidates on 'participating in an activity related to the subject'. The attitudes of the prospective teachers who did not participate in the scientific activity were found to be more positive than the participants. As summarize, based on the findings, it was determined that prospective teachers who read documentaries related to scientific subjects and researches and who read publications such as books and magazines have positive attitudes towards science.

#### 4. CONCLUSION AND SUGGESTIONS

It was concluded that the scientific attitudes of the prospective science teachers were close to the positive level. When the related literature is examined, there are studies with similar results (e.g. Önen, 2013). Teachers who have an active role in the upbringing of students have positive attitudes towards science and are important in terms of directing students. In addition, it was determined that teacher candidates who read publications such as scientific books and journals have positive attitudes towards science. In the light of the findings of the research process, it is thought that teacher candidates should be more involved in scientific documentaries in order to increase their interest in science during their education. Introducing the lives of scientists to individuals to give importance to science education (Ata, 1999). In addition, science teachers can be provided with access to publications such as scientific attitudes should be kept up-to-date in course and extracurricular settings for prospective teachers.

#### REFERENCES

- Ata, E. İlköğretim Öğrencilerinde Bilimsel ve Sosyal Tutum. Adapazarı Örneği. Yayınlanmamış Yüksek Lisans Tezi. Sakarya Üniversitesi Sosyal Bilimler Enstitüsü, 1999
- Başaran, İ. E. Eğitim Psikolojisi. Ankara: Bilim Matbaası, 1978.
- Demirbaş, M., & Yağbasan, R. (2005). Sosyal öğrenme teorisine dayalı öğretim etkinliklerinin, öğrencilerin bilimsel tutumlarının kalıcılığına olan etkisinin incelenmesi. *Uludağ Üniversitesi Eğitim Fakültesi Dergisi*, 18(2), 363-382.
- Demirbaş, M., & Yağbasan, R. (2006). Fen bilgisi öğretiminde bilimsel tutumların işlevsel önemi ve bilimsel tutum ölçeğinin Türkçe'ye uyarlanma çalışması. *Uludağ üniversitesi eğitim fakültesi dergisi*, 19(2), 271-299.
- Ergin, d. Y., & Özgürol, ö. G. M. B. (2011). Bilimsel tutum ve duygusal zeka arasındaki ilişki. 2 nd International Conference on New Trends in Education and Their Implications, 27-29 April, 2011 Antalya-Turkey
- Freedman, M.P. Relationship Among Laboratory Instruction, Attitude Toward Science and Achievement in Science Knowledge. Journal of Research In Science Teaching, 1997: 34, 4, 343-357.
- Irwin, L. H. Teachers' Role In Multicultural Education: Setting the Stage for Preservice Teachers. *Contemporary Education*, 1997: 68, (4), 217-219.
- Karasar, N. (2007) Bilimsel Araştırma Yöntemleri, Nobel Yayın, Ankara.
- Liaghatdar, M. J., Soltani, A. & Abedi, A. (2011). A validity study of attitudes toward science scale among Iranian secondary school students. *International Education Studies*. 4(4), 36-46.
- MEB (2018). Fen bilimleri dersi öğretim programi (İlkokul ve Ortaokul 3, 4, 5, 6, 7 ve 8.Sınıflar). Ankara: *Millî Eğitim Bakanliği Temel Eğitim Genel Müdürlüğü*. Taslak Program.
- Mıhladız, G. & Duran, M. (2010). İlköğretim Öğrencilerinin Bilime Yönelik Tutumlarının Demografik Değişkenler Açısından İncelenmesi. *Mehmet Akif Ersoy Üniversitesi Eğitim Fakültesi Dergisi*. 10(20), 100 121.

- Mumba, F. (2005). *Influence of explicit instruction and reflection on mathematics and science teaching fellows' views of the nature of science*. (Unpublished doctoral dissertation). Illionis State University. Urbana, Illinois.
- Önen, F. (2013). Fen bilgisi öğretmen adaylarının aktivite temelli bilimin doğası öğretimine yönelik görüşleri ile bu öğretimin bilimsel tutum ve süreç becerilerine etkisi. *The Journal of Academic Social Science Studies*, 6(7), 843-868. Doi number: http://dx.doi.org/10.9761/JASSS1671
- Öztürk, F. Ö. (2016). Bilimsel epistemolojik inançlar, bilimsel bilginin doğası hakkındaki görüşler ve bilimsel tutumlar üzerine bir çalışma: Abu Dhabi örneği. *Muğla Sutkı Koçman Üniversitesi Eğitim Fakültesi Dergisi*, 3(2).
- Regis, A., Albertazzi, P. G., Roletto, E. Concept Maps In Chemistry Education. *Journal of Chemistry Education*, 1996: 73, (11), 1084-1088.
- Schibeci, R.A. Selecting Appropriate Attitudinal Objectives for School Science. Science Education, 1983: 67, (5), 595-603.
- Tekin, H. (1996). Eğitimde Ölçme ve Değerlendirme. 9. Baskı, Ankara: Yargı Yayınları.
- Yılmaz, F. (2007). İlköğretim I. kademede bilimsel tutum ve davranış kazandırmada fen bilgisi dersinin etkililiğine ilişkin öğretmen görüşleri. İlköğretim Online. 6 (1), 113-126.



# Theoretical and Experimental Comparison of Bulk Module of Tetragonal Bi-2212 Superconductor Materials

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**Abstract:** In the present study, the solid state reaction method is preferred for the production of superconductor materials. During the procedure, the Bi-2212 superconductor samples doped with Nd (Neodymium) are produced. The main objective in the present study is to examine the effects of both of Nd doping and solid state reaction method, which was chosen as the production method, on the structural and mechanical properties of Bi-2212 superconductor materials. Especially, the effects of doping on the mechanical properties of materials are discussed over the bulk module. It is aimed to obtain information about the mechanical properties by comparing the bulk modules calculated theoretically and experimentally. Besides that, the XRD measurement is performed in order to characterize the structural properties.

Keywords: Solid state reaction method, tetragonal, bulk modulus, Bi-2212

## **1. INTRODUCTION**

Nowadays it seems that technological device, equipment, vehicles, machine and constructions which are manufactured and used for various industrial purposes are manufactured from materials such as metal, ceramic, glass, plastic, paper, composites that have different properties [1-4]. The selection of the most qualified, the most durable and the most economic materials is very important due to the service conditions, the applied load type and value and the place of use of each product by design engineers. The mechanic, electrical, optical, thermal, corrosion properties of materials must be known very well for selection of material.

Generally it is seen that two different load types as static or dynamic effect to materials. The various mechanical properties of materials are determined depending on these applied load type. For example, the fatigue toughness is one of important properties of the materials is identified by the behavior of the material under the applied dynamic load. On the other hand, the properties of materials such as the tensile strength, yield strength, fracture toughness, toughness, ductility, elastic modulus and hardness are determined by the behavior of the material under applied static load [5,6].

The aim of this study is to investigate the role of Nd addition on the structural, and mechanical properties of Bi-2212 ceramics. The microhardness and XRD measurements are investigated for mechanical and structural properties, respectively.

## 2. MATERIAL AND METHODS

In this study, conventional solid state reaction method is used to prepare the Nd added Bi-2212 samples. Nd<sub>2</sub>O<sub>3</sub> (Alfa Aesar Co., Ltd. 99.9 % purity) at the rate of x = 0.01, 0.02, 0.03, 0.04 and 0.05 % has been doped to Bi<sub>2</sub>Sr<sub>2</sub>CaCu<sub>2</sub>O<sub>x</sub> (Alfa Aesar Co., Ltd. 99.9 % purity) superconducting powder. Undoped sample is produced under the same conditions for comparison with doped samples. Precursor powders of Bi-2212 superconducting are grinded in an agate mortar for 8 hour in the air atmosphere and pressed under a force of 7 ton at room temperature. The pellets are subjected to the sintering process in the sintering time range of 72 at 840°C, in air atmosphere. The change of the crystal structure, phase impurity, texturing, grain size and lattice parameters of the Bi-2212 superconductors is examined by XRD measurements performed by a Bruker D8 Advance model X-ray diffractometer. Moreover, a SHIMADZU HVM-2 model digital microhardness tester is used to determine the mechanical properties of the bulk Bi-2212 superconductors. The produced powder samples containing Nd are named Nd1, Nd2, Nd3, Nd4, Nd5 and Nd0 of undoped sample.

#### 3. RESULTS AND DISCUSSION

#### Results

*XRD Measurements:* X-ray diffraction method was used to determine the crystal structures and lattice parameters of the materials. XRD measurements were performed by Bruker D8 Advance X-ray powder diffractometer using  $CuK_{\alpha}$  ( $\lambda = 1,541$ Å) radiation in the range of  $3^0 \le 2\theta \le 60^0$  at a scan speed of  $4^\circ$ /min. The grain size and lattice parameters can easily be analyzed by the diffraction patterns belonging to a superconducting material. Figure 1 shows XRD patterns for the superconductor ceramics. The characteristic peaks identifying with Bi- 2212 phase are displayed by (h k l) miller indices on the XRD graphs. In addition, the characteristic peak intensities increase with the annealing duration at the annealing temperature of 840°C, confirming that the crystallinity and crystal plane alignments of the superconducting samples improve significantly (Fig. 1). Lattice parameters *a* and *c* and grain size values are summarized in Table 1.

| Samples | c (Å) | a (Å) | Grain Size (Å) |
|---------|-------|-------|----------------|
| Nd0     | 30.54 | 5.35  | 107.86         |
| Nd1     | 30.49 | 5.37  | 97.29          |
| Nd2     | 30.45 | 5.38  | 85.41          |
| Nd3     | 30.43 | 5.39  | 61.85          |
| Nd4     | 30.26 | 5.40  | 47.38          |
| Nd5     | 30.06 | 5.41  | 34.33          |

Table 1. Lattice parameters a and c and grain size values

In this study, it is seen that the lattice parameters and crystal structure obtained for the Lu doped Bi-2212 system conformed with the literature (Table 1). In addition, grain sizes of the samples are calculated using the Warren-Scherrer equation [7]. As can be seen from Table 1, the Lu doping is decreased the grain size compared with the undoped sample.



Figure 1. XRD patterns of all samples

*Vickers Microhardness Measurements:* The most common and effective experiment carried out in order to determine the mechanical properties of the materials is the hardness measurement. The main reasons for this are that the procedure is simple and that it damages the sample less than the other procedures do. The other advantage is that the hardness of a material is directly proportionate to the other mechanical properties. For instance; the tensile strength of material is directly proportionate to the hardness. Thus, it is possible to have an idea about the strength of the material by measuring the hardness.

Vickers microhardness test is used in this study. In this test, indenter is immersed on the surface of the material under a load selected according to the material with a certain period and diagonal length of indentation is measured. Vickers microhardness value;

$$H_{\nu} = 1854.4 \, (F/d^2) \tag{1}$$

is calculated using Eqn 1. Here *F* is calculated as  $(d_1+d_2)/2$  and *d* are applied load and diagonal length, respectively. In this study, in order to investigate the aging on the mechanical properties, Vickers microhardness measurements are carried out. Measurements ae taken from sample surfaces. *F* is changed between 0.245-2.940 N for 10 s. Obtained values are designated with an average of 7 readings at different region of the sample surfaces.





The microhardness values obtained experimentally are presented in Table 2. Moreover, the diagram of the change in microhardness depending on the force applied ( $H_v$ -F) is presented in Figure 2. As seen in the figure, the microhardness values decreased in parallel with the load applied. This behavior is defined as ISE (Indentation Size Effect) in the literature. These materials exhibit both the elastic and plastic deformation. Furthermore, as it can be seen in Figure 2, the microhardness values does not change when the loads higher than 1.5 N are applied. In and after this region that is called the saturation region, there would be no significant change in the hardness value even if the load applied is increased [8-10]. While calculating the microhardness values, the bulk modules of the material were calculated by using Equation (2).

$$B = 81.9635 \,H_V \tag{2}$$

| Samples | <b>F</b> ( <b>N</b> ) | Hv (GPa) | B (GPa) |  |
|---------|-----------------------|----------|---------|--|
| Nd0     | 0.245                 | 0.434    | 35.57   |  |
|         | 0.490                 | 0.417    | 34.17   |  |
|         | 0.980                 | 0.397    | 32.53   |  |
|         | 1.960                 | 0.392    | 32.12   |  |
|         | 2.940                 | 0.390    | 31.96   |  |
| Nd1     | 0.245                 | 0.427    | 34.99   |  |
|         | 0.490                 | 0.400    | 32.78   |  |
|         | 0.980                 | 0.388    | 31.80   |  |
|         | 1.960                 | 0.385    | 31.55   |  |
|         | 2.940                 | 0.382    | 31.30   |  |
| Nd2     | 0.245                 | 0.418    | 34.26   |  |
|         | 0.490                 | 0.391    | 32.04   |  |
|         | 0.980                 | 0.378    | 30.98   |  |
|         | 1.960                 | 0.367    | 30.08   |  |
|         | 2.940                 | 0.364    | 29.83   |  |
| Nd3     | 0.245                 | 0.405    | 33.19   |  |
|         | 0.490                 | 0.363    | 29.75   |  |
|         | 0.980                 | 0.335    | 27.45   |  |
|         | 1.960                 | 0.312    | 25.57   |  |
|         | 2.940                 | 0.305    | 24.99   |  |
| Nd4     | 0.245                 | 0.318    | 26.06   |  |
|         | 0.490                 | 0.267    | 21.88   |  |
|         | 0.980                 | 0.248    | 20.32   |  |
|         | 1.960                 | 0.246    | 20.16   |  |
|         | 2.940                 | 0.238    | 19.50   |  |
| Nd5     | 0.245                 | 0.285    | 23.35   |  |
|         | 0.490                 | 0.249    | 20.40   |  |
|         | 0.980                 | 0.229    | 18.76   |  |
|         | 1.960                 | 0.218    | 17.86   |  |
|         | 2.940                 | 0.210    | 17.21   |  |

Table 2. Values of F, H<sub>v</sub> and B of all samples

In order to be able to compare the experimental results that we obtained to the theoretical results, an empirical approach is developed for the bulk module. In this approach, it is possible to obtain a projectable value in examining the bulk modules of all the compounds consisting of elements between I and VII. Given the results, there is an approximately scale for these materials.

The *d* here refers to the bond length of the crystal structure. The doping applied to the materials may cause local planar defects on the samples. These defects cause extra reflections arising from *n* different planes differing from the XRD patterns. These changes create changes in the mean length of bond.

$$B = 550d^{3}$$
Bond length  $d = \sqrt{\frac{a^{2}}{9} + c^{2} \left(\frac{1}{4} - u\right)^{2}}$ 
(3)
(4)

*u* is related with c/a.

$$u = 0.24\sqrt{1 - a^2/c^2} \tag{5}$$

The calculated values are different from the experimental values. This can be explained with the difference of samples' porosity from the estimated values. Moreover, the theoretically calculated B value depends on the bond length. The bond length depends on the lattice parameters. Since the crystal structure of examined materials does not change with the doping, no significant change is observed in the lattice parameters and consequently the bond lengths. Besides that, the bulk module values obtained experimentally are calculated with a force applied on the surface of sample. The possible accumulation at the particle borders or porosity changing with the doping directly affects the hardness measurements. Detecting this difference is almost impossible in theoretical calculations. Thus, the difference between the theoretically calculated B values and the experimentally obtained ones can be explained.



Figure 3. Comparasion of theoretical and experimental results Table 3. Obtained results from the measurements and calculations

| Samples | Bond<br>Length | u     | Theoretical<br>B | Experimental B<br>(Average) | Theoretical<br>H | Experimental H<br>(Average) |
|---------|----------------|-------|------------------|-----------------------------|------------------|-----------------------------|
| Nd0     | 1.917          | 0.260 | 78.055           | 33.27                       | 0.952            | 0.406                       |
| Nd1     | 1.924          | 0.260 | 77.153           | 32.48                       | 0.941            | 0.396                       |
| Nd2     | 1.928          | 0.260 | 76.704           | 31.43                       | 0.935            | 0.383                       |
| Nd3     | 1.932          | 0.260 | 76.262           | 28.19                       | 0.930            | 0.344                       |
| Nd4     | 1.936          | 0.260 | 75.793           | 21.58                       | 0.924            | 0.263                       |
| Nd5     | 1.940          | 0.260 | 75.320           | 19.51                       | 0.918            | 0.238                       |

### Discussion

When the obtained results are investigated, we can say that the characteristic peak intensities increase with the annealing duration at the annealing temperature of 840°C, confirming that the crystallinity and crystal plane alignments of the superconducting samples improve significantly in the XRD measurement. In addition, we can say that all superconducting materials exhibit both the elastic and plastic deformation. And the microhardness values does not change when the loads higher than 1.5 N are applied. Also the bulk module values obtained experimentally are calculated with a force applied on the surface of sample. The possible accumulation at the particle borders or porosity changing with the doping directly affects the microhardness measurements.

### 4. CONCLUSION

In the present study, the Lu doped Bi-2212 based high-temperature superconducting samples are produced using solis state reasction method and their structural and mechanical properties are analyzed. The results obtained are summarized below.

• In diffraction patterns obtained from XRD analyses, no remarkable second phase or peak related with Lu ion was observed. All of the phases obtained here are the characteristic phases of Bi-2212.

- It can be seen that the *a* lattice parameters of the samples increased and *c* lattice parameters decreased. The Lu doping is decreased the grain size compared with the undoped sample.
- The microhardness values decreaseed with increasing the applied load on the surface. That is, the material exhibited ISE behavior.
- The microhardness and bulk modulus results are calculated as experimentally and theoretically. When the obtained results are compared, it is determined that the theoretical B values are higher. This difference can be explained with the difference of samples' porosity from the estimations and the result that the crystal structure does not change with the Lu doping.

#### REFERENCES

- [1] Smith R L and Sandland G E (1922). An accurate method of determining the hardness of metals, with particular reference to those of a high degree of hardness. Proceed. Inst. Mech. Engin., 1: 623-641.
- [2] Khalil S M (2001). Enhancement of superconducting and mechanical properties in BSCCO with Pb additions. J. Phys. Chem. Solids, 62: 457-466.
- [3] Asikuzun E, Ozturk O, Cetinkara H A, Yildirim G, Varilci A, Yılmazlar M, Terzioglu C. (2012). Vickers hardness measurements and some physical properties of Pr<sub>2</sub>O<sub>3</sub> doped Bi-2212 superconductors. J. Mat. Sci. Mater. in Elect., 23: 1001-1010.
- [4] Khalil S M (2005). Role of rare-earth Ba<sup>2+</sup> doping in governing the superconducting and mechanical characteristics of Bi-Sr-Ca-Cu-O. Smart. Mater. and Struc., 14: 804.
- [5] Koralay H, Arslan A, Cavdar S, Ozturk O, Asikuzun E, Gunen A, Tasci A T (2013). Structural and Mechanical Characterizations of Bi1.75Pb0.25Sr2Ca2Cu3-xSnxO10+δ Superconductor Ceramics Using Vickers Microhardness Test. J. Mat. Sci. Mater. in Elect., 24: 4270-4278.
- [6] Martin Cohen L (1985). Calculation of bulk moduli of diamind and zinc-blende solids. Phys. Rev. B., 32: 7988-7991.
- [7] Asikuzun E, Donmez A, Arda L, Cakiroglu O, Ozturk O, Akcan D, Terzioglu C (2015). Structural and mechanical properties of (Co/Mg) co-doped nano ZnO. Cer. Inter., 41: 6326-6334.
- [8] Arda L, Ozturk O, Asikuzun E, Ataoglu S (2013). Structural and mechanical properties of transition metals doped ZnMgO nanoparticles. Powder Techn., 235: 479-484.
- [9] Elmustafa A A, Stone D S (2003). Nanoindentation and the indentation size effect: Kinetics of deformation and strain gradient plasticity. J. Mech. and Phy. Sol., 51: 357-381.
- [10] Ozturk O, Asikuzun E and Yildirim G (2013). The role of Lu doping on microstructural and superconducting properties of Bi2Sr2CaLuxCu2Oy superconducting system. J. Mat. Sci. Mater. in Elect., 24: 1274-1281.



# Effect of Cutting Parameters on Surface Roughness in Turning of 7075-T6 Aluminum Alloys

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**Abstract:** The purpose of this study is to investigate the effect of cutting parameters (cutting speed - feed rate - tool radius) on surface roughness in turning of 7075-T6 aluminum alloys. For this purpose, machining tests on 7075-T6 aluminum alloy was performed to CNC lathe.

In experimental studies, dry cutting was applied for 60 mm diameter samples. These tests were performed by choosing constant cutting depths (0.5 mm), three different cutting speeds (190-280-375 m/min), three different feed rate values (0.05-0.15-0.25 mm/rev) and two different tool corner radius (0.4-0.8 mm). In the experiments, carbide cutting tools commonly used for turning aluminum alloys are used.

The results showed that the surface roughness decreased associated with the increase of cutting tool corner radius, the increase of the cutting speed effected the surface roughness a little bit and also the increase in the feed rate increased the surface roughness greatly.

As a consequence, 0.8 mm cutting tool corner radius can be used at high cutting speeds. The feed rate value should be chosen according to the desired surface roughness value.

Keywords: 7075 Aluminium alloy, Turning, Cutting parameters, Surface roughness

## **1. INTRODUCTION**

In recent years, because of its unique combination of properties, aluminium has become one of the most versatile structural and engineering materials in the industry (Çakır et al., 2016). Aluminum alloys have many varieties due to the differences in chemical composition. One of these alloys is 7075 aluminum alloy. This alloy is an aluminum alloy containing zinc as the primary alloy element (Rao and Allamraju, 2017). Due to the good proportions between their weight and their mechanical properties, these materials are used for the production of different elements that make up automotive and aviation vehicles. A good surface quality is required for these industry products (Agustina et al., 2009).

The shaping of 7075 aluminum alloys can be carried out with each of the manufacturing methods applied to metals. Machining methods are also commonly used methods. The quality criteria of the products which are shaped by the machining methods are taken into account and the main criteria such as surface roughness, size accuracy and shape accuracy are taken into consideration. There are many parameters that affect quality. Some of these parameters are cutting speed, feed, depth of cut, cutting fluids and cutting tool geometries.

In the literature for the 7075 aluminum alloys, there are studies investigating the effects of cutting speed and feed to surface roughness by using different cutting tool corner radius. A. Çakır et al., the effect of cutting parameters on surface roughness have investigated with what 0,4 mm cutting tool corner radius, two different feed (0,1 - 0,2 mm/rev) and four different cutting speeds (150 - 300 m/min). The results show that the increase in feed rate clearly increases the surface roughness, while the increase in the cutting speed is less effective. M. Pul, the effect of cutting parameters on surface roughness have investigated with 0,8 mm cutting tool corner radius, three different feed (0,25 - 0,40 mm/rev) and four different cutting speeds (200 - 400 m/min). The experimental data obtained show that the increase of the feed rate increases the surface roughness clearly and that the cutting speed does not have any effect. T. Dişbudak and S. Sensoy, the effect of cutting parameters on surface roughness have investigated with 0,8 mm acutting speed (90 - 500 m/min). The research results show that

both cutting parameters increase the value of the surface roughness clearly. S. J. Raykar et al., the effect of cutting parameters on surface roughness have investigated with what 0,8 mm cutting tool corner radius, three different feed (0,1 - 0,3 mm/rev) and three different cutting speeds (200 - 370 m/min). The research results show that the increase of the feed rate increases the surface roughness clearly and that the cutting speed does not have any effect. I. Zagórski ve T. Warda, the effect of cutting parameters on surface roughness have investigated with what 0,2 mm cutting tool corner radius, six different feed (0,05 - 0,15 mm/rev) and three different cutting speeds (700 - 900 m/min). The research results show that the increase of the feed rate increases the surface roughness clearly and that the cutting speeds (700 - 900 m/min). The research results show that the increase of the feed rate increases the surface roughness clearly and that the cutting speed does not have any effect. In the turning of 7075 aluminum alloys, the recommended cutting parameters values for good surface quality are feed rate (0.1-0.15 mm/rev) and cutting speed (200-400 m/min).

In this study, the effect of cutting tool corner radius value on surface roughness was investigated by using 0.4 mm and 0.8 mm cutting tool corner radius which are widely recommended in the sector. Furthermore, the surface roughness with the recommended feed and cutting speed range values was examined. The data obtained were presented graphically and the relationship between surface roughness and process time was examined.

### 2. MATERIAL and METHODS

Experiments were carried out for different cutting parameters in the machining of 7075-T6 aluminium alloys. The results of chemical analysis and hardness value of the test material are given in Table 1.

| Matarial |     | Alloy | elements | %     |       |       |        |
|----------|-----|-------|----------|-------|-------|-------|--------|
| Material | HB  | Si    | Cu       | Zn    | Cr    | Mg    | Al     |
| 7075-T6  | 168 | 0.162 | 1.525    | 4.568 | 0.188 | 2.079 | 91.232 |

Table 1 Results of chemical analysis and hardness value of the test material.

The process of turning, to which the alloy was subjected in the study, was performed on the ECOCA - MT 312 CNC turning machine without cooling system. Turning was carried out with KC WNMG 080408 A12 and Sumitomo VBGT 160404R-Ay cutting tool inserts. Machining process was conducted at constant depth of cut (0,5 mm), and changing range for two different cutting tool corner radius of other technological parameters: f=0.05-0.15-0.25 mm/rev, Vc=190-280-375 m/min. The machined length was equal to L = 30 mm.

The surface roughness values were measured by a Mahr-MarSurf Ps1 surface roughness device.

### **3. RESULTS AND DISCUSSION**

The graph showing the effect of the cutting parameters on the surface roughness of the turning process applied to 7075-T6 samples is presented in Figure 1-2.



Figure. 1 Surface roughness relationship graph of feed rates for radius 0.4 and 0.8.

Figure 1 shows the increase in surface roughness values with increasing feed rate values. This result is consistent with the literature (Çakır, 2016; Pul, 2017; Dişbudak and Şensoy, 2014; Raykar, 2015). Surface roughness values are

considerably lower than the two studies in the literature (Çakır, 2016; Dişbudak and Şensoy, 2014). Surface roughness values are higher in 0.4 mm cutting tool corner radius. The results showed that smaller surface roughness values could be obtained with 0.8 mm cutting tool corner radius.



Figure. 2 Surface roughness relationship graph of cutting speeds for radius 0.4 and 0.8.

Fig. 2 shows that the effect of the cutting speed on the surface roughness is negligible. This result implies that there is no drawback in terms of surface roughness of the work at high speeds taking into account the cutting tool life.

In machining, as well as achieving the desired quality, the processing time is also a very important value. Therefore, the effect of the cutting parameters on the machining time is examined in figure 3.



Figure. 3 Cutting parameters – machining time relationship graph

Figure 3 shows the result that the cutting speed increase decreases the machiniging time as expected. The effect of 0.05 mm/rev feed rate on the machining time is very high. Other feed rate values affect the machining time much less.

The surface roughness values required in the industry vary according to the working conditions of the workpiece. Therefore, the relationship between surface roughness and machining time was examined and the results are presented in figure 4.



Figure. 4 Surface roughness - machining time relationship graph

Figure 4 shows that the desired surface roughness value has a significant effect on the machining time. Especially when a surface roughness of less than 1 micrometer is desired, the machining time increases by 2.5 times. The effect of the range of 1-4 micrometer surface roughness values on the machining time is much less than the surface roughness value of less than 1 micrometer.

### 4. CONCLUSIONS

The results of the effects of the cutting parameters on the surface roughness and the machining time in the experimental studies applied to 7075-T6 aluminum alloy are as follows:

- Increased feed rate increased surface roughness.
- The effect of the cutting speed on the surface roughness is negligible.

- The effect of 0.05 mm/rev feed rate on the machining time is very high. Other feed rate values affect the machining time much less.

- Optimal cutting parameters:

Vc=375 m/min - f=0,05 mm/rev (0,4 and 0,8 Radius) for Ra<sub>min</sub>=0,5  $\mu$ m,

Vc=375 m/min - f=0,15 mm/rev (0,8 Radius) for Ramin=1 µm,

Vc=280 m/min - f=0,25 mm/rev (0,8 Radius) for Ramin=2 µm,

Vc=375 m/min - f=0,15 mm/rev (0,4 Radius) for Ramin=2 µm,

Vc=375 m/min - f=0,25 mm/rev (0,8 Radius) for Ramin=3 µm

and Vc=375 m/min - f=0,25 mm/rev (0,4 Radius) for Ra<sub>min</sub>=4  $\mu$ m.

- Smaller surface roughness values could be obtained with 0.8 mm cutting tool corner radius.

#### REFERENCES

- Agustina B., Rubio E. M., Villeta M. and Sebastián M. A., 2009. Analysis Of The Surface Roughness Obtained During The Dry Turning Of UNS A97050-T7 Aluminium Alloys. In: Third Manufacturing Engineering Society International Conference, 03 December, pp. 11-20 (<u>https://doi.org/10.1063/1.3273621</u>)
- Çakır A., Yağmur S., Kavak N., Küçüktürk G. and Şeker U., 2016. The effect of minimum quantity lubrication under different parameters in the turning of AA7075 and AA2024 aluminium alloys. The International Journal of Advanced Manufacturing Technology 84: 2515–2521.
- Dişbudak T. And Şensoy S., 2014. An Analysis on the Effects of Cutting Parameters over Surface Roughness of 7075 Aluminum during Turning Operation. Journal of Engineering and Technological Sciences 2(1): 13-29
- Pul M., 2017. Comparison of Surface Roughness and Tool Wear in Turning of 7075, 6061 and 2024 Aluminum Alloys. International Journal of Engineering Research and Development 9(2): 65-75
- Rao K. S. S. and Allamraju K.V., 2017. Effect on Micro-Hardness and Residual Stress in CNC Turning Of Aluminium 7075 Alloy. Materials Today: Proceedings 4: 975–981.
- Raykara S.J., D'Addonab D.M. and Manea A.M., 2015. Multi-objective optimization of high speed turning of Al 7075 using grey relational analysis. Procedia CIRP 33: 293 298
- Zagórski I. and Warda T., 2018. Effect of Technological Parameters on The Surface Rougness of Aluminium Alloys after Turning. Advances in Science and Technology Research Journal 12(2): 144-149



# Determination the Effectiveness of Acidic Pumice, Waste Glass Powder, Blast Furnace Slag and Colemanite Ore Waste on some Engineering Properties of Concretes by Taguchi Method

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**Abstract:** In recent years, it is very common to use mineral materials as supplementary cementitious materials for the concrete production. By this way, producers aim to reduce the cement usage amount, make more economic and environmental-friendly concretes. Because, cement is the most expensive material among the concrete components. Also, cement production processes have hazardous effects for the environment such as carbon dioxide emission. At the same time, some of these mineral materials are industrial by-products, which are considered as wastes if they cannot be utilized. In this study, as mineral materials, acidic pumice (AP), waste glass powder (GP), ground granulated blast furnace slag (GGBFS) and colemanite ore wastes (CW) were used. AP, GP and GGBFS were used 5%, 10% and 20% instead of cement. These amounts are 1%, 3% and 5% for CW, due to the set-retarding effect of boron mineral in cement. 50x50x50 mm cubic concrete samples were produced by using these materials. Concrete samples were cured for 180 days. After curing period, samples were subjected to compressive strength and ultrasonic pulse velocity tests. Besides, unit weights, water absorption and capillary water absorption values were measured. In order to reduce the number of the experiments for the combinations, an L<sub>16</sub> Taguchi design was used. By this way, the effect level of the factors (mineral materials) can be determined. Besides, in order to determine the exact effectiveness percentage of each material, ANOVA was performed to the results. The results indicated that, for all the experiments, the effectiveness rankings of the mineral additives are; AP, GP, GGBFS and CW, respectively.

Keywords: Pumice, blast furnace slag, waste glass, waste colemanite, Taguchi

## **1. INTRODUCTION**

Concrete is the most widely used construction material due to its cheapness and good engineering properties. It is consisting of cement, aggregates and water. In order to improve the engineering properties of hardened concrete, different additives are used (Nagrockiene et al. 2017). By using these additives, it is possible to solve any problems about obtaining concretes with specified characteristics. Additive usage provides higher strengths, lower permeability and higher durability. Besides, they are generally used instead of cement (approximately 5-20%). Therefore, the cement consumption decreases (Akchurin et al. 2016).

Every year, million tons of glass wastes (GP) are generated and most of them are landfilled or recycled. Landfilled glass wastes cause a growing environmental problem due to non-biodegradable in nature (Mirzahosseini and Riding, 2014, Sadiqul Islam et al., 2017). Glass powder named as a pozzolanic material due to its chemical composition. Glass particles dissolve in the water and the combination of this solution and hydrated cement products forms calcium-silicate-hydrate (CSH) gels (Elaqra and Rustom, 2018). Therefore, fine glass powder is used for reducing alkali-silica reaction, decreasing shrinkage or as cement replacement material due to its highly pozzolanic nature (Jani and Hoglan, 2014, Pereira et al. 2012, Vijayakumar et al. 2013).

Ground granulated blast furnace slag (GGBFS) is one of the most common supplementary cementitious materials (Tan et al., 2019). It is a by-product acquired in the production of pig iron (Özbay et al., 2016). It is used in order to reduce clinker usage in the cement production which its production has economic and environmental drawbacks (Wu et al., 2018). Besides, it is used as a cement replacement material due to its pozzolanic reaction (Tan et al., 2019). The common usage of GGBFS is generally related to the durability advantages provided. It is reported that GGBFS usage reduces the permeability (Chia and Zhang, 2002), alkali-silica reaction (Kwon, 2005), sulfate attack (Higgins, 2003).

Colemanite is one of the most common boron minerals. Turkey has rich boron reserves and most of them are colemanite. The boron minerals are subjected to concentration at production sites. As a result of this process, some boric acid containing waste occurs and these wastes create environmental problems such as groundwater contamination (Sevim, 2011). In the literature, there are limited number of studies on colemanite waste (CW) utilization in concrete. It is reported that colemanite waste can be used up to 5% in cementitious materials and increases the compressive strength in long term (Olgun et al., 2007, Kula et al., 2001).

Pumice is a volcanic origin natural material. It has a very porous and glassy structure. Besides the chemical composition of pumice shows that it is rich by silicate and alumina (Khandaker, 2004). Pumice is used in concrete due to its thermal and acoustic properties. Besides, low density of pumice makes it attractive in lightweight concrete production (Gencel, 2015).

In this study, acidic pumice, waste glass powder, GGBFS and colemanite ore waste were used as cement replacement material. A Taguchi design was prepared in order to reduce the number of the experiments and determine the effectiveness levels of the additives on the properties of concretes. Compressive strength, ultrasonic pulse velocity, unit weight, waster absorption and capillary water absorption tests were performed on 180 days cured samples.

## 2. MATERIAL AND METHODS

#### Materials

In the study CEM I 42.5 grade ordinary Portland cement was used. Acidic pumice (AP) was obtained from Kayseri territory. CW was obtained from ETI Mine Works, GP was obtained from MARCAM Heating Co. in Kahramanmaraş region and GGBFS was obtained from ISDEMIR (Iskenderun Iron and Steel Works Co.) (Fig. 1). The chemical and physical properties of the materials are given in Table 1.

| Compound                                    | Cement | AP    | GGBFS | GP    | CW    |  |  |  |  |
|---|--------|-------|-------|-------|-------|--|--|--|--|
| (%)   |        |       |       |       |       |  |  |  |  |
| Chemical Properties                         |        |       |       |       |       |  |  |  |  |
| SiO <sub>2</sub>                            | 18.63  | 68.25 | 40.29 | 71.12 | 34.54 |  |  |  |  |
| Al <sub>2</sub> O <sub>3</sub>              | 5.38   | 14.80 | 12.56 | 0.81  | 9.82  |  |  |  |  |
| Fe <sub>2</sub> O <sub>3</sub>              | 2.80   | 3.06  | 1.13  | 0.23  | 5.11  |  |  |  |  |
| CaO   | 63.39  | 2.92  | 35.20 | 9.27  | 12.57 |  |  |  |  |
| MgO   | 3.41   | 1.01  | 8.35  | 4.07  | -     |  |  |  |  |
| SO <sub>3</sub>                             | 2.47   | -     | -     | 0.24  | -     |  |  |  |  |
| Na <sub>2</sub> O+K <sub>2</sub> O          | 0.60   | 7.03  | -     | 13.13 | -     |  |  |  |  |
| <b>B</b> <sub>2</sub> <b>O</b> <sub>3</sub> | -      | -     | -     | -     | 18.85 |  |  |  |  |
| *LOI (%)                                    | 0.53   | -     | -     | -     | 10.62 |  |  |  |  |
| Physical Prope                              | erties |       |       |       |       |  |  |  |  |
| Specific                                    | 3.14   | 2.26  | 2.88  | 2.62  | 2.42  |  |  |  |  |
| Gravity                                     |        |       |       |       |       |  |  |  |  |
| Fineness                                    | 3310   | 3212  | 5500  | 3682  | 3415  |  |  |  |  |
| (cm <sup>2</sup> /g)                        |        |       |       |       |       |  |  |  |  |

Table 1. Chemical and physical analysis of materials

\*Loss on ignition



a) Acidic pumice

(b) Waste glass powder



(c) Ground granulated blast furnace slag



(d) Colemanite ore waste

Figure 1. Mineral additives

As aggregates 0-4mm crushed sand and 4-10mm crushed stone were used. The specific gravities are 2.71 and 2.78, water absorptions are 1.8% and 1.03%, fineness moduli are 3.75 and 6.82 for fine and coarse aggregate, respectively. The grain distribution of the mixture aggregate is given in Fig. 2.



Figure 2. Gradation curve of mixture aggregate

## Methods

In this study, Taguchi experimental design method was used. The main purpose of using experimental design methods is to provide maximum information with minimum experiments (Taguchi et al., 1993). In this study an  $L_{16}$  orthogonal design was used. In this design, as factors, mineral additives and as levels, the usage percentages were considered. Table 2. shows the factors and their levels.

Table 2. Factors and their levels for  $L_{16}$  design

| Factors  | Level 1 | Level 2 | Level 3 | Level 4 |
|----------|---------|---------|---------|---------|
| AP, %    | 0       | 5       | 10      | 20      |
| GP, %    | 0       | 5       | 10      | 20      |
| GGBFS, % | 0       | 5       | 10      | 20      |
| CW, %    | 0       | 1       | 3       | 5       |

The mixture proportions due to  $L_{16}$  design shown in Table 3. In this table reference sample, AP, GP, GGBFS and CW coded as "R", "P", "G", "S" and "C", respectively. Water, fine aggregate and coarse aggregate amount kept constant in all the mixtures as 160kg/m<sup>3</sup>, 740kg/m<sup>3</sup> and 1248kg/m<sup>3</sup>, respectively.

Table 2. Mixture proportions

| Trial | Code | AP | GP | GGBFS | CW | Cement | Water | FA  | CA   |
|-------|------|----|----|-------|----|--------|-------|-----|------|
| 1     | R    | 0  | 0  | 0     | 0  | 320    | 160   | 740 | 1248 |
| 2     | GSC1 | 0  | 5  | 5     | 1  | 284.8  | 160   | 740 | 1248 |
| 3     | GSC2 | 0  | 10 | 10    | 3  | 246.4  | 160   | 740 | 1248 |
| 4     | GSC3 | 0  | 20 | 20    | 5  | 176    | 160   | 740 | 1248 |
| 5     | PSC1 | 5  | 0  | 5     | 3  | 278.4  | 160   | 740 | 1248 |

| 6  | PGC1  | 5  | 5  | 0  | 5 | 272   | 160 | 740 | 1248 |
|----|-------|----|----|----|---|-------|-----|-----|------|
| 7  | PGS1  | 5  | 10 | 20 | 0 | 208   | 160 | 740 | 1248 |
| 8  | PGSC1 | 5  | 20 | 10 | 1 | 204.8 | 160 | 740 | 1248 |
| 9  | PSC2  | 10 | 0  | 10 | 5 | 240   | 160 | 740 | 1248 |
| 10 | PGSC2 | 10 | 5  | 20 | 3 | 198.4 | 160 | 740 | 1248 |
| 11 | PGC2  | 10 | 10 | 0  | 1 | 252.8 | 160 | 740 | 1248 |
| 12 | PGS2  | 10 | 20 | 5  | 0 | 208   | 160 | 740 | 1248 |
| 13 | PSC3  | 20 | 0  | 20 | 1 | 188.8 | 160 | 740 | 1248 |
| 14 | PGS3  | 20 | 5  | 10 | 0 | 208   | 160 | 740 | 1248 |
| 15 | PGSC3 | 20 | 10 | 5  | 5 | 192   | 160 | 740 | 1248 |
| 16 | PGC3  | 20 | 20 | 0  | 3 | 182.4 | 160 | 740 | 1248 |

\*FA: Fine aggregate CA: Coarse aggregate

Boron containing minerals has retarding effect on cement hydration. Therefore, CW cannot be utilized in higher amounts such as other minerals. According to Table 3, fresh mixtures produced and cast into 50x50x50mm cubic molds. After one day, samples demolded and cured in lime saturated water for 180 days. After curing period, unit weights, water absorption and capillary water absorption values were determined. Besides, samples were subjected to ultrasonic pulse velocity and compressive strength tests. The results analyzed according to Taguchi experimental design analyze and the effectiveness of each factor on tests was determined. In order to obtain the contribution level of each factor on results, ANOVA was performed.

#### **3. RESULTS AND DISCUSSION**

#### **Unit Weight**

The results of the unit weight test are given in Fig. 3.

The unit weights of the samples changes between 2.2-2.3g/cm<sup>3</sup>. There is no significant change between samples by mineral addition. The highest value was obtained from PGS1 and the lowest value was obtained from PGSC3.



Figure 3. Unit weights of samples

### Ultrasonic Pulse Velocity (UPV)

The UPV results are given in Figure 4.



Figure 4. Ultrasonic pulse velocity results

The results of UPV changes between 4688-5529m/s. The reference sample is 5496m/s. The highest value was obtained from PSC1 and the lowest was obtained from PGSC3. This can be related to the substitution amount with cement and the pumice content. Pumice is a very porous material and higher the pumice amount increases the pore amount in the structure.

### **Compressive Strength**

The results of compressive strength test are given in Fig. 5. Fig. 5(a) shows the real values and Fig. 5(b) shows the relative values according to the reference sample.

The reference sample has 52.5MPa compressive strength value. The strength values change between 37.4, which was obtained from PGSC3 and 54.4, which was obtained from PSC1. The highest value has a relative value of 103.62% while the lowest one 71.24%.



Figure 5. Compressive strength results

### **Relationship between Compressive Strength and UPV**

The relationship between compressive strength and UPV is given in Figure 6.



Figure 6. Relationship between compressive strength and UPV

UPV is an indication of pore structure. The main principle of the test is ultrasonic waves go across the porous sample slower and in a denser material the velocity of the wave is faster. It can be said that the relation between pore structure and compressive strength also creates a relation between UPV and compressive strength. Figure 6 shows that there is a strong relation between compressive strength results and UPV. The  $R^2$  value is approximately 0.96.

### Water Absorption and Capillary Water Suction

The results of water absorption and capillary water absorption are given in Fig. 7.



Figure 7. Water absorption and capillary water absorption results

The highest water absorption and capillary absorption value was obtained from PGSC3 and the lowest values were obtained from PSC1. The highest water absorption value is 5.29% and the highest capillarity coefficient is  $17.15g/cm^2s^{-1}$ . The lowest water absorption value is 4.01% while reference value is 4.12%. The lowest capillarity coefficient is  $5.99g/cm^2s^{-1}$  while reference value is  $8.44g/cm^2s^{-1}$ .

#### **Design of Experiments**

The signal/noise (S/N) ratios according to the Taguchi analyze are given in Tables 3-6 for each test. At UPV and compressive strength larger is better option was chosen and for water and capillary water absorption smaller is better option was chosen.

| Level | AP     | GP     | GGBFS  | CW     |  |
|-------|--------|--------|--------|--------|--|
| 1     | 74.48  | 74.48* | 74.38* | 74.30  |  |
| 2     | 74.51* | 74.36  | 74.28  | 74.33* |  |
| 3     | 74.27  | 74.24  | 74.31  | 74.26  |  |
| 4     | 73.70  | 73.88  | 73.99  | 74.07  |  |

Table 3. S/N ratios for UPV results

| Rank | 1 | 2 | 3 | 4 |
|------|---|---|---|---|
|      |   |   |   |   |

\*Optimum level

| Level | AP     | GP     | GGBFS  | CW     |  |
|-------|--------|--------|--------|--------|--|
| 1     | 33.83  | 33.86* | 33.56* | 33.40  |  |
| 2     | 33.94* | 33.59  | 33.42  | 33.53* |  |
| 3     | 33.37  | 33.24  | 33.47  | 33.37  |  |
| 4     | 32.11  | 32.56  | 32.79  | 32.94  |  |
| Rank  | 1      | 2      | 3      | 4      |  |

Table 4. S/N ratios for compressive strength results

\*Optimum level

Table 5. S/N ratios for water absorption results

| Level | AP      | GP      | GGBFS   | CW      |  |
|-------|---------|---------|---------|---------|--|
| 1     | -12.80* | -12.84* | -12.93* | -13.14  |  |
| 2     | -12.81  | -12.99  | -13.05  | -13.13* |  |
| 3     | -13.20  | -13.25  | -13.23  | -13.21  |  |
| 4     | -14.13  | -13.86  | -13.74  | -13.46  |  |
| Rank  | 1       | 2       | 3       | 4       |  |

\*Optimum level

Table 6. S/N ratios for capillary water absorption results

| Level | AP     | GP     | GGBFS          | CW             |  |
|-------|--------|--------|----------------|----------------|--|
| 1     | 80.35* | 80.21* | 79.10          | 78.41          |  |
| 2     | 79.87  | 79.11  | <b>79.98</b> * | 79.22          |  |
| 3     | 78.02  | 78.37  | 78.29          | <b>79.40</b> * |  |
| 4     | 76.17  | 76.72  | 77.05          | 77.38          |  |
| Rank  | 1      | 2      | 3              | 4              |  |

\*Optimum level

According to the Taguchi results, the optimum mixture for UPV,  $P_0G_0S_0C_1$ , for compressive strength,  $P_5G_0S_0C_1$ , for water absorption,  $P_0G_0S_0C_1$  and for capillary water absorption,  $P_0G_0S_5C_3$ . The results indicate that for all of the tests, AP is the most effective parameter.  $2^{nd}$  is GP,  $3^{rd}$  is GGBFS and the last one is CW. In order to determine the contribution percentage, ANOVA was performed. The contribution percentages are given in Table 7.

Table 7. Contribution level of admixtures on test results according to ANOVA

| Admixture | Rank | UPV(%) | Compressive  | Water          | <b>Capillary Water</b> |
|-----------|------|--------|--------------|----------------|------------------------|
|           |      |        | Strength (%) | Absorption (%) | Absorption (%)         |
| AP        | 1    | 53.91  | 55.54        | 49.98          | 42.16                  |
| GP        | 2    | 25.65  | 25.14        | 25.91          | 24.84                  |
| GGBFS     | 3    | 11.47  | 9.96         | 16.18          | 18.08                  |
| CW        | 4    | 3.80   | 5.22         | 3.10           | 9.80                   |

ANOVA results show the contribution percentages of the materials on test. Accordingly, AP has the highest contribution levels on every test which are over 40% for all test. The second highest contribution was obtained from GP, which is approximately around 25% for all the tests. Third effective parameter is GGBFS which is effective between 9.96% and 18.08%. The last parameter is CW, which has a contribution of 9.80% at most.

### 4. CONCLUSION

According to the experimental study and the results of the experimental design the conclusions can be listed as follows;

• According to Taguchi results, AP is the most effective parameter for all tests. This is due to the porous structure of AP. ANOVA indicated that the contribution level of the AP on the results are over 40%.

- Second effective parameter is GP. The contribution level to the results is around 25%.
- The third effective parameter is GGBFS. The contribution level changes between 9.96%-18.08%
- The lowest effectiveness was obtained from CW. This result can be related to the lower addition amount of this admixture.

#### REFERENCES

- Akchurin T K, Tukhareli V D, Pushkarskaya O Yu (2016). The modifying additive for concrete compositions based on the oil rafinery waste. Procedia Engineering, 150: 1485-1490.
- Chia K S, Zhang M H (2002). Water permeability and chloride penetrability of high-strength lightweight aggregate concrete. Cement and Concrete Research, 32: 645-659.
- Elaqra H, Rustom R (2018). Effect of using glass powder as cement replacement on rheological and mechaical properties of cement paste. Construction and Building Materials, 179: 326-335.
- Gencel O (2015). Characteristics of fired clay brics with pumice additive. Energy and Buildings, 102: 217-224.
- Higgihns D D (2003). Increased sulfate resistance of GGBS concrete in the presence of carbonate. Cement and Concrete Composites, 25(8): 913-919.
- Jani Y, Hoglan W (2014). Waste glass in the production of cement and concrete a review. Journal of Environmental Chemical Engineering, 2(3): 1767-1775.
- Khandaker M A H (2004). Properties of volcanic pumice based cement and lightweight concrete. Cement and Concrete Research, 34: 283-291.
- Kula I, Olgun A, Erdoğan Y, Sevinc V (2003). Effects of colemanite waste, coal bottom ash, and fly ash on the properties of cement. Cement and Concrete Research, 31: 491-494.
- Kwon Y J (2005). A study on the alkali-aggregate reaction in high-strength concrete with particular respect to the ground granulated blast-furnace slag effect. Cement and Concrete Research, 35: 1305-1313.
- Mirzahosseini M, Riding K A (2014). Effect of curing temperature and glass type on the pozzolanic reactivity of glass powder. Cement and Concrete Research, 58: 103-111.
- Nagrockiene D, Girskas G, Skripkiunas G (2017). Properties of concrete modified with mineral additives. Construction and Building Materials, 135: 37-42.
- Olgun A, Kavas T, Erdoğan Y, Once G (2007). Physico-chemical characteristics of chemically activated cement containing boron. Building and Environment, 42: 2384-2395.
- Özbay E, Erdemir M, Durmuş H İ (2016). Utilization and efficienct of ground granulated blast furnace slag on concrete properties A review. Construction and Building Materials, 105: 423-434.
- Pereira L A, Castro-Gomes J P, Santos P M S (2012). The potential pozzolanic activity of glass and red clay ceramic waste as cement mortar components. Construction and Building Materials, 31: 197-203.
- Sadiqul Islam G M, Rahman M H, Kazi N (2017). Waste glass powder as partial replacement of cement for sustainable concrete practice. International Journal of Sustainable Built Environment, 6: 37-44.
- Sevim U K (2011). Colemanite ore waste concrete with low shrinkage and high split tensile strength. Materials and Structures, 44: 187-193.
- Taguchi G, Yokoyama Y, Wu Y (1993). Taguchi methods. Design of Experiments. American Supplier Instute, pp: 59-63.
- Tan H, Nie K, He X, Guo Y, Zhang X, Deng X, Su Y, Yang J (2019). Effect of organic alkali on compressive strength and hydration of wet-grinded granulated blast furnace slag containing Portland cement. Construction and Building Materials, 206: 10-18.
- Vijayakumar G, Vishaliny H, Govindarajulu D (2013). Studies on glass powder as partial replacement of cement in concrete production. International Journal of Emerging Technology and Advanced Engineering, 3(2): 153-157.
- Wu M, Zhang Y, Ji Y, Liu G, Liu C, She W, Sun W (2018). Reducing environmental impacts and carbon emissions: Study on effects of superfine cement particles on blended cement containing high volume mineral admixtures. Journal of Cleaner Production, 196: 358-369.



# The Effect of Co Doping on Structural and Mechanical Properties of Y123 Phase

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**Abstract:** In this study, different proportions of Co nanoparticles are substituted with Cu in the Y123 structure produced by solid state reaction method (YBa<sub>2</sub>Cu<sub>3-x</sub>Co<sub>x</sub>O<sub>7- $\delta$ </sub>, x=0.10 ve 0.15). Changes in the structural and mechanical properties on the Y123 structure were investigated. Lattice parameters of the samples were calculated by X-ray diffraction (XRD) analysis. In this way, the effects of Co atom on the crystal structure of Y123 phase were examined. In addition, the surface morphology of the samples were examined by scanning electron microscopy (SEM). For the analysis of mechanical properties, Vicker's microhardness analysis was performed by using different loads and the microhardness values are obtained increased with the doping ratio. The samples produced showed Indentation Size Effect (ISE) behavior. The elastic (Young) modulus and yield strength values were calculated by using the data obtained from Vickers microhardness analysis. In addition, Meyer's law, Hays-Kendall (HK) approach and indentation-induced cracking (IIC) model were applied to obtain the independent microhardness values. It was concluded that the most suitable model for explaining RISE behavior is the IIC model for the produced samples.

Keywords: YBCO, Y123, high temperature superconductors, solid state reaction method, Meyer's law, HK, IIC

## **1. INTRODUCTION**

After the discovery of high temperature superconductors (HTS) which have a transition temperature value above the fluid helium temperature, the YBCO family has become a very popular field of study. The fact that the YBCO family has different phases and each phase has a different critical temperature value has led to the growth of this study area. In many studies, Y123 phase was preferred and different elements were added to this phase and their properties were examined. Structural, electrical, mechanical, magnetic and many other features have been examined and the front of it is used for different technological applications (Supandanaison 2018, Matskevich 2014, Dihom 2017, Ramil 2016, Kurnaz 2017).

As a result of the contributions made for the Y123 phase; lattice parameters of the crystalline structure, critical temperature value, magnetic levitation properties, mechanical properties are examined. Examination of these changes is important for shaping the material according to the area of use.

The solid-state reaction method is one of the most preferred production methods among other methods. The critical temperature values of some of the samples produced and doped by this method increase while some others decrease. Similarly, there are also changes in the crystal lattice parameters (Guner 2019, Yılmaz 2012, Khalid 2018, Pinemtel 2014).

In some studies in which the effect of Co contribution on Y123 phase was examined, it was seen that the value of superconducting transition temperature decreased with contribution. Here, the change in lattice parameters and structural defects were investigated and it was concluded that Co-Cu substitution caused a decrease in the critical temperature value (Liu 2002, Boudjema 2010, Anderson 1993).

In this study, the structural and mechanical properties of the samples obtained for the Co doped Y123 phase produced by solid state reaction were investigated.
## 2. MATERIALS AND METHODS

Co doped superconducting materials were prepared by using solid state reaction method. For obtaining samples;  $Y_2O_3$  (Yttrium (III) oxide %99.99, Alfa Aesar), BaCO<sub>3</sub> (Barium carbonate %99.95, Alfa Aesar), CuO (Copper (II) oxide %99.9995, Alfa Aesar) and Co-nanoparticle (Cobalt powder-325 mesh %99.5, Alfa Aesar) powders were used. Two different ratios were selected for the doping process (0.10 and 0.15% w) Powders were ground for 1 hour in agate mortar to obtain more stable mixing. Samples were calcined 3 times at 850°C for 24 hours in ash furnace. After each calcination powders grounded for 1 hour. Obtained powders were pressed into bulk form (13mm diameter and 2mm thickness). The bulk samples were placed in tube furnace on the alumina crucible for sintering process. Samples sintered at 930°C for 24 hours by heating rate of 5°C/min and then cooled to 500°C in 60 minutes. The samples were kept in the oxygen atmosphere for 5 hours at 500°C. Samples are named as Y123-U, Y123-10 and Y123-15 in accordance with doping ratio.

The XRD analysis of the samples performed by Broker D8 Advance X-ray diffractometer with CuK $\alpha$  in the range of  $2\theta$ =3-90°. Surface morphology investigated by FEI Quanta Feg 250 model scanning electron microscopy. Vicker's microhardness test performed to obtain mechanical properties of produced samples and Meyer's Law Hays-Kendall (HK) and Indentation-Induced Cracking (IIC) models applied for obtaining load independent hardness values.

# **3. RESULTS AND DISCUSSION**

## **Structural Analysis**

The XRD patterns of all samples produced in  $Y_1Ba_2Cu_{3-x}Co_xO_{7-\delta}$  general formula with the x = 0, 0.10 and 0.15 doping ratios are given in Figure 1. When XRD patterns were examined, it was seen that peaks indicated by miller indices were peaks of Y123 phase. The fact that Co peaks were not found in XRD patterns showed that Co atoms entered into orthorhombic Y123 structure by doping process. The displacement of Co<sup>+3</sup> (ionic radius 0.745Å) with Cu<sup>+2</sup> (ionic radius 0.73Å) played an important role in the introduction of Co atoms as a result of the doping process. Co atoms entering the structure caused some changes in peak intensity but did not corrupt orthorhombic structure.



Figure 1: XRD patterns and miller indices of produced samples

Table 1. Lattice parameters and particle size values

| Sample  | Grain Size (Å) | a (Å) | b (Å) | c (Å)  |
|---------|----------------|-------|-------|--------|
| Y123-U  | 28.95          | 3.815 | 3.882 | 11.683 |
| Y123-10 | 37.63          | 3.822 | 3.884 | 11.687 |
| Y123-15 | 38.13          | 3.826 | 3.879 | 11.691 |

Since Y123 is an orthorhombic structure; d the distance between the planes and the data such as the a, b, and c parameters were calculated by formula,

$$\frac{1}{d^2} = \frac{h^2}{a^2} + \frac{k^2}{b^2} + \frac{l^2}{c^2} \tag{1}$$

using the indices h, k, l from the XRD. Grain size, D, calculated using formulas

$$D = 0.941\lambda/B\cos\theta \tag{2}$$

$$B^2 = B_s^2 - B_m^2$$
(3)

here  $B_s$  is the full width half of maximum (FWHM) and  $B_m$  is 0,000007 (Pimentel 2014).

In the literature, the lattice parameters of Y123 structure are given as a=3.82Å, b=3.89Å and c=11.7Å respectively. It is seen that the lattice parameters of Y123 for superconducting samples with Co doping produced using solid state reaction method are in accordance with the literature. The grain size values given in Table 1 show that the particle size increases with the increase of Co doping.



Figure 2. SEM images of samples produced by solid state reaction method (a; Y123-U, b; Y123-10, c; Y123-15)

SEM analysis was performed to determine the particle boundaries, inter-particle distance and particle size of Co doped samples obtained by solid state reaction method. Figure 2 contains the SEM images of the samples. In Figure 2 large images were taken at 2500 magnification, while internal pictures were taken at 10000 magnification. From the SEM images, it is seen that the particle sizes increase with Co contribution. This change is in agreement with the particle size values calculated by XRD results. The variation of particle boundaries and inter-particle distance is clearly seen in Figure 2. In addition, the increased particle size with the doping also led to a decrease in porosity.

#### **Microhardness and Modelling**

It is important to determine the mechanical properties for a superconducting material. It is important that certain properties such as hardness, yield stress and elastic modulus be determined and optimized for proper operation. Vickers microhardness test is the most common method for the investigation of mechanical properties (Ozturk 2012).

The resistance of the material to the applied load on the surface is defined as hardness. The material hardness may vary with the processes performed during the production phase (Safran 2015).

The top angle of the pyramid shaped indenter tip used in the Vickers microhardness test is  $\theta$ =136°. Vickers microhardness value calculated by using formula,

$$H_V = 1854.4 \left(\frac{F}{d^2}\right) \tag{4}$$

Here, F is the load applied to the surface (F= 0.245, 0.490, 0.980, 1.960 ve 2.940N), and d is the average of the diagonal lengths of the indenter tip that is left on the material surface.

In addition, elastic modulus (E) and yield stress (Y) values of material were calculated by,

$$E = 81.9635 H_V$$
 (5)

$$Y \approx H_V / 3 \tag{6}$$

using empirical formulas.



Figure 4. Microhardnes graph of produced samples

The graph of the hardness values obtained for the samples in this study according to the applied load is given in Figure 4. This graph shows an increase in the hardness values with the increase of the contribution rate. In the literature, this behavior is defined as Reverse Indentation Size Effect (RISE) which is the increasing of the the microhardness with increasing of the applied load. Calculated datas are given in Table 2.

| Sample | <b>F</b> ( <b>N</b> ) | H <sub>V</sub> (GPa) | E (GPa) | Y (GPa) |
|--------|-----------------------|----------------------|---------|---------|
|        | 0.245                 | 2.881                | 236.15  | 0.960   |
|        | 0.490                 | 3.143                | 257.63  | 1.048   |
| ed     | 0.980                 | 3.420                | 280.28  | 1.140   |
| lop    | 1.960                 | 3.768                | 308.80  | 1.256   |
| Unc    | 2.940                 | 3.755                | 307.80  | 1.252   |
|        |                       |                      |         |         |
|        | 0.245                 | 3.313                | 271.51  | 1.104   |
|        | 0.490                 | 3.492                | 286.23  | 1.164   |
| 0      | 0.980                 | 3.887                | 318.59  | 1.296   |
| 3-1    | 1.960                 | 4.157                | 340.73  | 1.386   |
| Y12    | 2.940                 | 4.175                | 342.20  | 1.392   |
|        |                       |                      |         |         |
|        | 0.245                 | 3,378                | 276,86  | 1,126   |
|        | 0.490                 | 3,680                | 301,58  | 1,227   |
| Ŋ      | 0.980                 | 4,123                | 337,94  | 1,374   |
| 3-1    | 1.960                 | 4,255                | 348,79  | 1,418   |
| Y12    | 2.940                 | 4,296                | 352,11  | 1,432   |

Table 2. Microhardness, elastic modulus and stress values of Co doped samples produced by solid state reaction method

### Meyer's law

Meyeradırs law is usually used to describe the behavior of materials ISE (Indentation Size Effect). In this method, which is a simple expression between F and d, the formula

(7)

 $F = Ad^n$ 

is used. In this formula, the n, also known as the Meyer number, is the slope of the lnF-lnd graph (Figure 5). The point where the graph intercepts the y axis gives A value (Ozturk 2012, Safran 2015). If the number n is less than 2, it is proved that the material exhibits ISE behavior. The data obtained for this model are given in Table 3.



Figure 5. Variation of applied load lnF with diagonal lnd for the samples

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Table 3. Best-fit results of experimental data according to Meyer's law

| Samples | n        | lnA (GPa) | Hv (GPa)    |
|---------|----------|-----------|-------------|
| Undoped | 2,255103 | -7,10359  | 3,420-3,755 |
| Y123-10 | 2,225155 | -6,88184  | 3,887-4,175 |
| Y123-15 | 2,220995 | -6,82854  | 4,123-4,296 |

## Hays-Kendall approach (HK)

In this method, it is suggested that the minimum load value (W) is required to create permanent deformation. If the load applied to the surface does not exceed this value, permanent deformation will occur and only elastic deformation will occur (Ozturk 2018).

$$F - W_{HK} = A_{HK} d^2 \tag{8}$$

Here,  $A_{HK}$  is load independent constant and these values are obtained from F-d<sup>2</sup> graph (Figure 6). Load independent microhardness value for this model is,

$$H_{HK} = 1854.4A_{HK}$$
 (9)



Figure 6. Applied load versus the square of the impression semi-diagonal length for the samples

Table 4 shows the values obtained for Hays-Kendall model. When the values in the table are examined, it is seen that W value is negative for all samples. This shows that all samples exhibit RISE behavior. However, when the HHK values given in the table are examined, it is seen that these values are far from the plateau region. In this case, this model is not sufficient to determine the hardness.

Table 4. Best-fit results of experimental data according to HK model

| Samples | W(N)     | Ax10 <sup>-3</sup> | H <sub>HK</sub> (Gpa) | H <sub>v</sub> (GPa) |
|---------|----------|--------------------|-----------------------|----------------------|
| Undoped | -0,10744 | 2.11               | 3,913                 | 3,420-3,755          |
| Y123-10 | -0,09549 | 2.33               | 4,321                 | 3,887-4,175          |
| Y123-15 | -0,0816  | 2.38               | 4,414                 | 4,123-4,296          |

## Indentation-induced cracking (IIC) model

The IIC model has been developed to describe the RISE behavior. In this model, it is argued that the test load is balanced by the total sample resistance at maximum depth. This resistance consists of four elements; shift, deformation, elastic deformation In this model, the load-independent hardness is calculated by the formula,

$$H_V = K \left(\frac{F^{5/3}}{d^3}\right)^m \tag{10}$$

If the m value obtained from the slope of the  $\ln(F^{5/3}/d^3)$ -lnH<sub>V</sub> graph is smaller than 0.6, it is proved that the material exhibits RISE behavior (Ozturk 2012, Safran 2015, Ozturk 2018).



**Figure 7.** Plot of  $\ln H_V$  against  $\ln(F^{5/3}/d^3)$  according to IIC model for all the samples

When Table 5, which shows the data obtained in IIC model, is examined, it is seen that the value of m is less than 0.6. This supports RISE behavior. It is also seen that the load-independent hardness is in the plateau region. In this case, it is seen that the IIC model is a more useful method to prove the RISE behavior of the samples produced according to other models.

Table 5. Best-fit results of experimental data according to IIC model

| Samples | ${\rm K}~({\rm N}^{(3-5m)/3}~/\mu m^{(2-3m)})$ | m    | HIIC (GPa) | Hv (GPa)    |
|---------|--|------|------------|-------------|
| Undoped | 4,43   | 0,34 | 3,493      | 3,420-3,755 |
| Y123-10 | 4,32   | 0,31 | 3,895      | 3,887-4,175 |
| Y123-15 | 4,34   | 0,32 | 4,041      | 4,123-4,296 |

### 4. CONCLUSION

In the study, Co doped YBCO123 samples are produced by solid state reaction method. XRD, SEM and Vickers microhardness measurements are performed for all samples to investigate structural, morphologic and mechanical properties.

In the XRD results, it was observed that the peaks shifted to the low angle values in direct proportion to the doping ratio. It is predicted that this may be due to the tendency to disrupt the orthorhombic structure. In addition, the decrease in peak intensity with the doping indicates that the crystallinity has deteriorated. From the SEM images it was observed that, surface morphology changed with doping. Increasing of doping ratio increased the porosity of the samples surface.

The mechanical behavior of the samples was examined by microhardness analysis. As a result of the microhardness analysis, it was observed that the sample microhardness increased as the contribution rate increased. In all samples, the microhardness value increased with increasing load so that all samples exhibited RISE behavior. Experimentally obtained microhardness values were examined theoretically. As a result of these investigations, it was concluded that the IIC model is the most appropriate model for explaining RISE behavior.

#### REFERENCES

- Andersen, N., Andersen, J., Börjesson, L., Hadfield, R., Kakihana, M., & McGreevy, R. et al. (1993). Structure and superconductivity in Co doped YBa2Cu3O6+x. Journal Of Alloys And Compounds, 195, 327-330. doi: 10.1016/0925-8388(93)90749-d
- Boudjema, E., Mahtali, M., Chamekh, S., & Taoufik, A. (2010). The effect of Ca co-substitution in (Y1-xCax)Ba2 (Cu0.98Co0.02)3O7- $\delta$  (0  $\leq$  x  $\leq$  0.35) ceramic. Surface And Interface Analysis, 42(6-7), 996-999. doi: 10.1002/sia.3466
- Dihom, M., Shaari, A., Baqiah, H., Al-Hada, N., Kien, C., & Azis, R. et al. (2017). Microstructure and superconducting properties of Ca substituted Y(Ba1–Ca )2Cu3O7–δ ceramics prepared by thermal treatment method. Results In Physics, 7, 407-412. doi: 10.1016/j.rinp.2016.11.067
- Guner, S., Gorur, O., Celik, S., Dogruer, M., Yildirim, G., Varilci, A., & Terzioglu, C. (2019). Effect of zirconium diffusion on the microstructural and superconducting properties of YBa2Cu3O7-6 superconductors.
- Khalid, N., Kechik, M., Baharuddin, N., Kien, C., Baqiah, H., & Yusuf, N. et al. (2018). Impact of carbon nanotubes addition on transport and superconducting properties of YBa 2 Cu 3 O 7-δ ceramics. Ceramics International, 44(8), 9568-9573. doi: 10.1016/j.ceramint.2018.02.178
- Kurnaz, S., Çakır, B., & Aydıner, A. (2017). The effect of growth temperature on the irreversibility line of MPMG YBCO bulk with Y 2 O 3 layer. Cryogenics, 85, 51-57. doi: 10.1016/j.cryogenics.2017.05.010
- Liu, L., Dong, C., Zhang, J., & Li, J. (2002). The microstructure study of Co-doped YBCO system. Physica C: Superconductivity, 377(3), 348-356. doi: 10.1016/s0921-4534(01)01286-2
- Matskevich, N., & Wolf, T. (2014). Thermochemical investigation of YBa2Cu3O7–δ superconductor doped by lutetium. Journal Of Alloys And Compounds, 614, 415-419. doi: 10.1016/j.jallcom.2014.06.125
- Ozturk, O., Erdem, M., Asikuzun, E., Yildiz, O., Yildirim, G., Varilci, A., & Terzioglu, C. (2012). Investigation of indentation size effect (ISE) and micro-mechanical properties of Lu added Bi2Sr2CaCu2Oy ceramic superconductors. Journal Of Materials Science: Materials In Electronics, 24(1), 230-238. doi: 10.1007/s10854-012-0722-9
- Ozturk, O., Asikuzun, E., Tasci, A.T. et al. J Mater Sci: Mater Electron (2018) 29: 3957. https://doi.org/10.1007/s10854-017-8336-x
- Pimentel, J., Buitrago, D., Supelano, I., Parra Vargas, C., Mesquita, F., & Pureur, P. (2014). Synthesis and Characterization of the Superconductors Y3Ba5Cu8−x Fe x O18(0.0597 ≤ x ≤ 0.1255). Journal Of Superconductivity And Novel Magnetism, 28(2), 509-512. doi: 10.1007/s10948-014-2742-6
- Ramli, A., Shaari, A., Baqiah, H., Kean, C., Kechik, M., & Talib, Z. (2016). Role of Nd2O3 nanoparticles addition on microstructural and superconducting properties of YBa2Cu3O7-δ ceramics. Journal Of Rare Earths, 34(9), 895-900. doi: 10.1016/s1002-0721(16)60112-6
- Safran, S., Kılıç, A., Kılıçarslan, E., Ozturk, H., Alp, M., Asikuzun, E., & Ozturk, O. (2015). Mechanical, microstructural and magnetic properties of the bulk BSCCO superconductor prepared by two different methods. Journal Of Materials Science: Materials In Electronics, 26(4), 2622-2628. doi: 10.1007/s10854-015-2733-9
- Supadanaison, R., Panklang, T., Wanichayanan, C., Kaewkao, A., Nilkamjon, T., & Udomsamuthirun, P. et al. (2018). Determination of Cu 2+ and Cu 3+ by titration in Y134 and Y145 superconductor. Materials Today: Proceedings, 5(7), 14896-14900. doi: 10.1016/j.matpr.2018.04.026
- Yilmaz, M., & Dogan, O. (2012). Structural and superconducting properties in Y0.6Gd0.4Ba2(Nb)Cu3O7-y cuprates doped with niobium. Journal Of Rare Earths, 30(3), 241-244. doi: 10.1016/s1002-0721(12)60031-3



# The Structural, Morphological and Superconducting Properties of Co Doped Polycrystalline YBCO123 Superconductor Produced by Sol-Gel Method

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Abstract: The aim of this study is to investigate the effect of the Co nanoparticle on YBCO-123 system. Co nanoparticles are substituted with Cu in the YBa2Cu3-xCoxO7- $\delta$  high temperature superconductor (HTS) produced by sol-gel method (x=0.10 ve 0.15). Methanol, acetic acid were used as solvent and triethanolamine as thickener. The gelled solution formed as bulk after the calcination process and sintered at 930°C for 24 hours. The structural, superconductivity and mechanical properties of the produced samples were investigated. X-ray diffraction (XRD) analysis was performed to examine the crystal structure and to calculate the lattice parameters. The surface morphology of the samples was analyzed by scanning electron microscopy (SEM). Transition temperature of the samples has been determined by four probe resistance measurements (R-T). Critical transition temperature (Tc) have found to decrease with the increasing concentration of Co in compound.

Keywords: YBCO, Y123, HTS, sol-gel method, Co doping

# **1. INTRODUCTION**

Since the discovery of the YBCO compound system, a large number of researchers have conducted studies on the investigation and development of superconducting properties. A number of articles have been published to investigate the superconductivity and structural properties of YBCO samples. Many groups have made adding or substitution for developing the properties of the Y123 (Hamrita2014, Slimani2014, Josephson1962, Metin2016).

Gupta et al. (Gupta2010) made Co doping to Y123 system; lattice parameters increases with the contribution and critical temperature value has been increased with the increasing of the Co doping. Roa et al. (Roa2010) published their results by applying the stress-strain test on Y123 superconducting ceramic structures. Dadras et al. (Dadras2016) showed that in the Y123 phase produced by sol-gel method, critical temperature and transition temperature values was increased by Au doping. In another study of the same group (Dadras2015), the carbon nantube doped YBCO123 phase was shown decrease in the critical temperature value. Wang et al. (Wang2015) concluded that 930°C is the most suitable temperature that gives the highest critical temperature value for the sol-gel method as a result of the calcination process at different temperatures. Jin et al. (Jin2017) showed that with an increase in calcination temperature the oxygen content rises. They also showed that increased oxygen content caused a decrease in the resistivity.

In this study, we produced samples with sol-gel method and substituted Co nanoparticle with Cu. We aimed to investigate the structural, morphological and superconducting properties of the Co doped Y123 system.

# 2. MATERIALS AND METHODS

Samples prepared by using powders; (CH3CO2)3Y (Yttrium(III) acetate hydrate, %99.9, Sigma-Aldrich), (CH3COO)2Ba (Barium acetate, %99.99, Sigma-Aldrich), Cu(CO2CH3)2 (Copper(II) acetate, %99.99, Sigma-Aldrich) and Co-nanoparticle (Cobalt powder-325 mesh %99.5, Alfa Aesar). Methanol anhydrous (CH3OH 99.8%, Sigma-Aldrich) and acetic acid (Acetic acid glacial, CH3CO2H,  $\geq$ 99.85%, Sigma-Aldrich) were used as solvent and triethanolamine (Tris(2-hydroxyethyl)amine  $\geq$ 99.0, Sigma-Aldrich) as a thickener. The required weighted powders were

ground in agate mortar for 1 hour and put into alumina boats. Samples calcined at 850°C for 24 hours in Protherm PLT-120/5 model ash furnace. Calcination process repeated 3 times and after each calcination powders grounded in agate mortar for 1 hour. After the third calcination process, grounded powders form into 13mm diameter and 2mm thickness bulk by using cold press. The tablet samples were placed in Protherm PTF-15/45/450 model tube furnace on the alumina crucible and sintering process applied. Samples sintered at 930°C for 24 hours by heating rate of 5°C/min and then cooled to 500°C in 60 minutes. The samples were kept in the oxygen atmosphere for 5 hours at 500°C. Samples are named as C0, C10 and C15 in accordance with doping ratio.

The phase diagnosis and structural analysis were investigated by Bruker D8 Advance X-ray diffractometer in the range of  $2\theta$ =3-90° with CuK $\alpha$ . Surface morphology and elemental analysis investigated by FEI Quanta Feg 250 model scanning electron microscopy. Electrical resistance measurements performed by conventional four probe method in cryogenic system.

# 3. RESULTS AND DISCUSSION

The XRD patterns of all samples produced in  $Y_1Ba_2Cu_{3-x}Co_xO_{7-\delta}$  general formula with the x = 0, 0.10 and 0.15 doping ratios are given in Figure 1. When XRD patterns were examined, it was seen that peaks indicated by miller indices were peaks of Y123 phase. The fact that Co peaks were not found in XRD patterns and this showed that Co atoms entered into orthorhombic Y123 structure by doping process. The displacement of Co+3 (ionic radius 0.745Å) with Cu+2 (ionic radius 0.73Å) played an important role in the introduction of Co atoms as a result of the doping process. Co atoms entering the structure caused some changes in peak intensity but did not corrupt orthorhombic structure.



Figure 1. XRD patterns and miller indices of produced samples

Table 1. Lattice parameters and particle size values

| Sample | Grain Size (Å) | a (Å) | b (Å) | c (Å) |
|--------|----------------|-------|-------|-------|
| C0     | 28.21          | 3.82  | 3.86  | 11.61 |
| C10    | 32.90          | 3.83  | 3.87  | 11.74 |
| C15    | 32,99          | 3.85  | 3.86  | 11.75 |

Since Y123 is an orthorhombic structure; d the distance between the planes and the data such as the a, b, and c parameters were calculated by formula,

$$\frac{1}{d^2} = \frac{h^2}{a^2} + \frac{k^2}{b^2} + \frac{l^2}{c^2} \tag{1}$$

using the indices h, k, l from the XRD. Grain size, D, calculated using formulas

$$D = 0.941\lambda/B\cos\theta \tag{2}$$

$$B^2 = B_s^2 - B_m^2 (3)$$

here  $B_s$  is the full width half of maximum (FWHM) and  $B_m$  is 0,000007 [9].

In the literature, the lattice parameters of Y123 structure are given as a=3.82Å, b=3.89Å and c=11.7Å respectively. It is seen that the lattice parameters of Y123 for superconducting samples with Co doping produced using sol-gel method are in accordance with the literature. As seen from Table 1, the grain size increased with Co doping ratio.



Figure 2. SEM images of samples produced by Sol-Gel method (a; C0, b; C10, c; C15)

SEM images was carried out to determine the particle spacing, particle boundaries and particle sizes of Co doped samples produced by sol-gel method. Figure 2 contains the SEM images of the samples. Large images in Figure 2 are taken at 2500 magnification, while internal pictures are taken at 10000 magnification.

From the SEM images of Co doped samples produced by the Sol-gel method, it is seen that the particle size increases with the increase of Co doping ratio and the samples getting less porosity by changing the grain boundaries. The change in particle size corresponds to the particle size values calculated by XRD results.

Electrical resistance measurements based on temperature inform us about the behavior of materials at low temperatures. To determine the critical temperature value of a superconductor material, temperature dependent electrical resistance measurement is performed. In these measurements,  $T_c^{onset}$  indicating that the critical temperature transition has begun and the  $T_c^{offset}$  offset values indicating that the critical temperature transition is over are obtained.

The results of electrical resistivity measurement of Co additive samples produced by sol-gel method are given in Figure 3. As a result of the electrical resistance measurements, the samples produced by sol-gel method showed superconductivity. In this way, the result is that Co atoms entering the structure do not break the orthorhombic structure. The critical temperature value of the materials was decreased with the increasing of the doping ratio. The YBCO structure will hold an excess of oxygen due to the fact that the Co doping creates an free electron in the system. The oxygen content in the YBCO superconductors determines the critical temperature. Therefore, each electron added to the system maintains oxygen in the structure. As a result, the amount of oxygen in the structure increases and the critical temperature values decrease. The values obtained from the electrical resistance measurement are given in Table 2.



Figure 3. Electrical resistance measurement of produced samples

CO sample has the highest transition temperature and the lowest superconductivity transition value ( $\Delta T_c$ ). While the transition temperature decreased with the increasing of the doping ratio, the superconductor transition value has increased. The room temperature resistance also increased with the doping ratio. In the graph of electrical resistance vs temperature, the binary transition is observed. This is thought to be due to the impurity phases which cause poor bonding in the grain boundaries.

Table 2. Electrical resistance measurement values

| G      | Critical T      | $\Delta T_{\mathcal{C}}(\mathbf{K})$ |                                |  |
|--------|-----------------|--------------------------------------|--------------------------------|--|
| Sample | $T_{C}^{onset}$ | $T_{C}^{offset}$                     | $T_C^{onset}$ - $T_C^{offset}$ |  |
| C0     | 92.46           | 90.00                                | 2.46                           |  |
| C10    | 89.05           | 72.71                                | 16.34                          |  |
| C15    | 72.34           | 43.49                                | 28.85                          |  |

## 4. CONCLUSION

We have investigated the structural, morphological and superconducting properties of Co nanoparticle doped YBCO123 samples produced by sol-gel method. From XRD results it obvious that crystal structure of YBCO123 have effected by Co doping ratio. The peak intensities of the doped samples decreased by increasing of doping ratio. Grain size and c lattice parameter of doped samples have increased with increasing of doping ratio. Particle sizes and particle surface have imaged by SEM and it is clear that Co doping have changed the surface morphology of YBCO123 phase. The important parameter for superconductors is to know critical temperature. Critical temperature values have obtained by electrical resistance measurements. Increasing of Co nanoparticle doping ratio have caused to decreasing of critical temperature values. However, the critical temperature transition value increased by doping ratio.

#### REFERENCES

Dadras, S., Soufiabadi, E., (2015). The mechanical properties investigation of the CNT doped YBCO high temperature superconductor with ANSYS finite element software. Iranian Journal of Science and Technology, Transaction A: Science, 39(A4), 543-549

- Dadras, S., Gharehgazloo, Z., (2016). Effect of Au nano-particles doping on polycrystalline YBCO high temperature superconductor. Physica B, 492, 45–49
- Gupta, S., Yadav, R.S., Lalla, N.P., Verma, G.D., Das, B., (2010). Microstructural and Superconducting Properties of YBa2Cu3xCoxO7-δ System. Integrated Ferroelectrics, 116:1, 68-81
- Hamrita, A., Slimani, Y., Ben Salem, M.K., Hannachi, E., Bessais, L., Ben Azzouz, F., Ben Salem, M., (2014). Superconducting properties of polycrystalline YBa2Cu3O7 – d prepared by sintering of ball-milled precursor powder. Ceramics International, 40, pp. 1461–1470
- Jin, F., Zhang, H., Wang, W., Liu, X., Chen, Q., (2017). Improvement in structure and superconductivity of YBa2Cu3O6+δ ceramics superconductors by optimizing sintering processing. Journal of Rare Earths, 35(1), 85-89
- Josephson, B.D. (1962). Possible new effects in superconductive tunneling. Physics Letters, 1, 7, pp. 251-253
- Metin, T., Tepe, M., (2016). The effect of Ag doping on the superconducting properties of Y3Ba5Cu8-xAgxO18-δ ceramics. Journal of Superconductivity and Novel Magnetism., 30(4), pp. 1083-1087
- Roa, J.J., Jiménez-Piqué, E., Capdevila, X.G., Segarra, M., (2010). Nanoindentation with spherical tips of single crystals of YBCO textured by the Bridgman technique: Determination of indentation stress-strain curves. Journal of the European Ceramic Society, 30, 1477–1482
- Slimani, Y., Hannachi, E., Ben Salem, M.K., Hamrita, A., Varilci, A., Dachraoui, W., Ben Salem, M., Ben Azzouz, F., (2014). Comparative study of nano-sized particles CoFe2O4 effects on superconducting properties of Y-123 and Y-358. Physica B: Condensed Matter, 450, pp. 7–15
- Wang, W., Chen, Q., Cui, Q., Ma, J., Zhang, H., (2015). Preparation of c-axis oriented YBa2Cu3O7 polycrystalline ceramics by solgel method. Physica C, 511, 1–3



# Analysis of Biodiesel Production from Hazelnut Oil: The Effect of Ultrasonic Bath Application on the Physical Properties

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Abstract: Energy that the world needs is faced with the danger of exhaustion due to the decrease in oil stocks. In addition, the current fossil fuel resources used to meet the rapidly increasing energy demand cause climate changes due to the  $CO_2$  gas released. This situation increases the interest for alternative and sustainable energy sources that are not based on petroleum every day. Therefore, biomass energy obtained from vegetable and animal origin materials for alternative energy sources, is very important. For this aim, one of the most important sources to be obtained is biodiesel obtained from vegetable oils. Biodiesel can be inferior in performance as an environmentally friendly fuel in terms of sulphur and carbon emissions compared to petroleum-based diesel. For this reason, it is used after mixed with the diesel at certain ratios. Therefore, in this study, it was aimed to determine the effect of ultrasonic sound waves on the physical properties of biodiesel produced from hazelnut oil to bring the properties of biodiesel to the natural range.

After measured the acidity of hazelnut oil, biodiesel was produced from naturel vegetable oil by transesterification reaction using methyl alcohol at 55°C by NaOH-catalyzed. The process was carried out in different exposure times and sound waves within the ultrasonic water bath. Thus, changes in the physical properties of biodiesel that belongs to changing of chemical structure have been tried to be determined. For this aim, biodiesel samples were determinated using by FTIR, cloud and pour point, sulphur concentration etc. qualitative and quantitative analysis.

As a result, FTIR analyses showed that fatty acid-methyl esters formed by high efficiency from the hazelnut oil. It was found that there was a partial change in physical parameters such as flow-cloud point, and these values were found to be between sonicated biodiesel and diesel-related data. The ultrasonic bath method used had a good effect on shortening the long fat chains, which is the most important problem in biodiesel production, as well as chemical methods. It also has a high application potential as a faster method since it does not include the step of removing chemicals.

Keywords: Biodiesel, hazelnut oil, ultrasonic waves, transesterification reaction, sustainable energy

# **1. INTRODUCTION**

As a result of the rapid increase in the energy demand of the world's population, interest in new alternative energy sources is increasing day by day. Therefore, instead of the existing energy sources used as fuel; studies are carried out to meet energy needs from alternative, sustainable and environmental sources that are not based on petroleum. One of the most important sources in which alternative energy can be provided in this area is to obtain biodiesel from plant-based substances by transesterification reactions. Although biodiesel is an environmentalist fuel in terms of sulfur and carbon emission compared to petroleum-based diesel, it may be insufficient in performance. For this reason, it is used by mixing with diesel in certain proportions at today. In this study, hazelnut oil was preferred due to the fact that its physical properties such as viscosity, density and thermal value are better than diesel fuel.

Keskin et al., who obtained biodiesel from hazelnut oil, investigated torque, power and specific fuel consumption according to the diesel engine. They found that the engine performance values of the biodiesel obtained from the hazelnut are close to the diesel fuel values (Kesgin, 2006). Oğuz established a plant to convert hazelnut oil into biodiesel, and the fuel properties of biodiesel that he obtained were investigated in a diesel engine. As a result of the research, the fuel properties of biodiesel compared with diesel fuel and stated that there is no difference (Oguz, 2004).Sanches and his friends examined the chemical structure of hazelnut oil, the equivalent carbon number of some chemical compounds in hazelnut oil levels and triacylglycerol carbon numbers, compared with the results obtained with other vegetable oils

samples in his study (Sanches, 2003). In one study, biodiesel with NaOH-catalyzed methyl alcohol at  $55 \,^{\circ}$  C was obtained by taking into account the test work and fuel characteristics of diesel engines for the determination of the most suitable oils in biodiesel extraction from vegetable oils (Leung et al., 2010; Ma and Hanne, 1999).

# 2. MATERIAL AND METHODS

In this study, the effect of ultrasonic sound waves on the physical properties of biodiesel has been investigated by obtaining biodiesel by using the base catalyzed transesterification reaction from hazelnut vegetable oil, which is known to be used in biodiesel extraction. For this purpose, biodiesel was obtained from vegetable oils with NaOH catalyzed methyl alcohol at 55°C.

It was conducted that process is applied in a water bath that emits ultrasonic sound waves, different exposure times such as 60, 120 and 180 minutes. In this study, biodiesel that obtained from hazelnut oil was carried out following the following steps.

# **Determination of the Amount of Free Fatty Acid**

Free fatty acid refers to free fatty acids in the structure of the oil, which are not bound to the triglyceride structure. Excess of free fatty acids is not desirable because of the increasing effect of saponification in biodiesel production process and should not exceed 1% in general (EN14104). For this purpose, the amount of free fatty acid in oil has been determined primarily. The amount of free fatty acid was determined by titration of the oil sample with 0.1 N KOH prepared in ethanol. In the study, because of the results remained below 1%, that is, 0.33%, at the free fatty acidity determination, biodiesel production was started as the next step.

## **Biodiesel Production**

Therefore, during transesterification, glycerine is removed from the vegetable oil, the fat becomes finer, and its viscosity decreases. For this purpose, biodiesel samples obtained from vegetable origin oils were applied systematically which the following operations and the conformity of the biodiesel, which obtained according to the standards was investigated. The stages of transesterification of hazelnut oil are given in Figure 2.1. For transesterification, methyl alcohol with methyl alcohol that molar mass and stearic barrier was the lowest were used.



Figure 2.1. Transesterification reaction equation (NREL, 2004)

Sodium hydroxide that is used as catalyst was dissolved in alcohol. After the alcohol /catalyst mixture was filled into the closed reaction vessel, vegetable oil was added. The system was completely closed to the atmosphere to prevent the loss of alcohol. The reaction mixture was kept at a certain temperature to accelerate the reaction and the reaction was carried out. After the reaction was completed, two main products glycerin and biodiesel were obtained. Since the density of the glycerine phase was much greater than that of the biodiesel phase, these two phases could be separated by gravity and the glycerin phase was easily removed from the bottom of the precipitation vessel (Figure 2.2) (URL-1)



Figure 2.2. Transesterification process of hazelnut oil: a) Transesterification process b, c) Biodiesel-glycerol separation process.

# **Sonication Process**

In this study, after obtaining biodiesel, physical and chemical changes in the structure of biodiesel were investigated by using ultrasonic bath. After 1000 ml hazelnut oil biodiesel has been placed into closed glass containers, 100% ultrasonic sound waves applied at 80 Hz (Elmasonic P). Biodiesel samples were given code according the exposure time in the the sonication bath (Table-2.1).

# Table 2.1. Biodiesel sample codes

| F0: Ultrasonic untreated nut oil           |
|--|
| F1: 60 minutes ultrasonic treated nut oil  |
| F2: 120 minutes ultrasonic treated nut oil |
| F3: 180 minutes ultrasonic treated nut oil |

# **FTIR Analysis**

Biodiesel samples synthesized from hazelnut oil were tested with Bruker Optics ALPHA model FT-IR spectrometer and specific peaks were determined for biodiesel. In addition, the element content and sulphur percentage in biodiesel samples were determined by XRF.

# **Cloud and Pour Point Analysis**

The cloud and pour point was measured in order to determine the cold flow characteristics of the fuels using by Koehler Cloud and Pour Point Equipment (NRELL 2009). According to TS EN 590: 2013 + A1 standards and EN 23015 test method, the cloud and pour point analysis had been done using by of the diesel can vary from -10 to -34 <sup>o</sup>C according to the climate types.

# 3. RESULTS AND DISCUSSION

# **Elemental Analysis and Sulphur Contents**

In the elemental analysis of biodiesel samples obtained from hazelnut oil by XRF device, the amount of sulfur was examined and the values obtained as expected were around 0%. The sulfur content of the obtained biodiesel was compared with the petrochemical diesel sample. Non-sulfur biodiesel samples are important for elimination of environmental pollution factors. While the sulfur percentage of diesel fuel was 0%, the sulfur percentage of hazelnut oil biodiesel was found as 0.2579%.

# **Results of FTIR Analysis**

According to FTIR analysis, it was determined that the transesterification reaction with hazelnut oil was carried out successfully and the esterification rate was similar in biodiesel samples obtained from hazelnut oil. The absorption peaks and their properties for biodiesel samples are given in Table 3.1.

| Number of waves (cm-1) | Group Vibrations   | Vibration Type                 | Absorption |
|------------------------|--------------------|--------------------------------|------------|
|                        |                    |                                | intensity  |
| 3005                   | =С-Н               | Stress                         | Middle     |
| 2922                   | -CH <sub>2</sub>   | Asymmetric stress vibration    | Severe     |
| 2853                   | -CH <sub>2</sub>   | Symmetrical stress vibration   | Severe     |
| 1741                   | -C=O               | Stress (typical for esters)    | Severe     |
| 1500-900               | Fingerprint zone   |                                |            |
| 1460, 1461,            | -CH <sub>2</sub>   | Scissor Type strain            | Middle     |
| 1446-1435              | -CH <sub>3</sub>   | Asymmetric stress (specific    | Middle     |
|                        |                    | to biodiesel)                  |            |
| 1360, 1361             | -CH <sub>2</sub>   |                                | Middle     |
|                        |                    | Bending vibration              |            |
| 1196                   | -O-CH <sub>3</sub> | Stress (typical for biodiesel) | Middle     |
| 1168                   | C-O-C              | Anti-symmetrical strain        | Middle     |
|                        |                    | vibration                      |            |
| 1016                   | C-O-C              | Symmetrical stress vibration   | Middle     |
| 722                    | -CH <sub>2</sub>   | Planar rocking                 | Middle     |

Table 3.1. Absorption peaks obtained by FT-IR spectroscopy and properties

FT-IR spectrum of the biodiesel sample synthesized from hazelnut oil also is given in Figure 3.1. Although there was no change in the peak values in the fingerprint zone of the biodiesel sample prepared from hazelnut oil, there was no significant change in the intensity of some peaks. An increase in peak intensity showing the ester bonds in 1741 cm-1 in F0 sample was observed with increasing sonication time. Also, it is also understood from the broadband in the region of 3310 cm<sup>-1</sup> that alcohol groups or unsaturated bonds are formed in the structure F0 of the biodiesel sample.



Figure 3.1 Hazelnut Oil (F0) FT-IR spectra

In general, shear has been observed in specific peak regions of the ultrasonic treated and unexposed samples. It is noted that there is an increase in the peak intensity.

# **Cloud and Pour Point Results**

According to TS EN 590: 2013 + A1 standards and EN 23015-test method, the flow-cloud point of the diesel can vary from -10 to -34 <sup>o</sup>C according to the climate types. Table.3.2 gives the flow-cloud point and freezing point values of the biodiesel samples of the hazelnut oils treated at 0, 60, 120 and 180 minutes in the ultrasonic bath. Keskin and friends in his study (Keskin, 2006), hazelnut biodiesel pour point is -15 <sup>o</sup>C; in this study, these values were determined as -11 <sup>o</sup>C for hazelnut biodiesel not treated under the same conditions. The results obtained were approximately overlapping.

 Tablo 3.2 Cloud and pour point changes of the biodiesel samples treated in 0, 60, 120 and 180 minutes in ultrasonic bath

| Biodiesel Sample | Cloud and Pour Point | Freezing Point |
|------------------|----------------------|----------------|
| F0               | -11                  | -12            |
| F1               | -11                  | -13            |
| F2               | -11                  | -14            |
| F3               | (-11 ve -12)         | -13            |

### 4. CONCLUSION

Since the amount of free fatty acid for hazelnut oil is below 1%, it is not necessary to take any additional action to reduce free fatty acid. This shows that the refining of oils is well done.

In all biodiesel samples, peaks of various intensity were detected in FT-IR spectrum around 1741 cm-1. This peak value shows the ester bonds in the structure. Although an increase in peak intensity was observed with increasing sonication time, it was found that the peak value approached untreated samples which were indexed with "0" when the sonication time was 3 hours.. This has shown that the duration of the sonication process stimulates structural changes differently. Short-chain paraffin groups were formed up to a certain exposure time, whereas these paraffin groups were found to form alcohol or phenolic structures undergoing new transformations with increased sonication time. It was found that peaks compatible with the ester structure of biodiesel were observed in the FT-IR spectrum and no peaks were observed due to impurities or non-removal of alcohol. FTIR analysis showed that the rate of fatty acid-methyl esters formed changed with the sonication process increased the production rate of fatty acid-methyl ester and increased the biodiesel efficiency and the physical properties were closer to the diesel. In the biodiesel sample obtained from hazelnut oil, it was observed that the exposure time of the cloud and pour point was not effective and the cloud and pour point remained constant. In freezing point analysis, the best value was obtained in F2, 120 min treated hazelnut sample.

In this respect, the sonication of the samples after biodiesel production by transesterification process has shortened the chain lengths of the fatty acid-methyl esters, which supported the approaching of the physical properties to the diesel

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#### REFERENCES

- Keskin, A., Aydın, K., 2006. Fındık Yağı biodizel üretimi ve dizel motorlarda alternatif yakıt olarak kullanımınnın araştırılması, Gazi Ü.Müh. Mim.Fak.Dergisi, 21, 367-372.
- Leung, D. Y. C., Wu, X., Leung M. K. H., 2010. A review on biodiesel production using catalyzed transesterification. Applied Energy, 87, 1083-1095.
- Ma, F., Hanna M.A., 1999. Biodiesel production: A review. Bioresour Technol. 70: 1-15
- NREL, 2004. Subcontractor Report Biodiesel Production Technology, Etitor; NREL National Renewable Energy Laboratory, (NREL/SR-510-36244).
- NREL, 2009. Biodiesel Handling and Use Guide, 4th Ed., National Renewable Energy Laboratory NREL/TP-540-43672 Revised, USA.,1-53
- Oğuz H., 2004. Tarım kesiminde yaygın olarak kullanılan dizel motorlarında fındık yağı biyodizelinin yakıt olarak kullanım imkanlarının incelenmesi. Doktara Tezi, Konya,
- Sanchez, P.L.B., Camacho, M.L., Aparicio, R.A., 2003. Comprehensive study of hazelnut oil composition with comparisons to other vegetable oils, particulary olive oil" Europaean Food Research and Technology, 218,1319.
- URL-1: http://www.solarenerji.com/biodizel-tesis-installation



# Determination of Radiogenic Heat Production in Soil Samples from Industrial City Karabük, Turkey

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Abstract: The kinetic energy of the emitted particles (alpha and beta) and the gamma-ray associated with the different radioactive decay processes is absorbed in the soils and rocks finally transformed into heat. The most important source of the earth's internal heat is the heat produced by the decay of long-lived radionuclides such as uranium, thorium, and potassium. The heat produced by radioactive decay in soils and rocks is of fundamental importance in understanding the thermal history of the earth and interpreting the continental heat flux data. In the study, the concentrations of uranium, thorium, and potassium in soil samples collected from industrial city Karabük located Western Black Sea of Turkey were analyzed using an energy dispersive X-ray fluorescence (EDXRF) spectrometry. Radiogenic heat contributions of soil samples were determined based on the soil's U (in mg/kg), Th (in mg/kg) and K (in %) concentrations and soil density. The values of radiogenic heat production (RHP) estimated for soil samples varied from 0.3 to 1.4  $\mu$ W/m3 with an average of 0.5  $\mu$ W/m3. The results indicate that the contribution and rate of heat production of 238U, 232Th and 40K in the samples vary significantly with geological locations, with 238U as the major element which predominates in heat production.

Keywords: Long-lived radionuclides, radiogenic heat production, soil, Karabük.

# **1. INTRODUCTION**

The rheology (Rheology, a special science that examines the behavior of bodies under load, deformation and time factors) of solids is a function of temperature. For this reason, the thermal structure of the earth's crust must be known in order to understand the mechanical behavior of the earth's crust. The thermal structure of the Earth's crust determines its tectonics and its seismicity. Therefore, the temperature is one of the most important physical parameters in the place. Despite this, our knowledge of the thermal structure of the ground is not enough. Difficult to apply and expensive temperature measurements are usually made at shallow depths and are under the influence of groundwater movements, climatic conditions, topography, and local geology.

Because of these effects on the measurements, it is very difficult to obtain reliable information about the temperature distributions in the deep part of the earth's crust. For this reason, various mathematical relations are tried to be calculated by using the model studies to calculate the temperatures in the deeper parts of the earth. These relations provide the geotherm graphics which give the variation of the temperature with depth. There are two components of the existing heat energy in the earth's crust. These are the heat energy transmitted from the mantle to the shell and the heat energy released by the decomposition of the radioactive elements in the shell. Many researchers agree that radioactive decay is the most effective source of thermal evolution during the period from the formation of the earth to the present (Göktürkler, 2002; Turcotte and Schubert, 1982; Mayhew, 1982)

The radioactive elements that cause this are divided into two groups as short and long half-life isotopes. The short half-life radioactive isotopes are 26Al, 26Cl and 60Fe and long half-life isotopes are 235U, 238U, 232Th and 40K.

The short half-life isotopes were effective in the production of heat energy in the early stages of the formation of the earth. Those with a long half-life are responsible for the heat energy produced in the process from the first periods to the present. As is known, with the decay of the radioactive elements, the emission of  $\alpha$  and  $\beta$  particles in the environment and electromagnetic wave propagation occur. Both the absorption of electromagnetic waves by other atoms in the environment and the collision of other atoms in the environment with the  $\alpha$  and  $\beta$  particles results in an increase in the kinetic energies

of these atoms, thus increasing the average kinetic energy of the medium and hence the temperature, called radiogenic heat generation (Göktürkler, 2002).

In this study, measurements of activity concentration of 238U, 232Th, and 40K were done using EDXRF spectroscopy. These activity concentrations were used to determine the contribution of each of the elements to radiogenic heat production.

## 2. MATERIALS AND METHODS

For analyses of soil samples, 18 sampling sites away from roads, cultivated locations, buildings, and trees were chosen from Karabük province. The samples, each about 1 kg in weight, were ground, homogenized and sieved to about 100 mesh screen. The samples were then placed for drying at 110 °C for 24 h to ensure that moisture is completely removed. The analysis survey was conducted using energy dispersive x-ray fluorescence (Spectro Xepos, Ametek). The target changer, with up to 8 polarization and secondary targets, offers many different excitation conditions ensuring the optimum determination of all elements from 11Na to 92U. A spectral resolution of less than 155 eV at Mn  $K\gamma$  is achieved. The sample chamber is equipped with a sample spinner for 40 mm sample cups (Anonymous, 2015; Kurnaz et al., 2016). The samples to be measured were passed through the crusher and sieved. Approximately 3 gr of powdered samples were used for the measurements. For the analysis of the powder samples, the "powder method" registered on the device was used.

If the density of the soil is  $\rho$  and the concentrations in Uranium (CU), thorium (CTh) and potassium (CK) are known, its radiogenic heat generation rate A can be determined using the values given (Shittu et al., 2016; Bubu and Ononugbo, 2017).

$$A[\mu Wm^{-3}] = 10^{-5*}\rho[kgm^{-3}]^*(9.52^*C_U[ppm] + 2.56^*C_{Th}[ppm] + 3.48^*C_K[\%])$$
(1)

#### **3. RESULTS AND DISCUSSION**

The concentrations of the radionuclides were presented in Table 1. Radiogenic heat production was calculated from the  $^{238}$ U,  $^{232}$ Th and  $^{40}$ K concentration using the formula (1) and presented in Table 1.

|             | 40                     | 40 222 228                 |               | Contribution                                       |       |       |       |
|-------------|------------------------|----------------------------|---------------|--|-------|-------|-------|
| Sample code | <sup>40</sup> K<br>(%) | <sup>232</sup> Th<br>(ppm) | 238U<br>(ppm) | Radiogenic heat production<br>(µW/m <sup>3</sup> ) | U     | Th    | K     |
| <b>S1</b>   | 0.8018                 | 2.2                        | 1.8           | 0.3  | 0.213 | 0.069 | 0.034 |
| <b>S2</b>   | 0.4802                 | 11.5                       | 8.9           | 1.4  | 1.039 | 0.360 | 0.020 |
| <b>S</b> 3  | 1.472                  | 9                          | 1.3           | 0.5  | 0.150 | 0.282 | 0.063 |
| <b>S4</b>   | 1.342                  | 6.7                        | 1.1           | 0.4  | 0.129 | 0.210 | 0.057 |
| <b>S</b> 5  | 1.733                  | 7.9                        | 2.2           | 0.6  | 0.256 | 0.248 | 0.074 |
| <b>S6</b>   | 1.079                  | 12                         | 4.0           | 0.9  | 0.470 | 0.376 | 0.046 |
| <b>S7</b>   | 0.4623                 | 2.8                        | 3.3           | 0.5  | 0.380 | 0.088 | 0.020 |
| <b>S8</b>   | 1.053                  | 5.6                        | 2.3           | 0.5  | 0.273 | 0.175 | 0.045 |
| <b>S9</b>   | 1.34                   | 5.6                        | 2.9           | 0.6  | 0.333 | 0.175 | 0.057 |
| S10         | 0.8018                 | 2.2                        | 2.3           | 0.4  | 0.266 | 0.069 | 0.034 |
| S11         | 0.1892                 | 3.8                        | 3.7           | 0.6  | 0.426 | 0.119 | 0.008 |
| S12         | 0.83                   | 5.3                        | 2.5           | 0.5  | 0.288 | 0.166 | 0.035 |
| S13         | 0.5533                 | 3                          | 2.6           | 0.4  | 0.302 | 0.094 | 0.024 |
| S14         | 0.366                  | 2.2                        | 1.5           | 0.3  | 0.172 | 0.069 | 0.016 |
| S15         | 0.2795                 | 1.8                        | 1.6           | 0.3  | 0.189 | 0.056 | 0.012 |
| S16         | 0.5061                 | 4.2                        | 4.9           | 0.7  | 0.573 | 0.132 | 0.022 |
| S17         | 0.167                  | 1.6                        | 2.3           | 0.3  | 0.263 | 0.050 | 0.007 |

Table 1. Concentrations of <sup>238</sup>U, <sup>232</sup>Th, <sup>40</sup>K and radiogenic heat production in soil samples of Karabük

| <b>S18</b> | 0.6759 | 4.5 | 5.1 | 0.8 | 0.592 | 0.141 | 0.029 |
|------------|--------|-----|-----|-----|-------|-------|-------|
|            |        |     | Av  | 0.5 | 0.351 | 0.160 | 0.033 |
|            |        |     | Min | 0.3 | 0.129 | 0.050 | 0.007 |
|            |        |     | Max | 1.4 | 1.039 | 0.376 | 0.074 |

The activity concentrations of <sup>238</sup>U ranged from 1.1 to 8.9 ppm. The lowest <sup>238</sup>U activity concentration was measured in sample S4 and the highest <sup>238</sup>U activity concentration was measured in sample S2. The activity concentrations of <sup>232</sup>Th ranged from 1.6 to 12 ppm. The highest <sup>232</sup>Th activity concentration was measured in sample S6 and the lowest <sup>232</sup>Th activity concentration was measured in sample S6 and the lowest <sup>232</sup>Th activity concentration was measured in sample S6 and the lowest <sup>232</sup>Th activity concentration was measured in sample S17. The values of radiogenic heat production (RHP) estimated for soil samples varied from 0.3 to 1.4  $\mu$ W/m<sup>3</sup> with an average of 0.5  $\mu$ W/m<sup>3</sup>. The results indicate that the contribution and rate of heat production of <sup>238</sup>U, <sup>232</sup>Th and <sup>40</sup>K in the samples vary significantly with geological locations. Considering the distribution of the radiogenic heat production elements contribution, <sup>238</sup>U is the major element which predominates in heat production (Table 1).

# 4. CONCLUSION

In the study area and samples, we cannot make a comparative analysis since radiological heat production is not calculated before. Our data provide a data infrastructure for geologists in radiogenic heat generation. This is important for tectonic movements, hydrocarbon, and geothermal energy in terms of resource evaluation in the working areas.

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## REFERENCES

- Anonymous (2015). Spectro Xepos Brochure, Ametek, http://www.spectro.com/products/xrf-x-ray-fluorescence-spectrometer/edxrf-spectrometer-xepos.
- Bubu A & Ononugbo C P (2017). Radiogenic Heat Production Due to Natural Radionuclides in the Sediments of Bonny River, Nigeria. Journal of Scientific Research & Reports, 17(6): 1-9.
- Göktürkler G (2002). Yerbilimlerinde Isı Transferi Modellemesi: Kararlı-Hal Kondüktif Isı İletimi. Deü Mühendislik Fakültesi Fen Ve Mühendislik Dergisi, 4(3): 67-80.
- Kurnaz A, Turhan Ş, Gezelge M, Hançerlioğulları A & Çetiner M A (2016). Elemental Composition of Soils Mixed with the Grape Molasses. Turkish Journal of Agriculture - Food Science and Technology, 4(9): 748-751.
- Mayhew M A (1982). Application of Satellite Magnatic Anomaly Data to Curie Isoterm Mapping. J. Geophys. Res., 87: 4846-4854.
- Shittu A, Hankouraou S & Ziyadat H (2016). Determination of radioactivity concentration and annual ommitted effective dose in drinking water collected from Local Borehole in Gombe, Nigeria. Sch. J. Phys. Math. Stat. 3(2): 56-65.
- Turcotte D.L., Schubert G. (1982): "Geodynamics -Application of continuum physics to geological problems", New York, Wiley, pp. 705.



# **Evaluation of Drilled Hole Quality in 2011-T6 Aluminium Alloy**

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Abstract: In this study, effect of cutting speed and feed rate on surface roughness and axial deviation hole quality values were investigated in the drilling process of AA 2011 Aluminum alloys. In the experimental studies, extrusion product AA 2011-T6 Aluminum alloy bars were used. 14 mm diameter uncoated carbide drilling tool was selected as cutting tool. Direct drilling method was used without any pauses in the hole drilling with the CNC milling machine. The cutting parameters; Cutting speeds (160, 200, 240 and 280 m/min), feed rates between 0.05-0.20 mm/rev were selected for hole quality. Cutting fluid was used in the drilling process. Three experiments were conducted for each parameters and mean values of 3 results were taken. Surface roughness (Ra) values of the samples were measured. Hole center axis measurements and hole axis deviation measurements were determined with 3 dimensional measurement system. The results clearly showed that the cutting speed and the feed rate have directly effect on surface roughness and the radial axial deviation values.

Keywords: 2011 Aluminum alloy, Drilling, Surface roughness, Axial deviation, Cutting parameters.

# **1. INTRODUCTION**

In 2xxx series aluminum alloys, copper (Cu) is used for main alloying element. If it is desired magnesium and other alloying elements (Mn, Si, Fe) can be added inside. 2xxx series aluminum alloys have high strength besides of good machinability, bad corrosion resistance and welding quality. 2xxx series are lighter than 7xxx series aluminum alloys so that preferred in aircraft outer panels and defense industry. 2011 aluminum alloy has A-rated machinability because of content of Bi and Pb addition inside. Bi and Pb have very low solubility in liquid and solid aluminum. Therefore, Bi and Pb precipitate in the grain boundaries as elemental metal or Bi-Pb alloys. 2011 aluminum alloys are used in complex parts which are produced with machining in aerospace, trailer and automotive industry. Extrusion process is mostly used for rod and bar profiles as raw materials before machining or forging operations for 2011 aluminum alloy (Koch and Antrekowitsch, 2011).

Drilling operations from machining methods comprise approximately 33 - 40% of the whole machining process (Bayraktar et al. 2017; Chaanthini et al, 2017). The hole surface quality results, such as surface roughness, axial misalignment, circularity, burr formation and hole size, vary depending on the process parameters in drilling operations (Bayraktar et al. 2017; Chaanthini et al, 2017; Abdelhafeez et al, 2015; Kurt et al, 2008; Giasin et al, 2016). Hole surface quality affects assembly and working conditions of work pieces. In precision mounting systems, the importance of hole axis improves the quality of the assembly, and installation with incorrect axes can be problem in the upcoming operations and mechanical properties of parts. The deflection in the hole axis affects the stress distribution and causes the parts exposed to high stresses. This high stress can decrease the expected life of the products. Bore surface roughness increases abrasive wear. In the design of plurality of parts, the aperture of the hole axis, perpendicularity of the hole axis, surface roughness of the hole, misalignment of the hole, circular tolerances are the most important parameters of product to get desired properties from the products where it will be used (Giasin et al, 2016).

In the literature research, some studies have been found about drilling of 2xxx series aluminum alloys. 2024 Aluminum alloy have similar properties with 2011 alloy. Kurt et al. studied with 30, 45 and 60 m/min parameters with Ø10 mm cutting tool. Cutting speeds, 0.15, 0.20, 0.25 mm/rev were selected and the results indicate that the surface roughness

changes with increasing cutting speed. Increase feed rate cause worse surface roughness. In the same study, the axial misalignment values increased with the increase in cutting speed and the increase in the axial offset (Kurt et al, 2008).

Giasin et al. studied with 100, 300, 600, 900 mm/min parameters with  $\emptyset$  6 mm cutting tool. Feed rates were selected as 1000, 3000, 6000, 9000 rev parameters in experiments and worked on the effects of cutting parameters on surface roughness and circularity (Giasin et al, 2016).

In this study, Extruded 2011 aluminum alloy rectangular bars that has better machinability than 2024 T6 Aluminum alloy was used in the drilling experiments to determine the effect of cutting speed and feed rate on surface roughness quality of the hole wall. 14 mm diameter uncoated carbide drilling tool used for CNC operations.

# 2. MATERIALS AND METHODS

### Material, Cutting Tool, Machine and Coolant

In the experimental studies, as test piece material, 2011 T6 aluminum alloy was used. Test piece has rectangular prism geometry with dimensions of 100 mm x 68 mm x 31 mm. At first 2011 aluminum billet was produced with direct chill casting method with 355 mm diameter. Billets were homogenized in batch furnace. 55 MN extrusion press were used to produce rectangular shape bard profile in ASAŞ Aluminum Plant. After extrusion process, solution heat treatment, quench and aging processes were conducted in NABERTHERM brand C40-version 5.02 heat treatment furnace. The solution was made. Water was quenched at room temperature. Aging was carried out under T6 conditions. Chemical composition of alloy is given in Table1 and mechanical properties of T6 condition of 2011 are given in Table2.

| Material | Si    | Fe    | Cu    | Mn    | Mg    | Zn    | Ni     |
|----------|-------|-------|-------|-------|-------|-------|--------|
| Wateria  | 0.248 | 0.243 | 5.843 | 0.014 | 0.001 | 0.004 | 0.008  |
| A A 2011 | Cr    | Pb    | Sn    | Ti    | Sb    | Other | Al     |
| AA 2011  | 0.011 | 0.202 | 0.008 | 0.032 | 0.012 | 0.343 | 93.031 |

 Table 2. Mechanical properties of test material

| Material | $R_p (N/mm^2)$ | $R_m (N/mm^2)$ | Elongation (%) | Hardness (HB) |
|----------|----------------|----------------|----------------|---------------|
| AA 2011  | 263            | 415            | 15             | 124           |

Cutting tool that is available for DIN 6589 (similar to DIN 1897), with two cutting blades, 3xD (3 times depth drill diameter), complete carbide, uncoated, cutting angle 118°, burr angle (helix angle) 30° and diameter of Ø14 mm drilling tool was selected for experiments. Measurements of the drilling hole were measured with QUİCKCHECK.MPC brand equipment. Drilling tool diameter, helix angle, cutting angle and helical groove depth were measured 13.986 mm, 30.10°, 7.31° and 4.45 mm.

According to the manufacturer's recommendations, the cutting data were evaluated for external factors such as the stability of tools and tool holders, material and machine type.

MAZAK VTC300-II which has 1610 mm x-axis, 510 mm y-axis and 610 mm z-axis working area and fully automatic CNC milling machine were used in drilling operations. Testing materials were fasten up with hydraulic clamp.

OUMETA brand water mixable ALUMET AL100 was used as a coolant and lubricant. ALUMET AL100 is a product specially developed for the machining of aluminum and aluminum alloys.

## Surface Roughness and Axial Misalignment Measurements

Roughness measurements were done with AHRSURFSD26 roughness meter which has MAHRSURF M400 main module processor and Bluetooth communication system. Ra value was used for surface roughness assessment. Measurements were taken from origin and end of holes in two positions. 17.5 mm scanning area was used in the measurements.

For measurement of hole axial misalignment, LK INTEGRA brand Metris with a 3D measurement system model that has precision of  $0.15\mu m$  was used in ambient environment of  $20^{\circ}C \pm 1^{\circ}C$ . The system software of device is CAMIO 4.5 version. In order to prevent measurement error, data were collected from 7 different distances such as beginning of hole

and 0.3, 5.0, 10.0, 15.0, 20.0, 25.0 and 30.0 mm away from the hole. Measurements were taken from 4 different points for each hole sample. Verification and cross control measurements were also taken to verify the results. Schematic representation of the axial misalignment evaluation is presented in Figure 1.



Figure 1. Schematic representation of Radial axial deflection assessment

# **Experimental Procedure**

Before the starting of experiments 2011 T6 aluminum alloy bars were brought into the machine environment one day before to ensure the test material's and environment's temperature same. Brand new clamping pliers were used in the machine to inhibit possible negative effects of oscillations and vibrations. Vibration and instability were prevented by using hydraulic clamp.

The machine manufacturers and industries' most used cutting speed and feed rate were accepted as reference in experimental procedure. Recommended parameters of drill manufacturer for  $V_c$  is 180 m/min and  $f_n$  is 0.15 mm/rev. Experimental study parameters are given in Table 3.

In experiments cutting speed and feed rate parameters were investigated from minimum to maximum rates. Vibration control was made for MAZAK VTC300-II and Spindle vibration was checked before operations. It is assumed that there is no vibration from the machine for the parameters selected as 12,000 rpm, and 0.71 mm/s in the range of 10 - 1000 Hz.

| Material | Cutting Tool                   | Feed rate $F_n$ ( mm/rev) | Cutting speed V <sub>c</sub> ( m/min) |
|----------|--------------------------------|---------------------------|---------------------------------------|
| Al 2011  |                                |                           | 160                                   |
|          | Ø14mm 118°<br>Uncoated carbide | hatwaan 0.05.0.20         | 200                                   |
|          |                                | between 0.03-0.20         | 240                                   |
|          |                                |                           | 280                                   |

Table 3. Experimental study parameters (4x4 = 16 variables)

# 3. RESULTS AND DISCUSSION

# Effect of Feed Rate and Cutting Speed on Surface Roughness

The effect of feed rate on surface roughness are given in Figure 2. The figure shows that increase in feed rate increases surface roughness as well. At the same time, as the cutting speed increases, the difference between feed rate and the surface roughness values increases. The results obtained from the experiments are generally consistent with the results of 2024 Aluminum alloy studies in literature (Abdelhafeez et al, 2015; Giasin et al, 2016). In these experiments, approximately same results were obtained from the origin and end of the drilled holes.

The effect of cutting speed on surface roughness are given in Figure 3. The surface roughness values increase for all feed rates with increasing cutting speed. Figure 3 shows the values of A, B and C mm/rev feed rates for each cutting speed, surface roughness results are very close to each other but for D mm/rev feed rate result showed surface roughness are higher than others. C mm/rev feed rate is the most suitable value for the best surface roughness as it is suggested by the manufacturer of the drilling tool. Surface roughness value  $R_a$  changes 83% for Cutting speed from 160 m/min to 280 m/min the lowest surface roughness was measured C mm/rev feed rate and 160 m/min cutting speed parameters.



Figure 2. Feed rate - surface roughness relation graphics



Figure 3. Cutting speed - surface roughness relation graphics

Figure 2 and 3 showed that surface roughness it is observed that the machinability of AA2011 is high.

# Effect of feed rate and cutting speed on axial misalignment

The friction caused by drilling, cutting force and torque cause misalignment in the drilled hole axis. The main reasons for this misalignment are type of material, microstructure and mechanical properties.

During the experiments, the deviation values for different feed rate and cutting speeds were measured in the x and y coordinate planes. The evaluation of the results was made by combining the deviation values in the x and y coordinate planes and the results are given in Figure 4, 5, 6, 7, 8, 9, 10 and 11.

The graphical results of the feed rate parameter showing the effect of axial offset are given in Figure 4, 5, 6, and 7. The effect of feed rate on the axial misalignment was different for each cutting speed conditions. For A mm/rev progress was similar to each other at all cutting speeds and resulted in smaller axial misalignment values. For B mm/rev (without 200 m/min) progress was observed that smaller misalignments than A mm/ revolution for C mm/rev and D mm/rev. The results of the axial misalignment of the feeds are the highest values at all cutting speeds. The data of Figure 4 shows that as a general expression without B mm/rev, when feed rate increase, the axial misalignment values increases also. Increasing in the feed rate increases the forces and moments on the tool, and this situation causes increasing of axial misalignment values, so the results are available for this study.

The smallest axial misalignment values were measured at 240 m/min when the entire hole depth is evaluated, cutting speed and B mm/rev feed rate obtained under parameter condition.



Figure 4. V<sub>c</sub>=160m/min feed rate – Radial axis deviation graphics



Figure 5. V<sub>c</sub>=200m/min feed rate – Radial axis deviation graphics



Figure 6. V<sub>c</sub>=240m/min feed rate – Radial axis deviation graphics



Figure 7. Vc=280m/min feed rate - Radial Axis deviation graphics

Effect of cutting speed on axial misalignment is given in Figure 8, 9, 10 and 11. It is observed that increase of cutting speed decreases axial misalignment values when it is considered all of the drilled hole depth from all Figure. These results for AA2024 are higher in literature studies (Abdelhafeez et al, 2015).



Figure 8. fn=A mm/rev, Cutting speed – Radial axis deviation graphics



Figure 9. fn=B mmm/rev, Cutting speed – Radial axis deviation graphics



Figure 10. fn=C mm/rev, Cutting speed – Radial axis deviation graphics



Figure 11. fn=D mm/rev, Cutting speed – Radial axis deviation graphics

For 280 m/min cutting speed, very low axial misalignment results were obtained up to 25 mm of drilled hole. When axial misalignment results are compared with 2024 and 2011 aluminum alloys, it is determined that 2011 aluminum alloy's

axial misalignment results are lower than AA2024. This result confirms that the 2011 aluminum alloy has a better machinability than 2024 aluminum alloy.

# 4. CONCLUSION

In this study, 2011 aluminum alloy bar profile that produced firstly with direct chill casting method in billet form. After that extruded to rectangular geometry bar profile. 2011 bar profiles are drilled with uncoated carbide drilling tool in CNC milling machine. Effects of cutting speed and feed rate parameters on surface roughness were investigated. The results are given below;

- 1. 2011 aluminum alloy has good machinability according to results obtained from this study.
- 2. When feed rate increases in CNC machine, surface roughness value increases as well.
- 3. Cutting speed is more effective parameter than feed rate on surface roughness. It is observed that increasing cutting speed at all feed rates effects directly surface roughness.

4. The smallest surface roughness results were measured with C mm/rev feed rate and 160 m/min cutting speed parameters.

5. The effect of feed rate on axial misalignment is different for each cutting speed conditions. It can be said that as a general expression, the increase in feed rate can increase axial misalignment too.

6. It is observed that increase of cutting speed decreases axial misalignment values when it is considered all of the drilled hole depth.

7. The smallest axial misalignment value is obtained by 240 m/min cutting speed and B mm/rev feed rate parameters.

8. In axial misalignment inspections, 280 m/min cutting speed has very low axial misalignment results up to a depth of 25 mm for all feed rates.

9. Both of Drilling depth and axial misalignment values are linear increasing.

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#### REFERENCES

- Abdelhafeez A.M., Soo S.L., Aspinwall D.K., Dowson A. and Arnold D., 2015. Burr formation and hole quality when drilling titanium and aluminum alloys. Procedia CIRP 37: 230 235
- Bayraktar Ş., Siyambaş Y. and Turgut Y, 2017. Delik delme prosesi: bir araştırma. Sakarya University Journal of Science 21(2): 120-130
- Chaanthini M. K., Shanmugam M. and Sanjivi A., 2017. Study on Hole Quality in Drilling AA 6063 Plate under CryogenicPre-Cooling Environment. Materials Today: Proceedings 4: 7476–7483
- Giasin K., Hodzic A., Phadnis V. and Soberanis S.A., 2016. Assessment of cutting forces and hole quality in drilling Al2024 aluminum alloy: experimental and finite element study. The International Journal of Advanced Manufacturing Technology 87: 2041–2061
- Koch S. and Antrekowitsch H., 2011. Investigations of lead-free aluminum alloys for machining. World of Metallurgy ERZMETALL 64(1): 26-30
- Kurt M., Kaynak Y. and Bağcı E., 2008. Evaluation of drilled hole quality in Al 2024 alloy. The International Journal of Advanced Manufacturing Technology 37: 1051-1060



# Investigation of structural and micromechanical properties of Cu/Fe doped ZnO semiconductor nano particles prepared using sol-gel method

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Abstract: In this study, Cu/Fe doped ZnO based semiconducting nano particles are produced by the sol-gel technique that is the most widely used method for preparing nano size materials. The effect of doping on structural and micromechanical properties of the ZnO semiconducting nano particles are investigated in detail. For comparison, undoped sample is prepared in the same conditions. X-ray diffraction analysis (XRD) is used to determine phase analysis and lattice parameters of the semiconducting nano materials and Vickers microhardness measurements are made for mechanical properties. In addition, different microhardness models in the literature are performed for all samples.

Keywords: ZnO, sol-gel, doping, Vickers

# **1. INTRODUCTION**

Technology and science are an integral part of life for humans. In this context, it is necessary and sufficient to use science to implement developing and advancing technology. As people's living conditions diversified and increased, new requirements emerged and these requirements brought with them new technological pursuits. In most of the major breakthroughs that are performed, the importance of materials used in developed products has been very great and this case has led to the emergence of material technology. As the requirements for technological devices increased day by day, the expected features of the used materials increased as well. As a result of all these requirements, scientists have improved their study at the point of producing technological materials that are constantly evolving and able to respond to the needs of time. Therefore, every new material and technique developed are of great importance [1-3].

The mechanical behavior of the materials reflects the deformation or resistance of material against the force or load applied. Depending on the force applied to the material, two types of deformation occur; i) elastic (non-permanent change) and ii) plastic (permanent change). The elastic deformation refers to the change of distance between the atoms of material, on which the force is applied, without any separation between the adjacent atoms. When the applied force is removed, then the object gains its previous form. If the stress created on the material by the force applied exceeds beyond the elasticity limit of the material, then the permanent deformation called plastic deformation occurs [4,5].

The most common and effective experiment carried out in order to determine the mechanical properties of the materials is the hardness measurement. The main reasons for this are that the procedure is simple and that it damages the sample less than the other procedures do. The other advantage is that the hardness of a material is directly proportionate to the other mechanical properties. For instance; the tensile strength of materials is directly proportionate to the hardness. Thus, it is possible to have an idea about the strength of the material by measuring the hardness.

In the present study, the Cu/Fe doped ZnO-based semiconducting samples are produced using sol-gel method and their structural and mechanical properties are analyzed. The mechanical properties of the samples are determined using Vickers microhardness, and the surface morphology, crystal structure characteristics, and lattice parameters are determined using XRD measurements.

## 2. MATERIAL AND METHODS

Zn<sub>0.95-x</sub>Cu<sub>0.05</sub>Fe<sub>x</sub>O system is prepared as polycrystalline nanoparticles with various compositions (0.01<x<0.05) using sol–gel techniques. Zinc acetate dihydrate and Cu and Fe acetate are used as precursors. Methanol and acetic acid are used as a solvent to preparing a homogenous solution. After weighing at appropriate rates, the precursors and solvents are stirred using the heater magnetic stirrer at 60°C for 8 h until a transparent solution is obtained. Then Zn<sub>0.95-x</sub>Cu<sub>0.05</sub>Fe<sub>x</sub>O powders are preheated at 300°C for 30 min in air and grounded. Grounded nanoparticles are pressed under 4 tons for 5 min into disk shaped compacts with a thickness of 2 mm and a diameter of 10 mm. Finally, samples are annealed at 600°C for 30 min. The change of the crystal structure, phase impurity, texturing, grain size and lattice parameters of the Bi-2212 superconductors is examined by XRD measurements performed by a Bruker D8 Advance model X-ray diffractometer. Moreover, a SHIMADZU HVM-2 model digital microhardness tester is used to determine the mechanical properties.

## **3. RESULTS AND DISCUSSION**

### Results

*XRD Measurements:* X-ray diffraction method was used to determine the crystal structures and lattice parameters of the materials. XRD measurements were performed by Bruker D8 Advance X-ray powder diffractometer using  $CuK_{\alpha}$  ( $\lambda = 1,541$ Å) radiation in the range of  $3^{0} \le 20 \le 60^{0}$  at a scan speed of 4°/min. The lattice parameters can easily be analyzed by the diffraction patterns belonging to a semiconducting material. Figure 1 shows XRD patterns for the materials. The characteristic peaks identifying with ZnO phase are displayed by (h k l) miller indices on the XRD graphs. The hegzagonal ZnO stuctures have been observed to form for all doping ratios as well as no Cu and Fe phases have been detected. This is an indication that the additive materials (Cu and Fe) have entered the structure [6]. Lattice parameters *a* and *c* and grain size values are summarized in Table 1. With rising doping ratio of Fe, *a* and *c* lattice parameters are partially decreased. This is an expected result because the ionic radius of Fe<sup>+3</sup>(0.64Å) is greater than the ionic radius of Zn<sup>+2</sup> (0.74Å).



Figure 1. XRD patterns of all samples

Table 1. Lattice parameters a and c values

| a (Å) | c (Å)  |   |
|-------|--|---|
| 3.73  | 5.20   |   |
| 3.24  | 5.18   |   |
| 3.25  | 5.21   |   |
| 3.24  | 5.18   |   |
|       | <b>a (Å)</b><br>3.73<br>3.24<br>3.25<br>3.24 | a (Å)       c (Å)         3.73       5.20         3.24       5.18         3.25       5.21         3.24       5.18 |

*Mechanical Characterization of Materials:* Microhardness measurements are carried using digital microhardness tester to determine the effect of Cu/Fe doping on the mechanical properties of ZnO based sem, conducting system at room temperature. In this study, Vickers microhardness test is used for measurements. The Vickers microhardness values  $(H_v)$  of different applied loadings in the range of 0.245-2.940 N for 10s can be calculated using the Eqn.1. This process is repeated 7 times and average value of hardness is found.

$$H_{\nu} = 1854.4 \,\frac{F}{d^2} \tag{1}$$

It can be seen from Fig. 2 that the microhardness values increase with increasing applied load. This result indicates that all materials exhibit *RISE* (Reverse Indentation Size Effect) behavior. In addition, the microhardness values decrease with increasing Fe doping.



**Figure 2.** Change of Hv versus applied load F **Table 2.** Microhardness values of all samples

| Samples     | F<br>(N) | Hv<br>(GPa) | Plateau Region<br>(GPa) | E<br>(GPa) | Y<br>(GPa) |
|-------------|----------|-------------|-------------------------|------------|------------|
| Undoped ZnO | 0.245    | 0.651       | 1.290                   | 53.35      | 0.217      |
|             | 0.490    | 0.962       | -                       | 78.84      | 0.320      |
|             | 0.980    | 1.140       | 1.520                   | 93.43      | 0.380      |
|             | 1.960    | 1.290       |                         | 105.73     | 0.430      |
|             | 2.940    | 1.320       |                         | 108.19     | 0.441      |

| Zn94Cu05Fe01O | 0.245 | 0.570 | 1.085 | 46.71 | 0.192 |
|---------------|-------|-------|-------|-------|-------|
|               | 0.490 | 0.734 | -     | 60.16 | 0.244 |
|               | 0.980 | 0.950 | 1.102 | 77.86 | 0.316 |
|               | 1.960 | 1.085 |       | 88.93 | 0.361 |
|               | 2.940 | 1.102 |       | 90.32 | 0.367 |
| Zn93Cu05Fe03O | 0.245 | 0.412 | 1.010 | 33.76 | 0.137 |
|               | 0.490 | 0.676 | -     | 55.40 | 0.225 |
|               | 0.980 | 0.924 | 1.099 | 75.73 | 0.308 |
|               | 1.960 | 1.010 |       | 82.78 | 0.336 |
|               | 2.940 | 1.099 |       | 90.07 | 0.366 |
| Zn92Cu05Fe05O | 0.245 | 0.339 | 0.577 | 27.78 | 0.113 |
|               | 0.490 | 0.479 | -     | 39.26 | 0.159 |
|               | 0.980 | 0.532 | 0.379 | 43.60 | 0.177 |
|               | 1.960 | 0.577 |       | 47.29 | 0.192 |
|               | 2.940 | 0.579 |       | 47.45 | 0.193 |
|               |       |       |       |       |       |

# Analyses and Modelling of Microhardness

# Meyer's Law

The slope of *InF-Ind* graph (Fig 3) gives the value of  $n_k$  and we can see that, it is greater than 2 for all samples. It is confirmed that the microhardness increases with applied load and the material exhibits RISE behavior [7-9] (Table 3).



Figure 3. Variation of applied load *lnF* with diagonal length *lnd* for the samplesTable 3. Best-fit results of experimental data according to Meyer's law

| Samples            | Meyer number   | InK    |
|--------------------|----------------|--------|
|                    | n <sub>k</sub> | (GPa)  |
| <b>Undoped ZnO</b> | 2.71           | -10.12 |
| Zn94Cu05Fe01O      | 2.73           | -10.46 |
| Zn93Cu05Fe030      | 3.09           | -12.00 |
| Zn92Cu05Fe05O      | 2.47           | -10.18 |

#### Proportional Sample Resistance (PSR) Model

The resulting data are summarized in Table 4. As can be seen from this table, the values of  $\gamma$  are negative, an expected result for materials that exhibit RISE behaviour. This situation confirms that only plastic deformations occur in these samples which shows *RISE* behavior. There is no elastic deformation. Also, plateau values of samples are far from microhardness values calculated by PSR model [10,11].



Figure 4. Plot of *F*/*d* versus *d* for the samples

Table 4. Best-fit result of experimental data according to PSR model

| Samples       | a x10 <sup>-3</sup><br>(N) | β x10 <sup>-5</sup><br>(N/μm) | H <sub>PRS</sub><br>(GPa) | Hv<br>(GPa) |
|---------------|----------------------------|-------------------------------|---------------------------|-------------|
| Undoped ZnO   | -14.07                     | 94.58                         | 1.753                     | 1.290-1.320 |
| Zn94Cu05Fe01O | -13.60                     | 80.19                         | 1.487                     | 1.085-1.102 |
| Zn93Cu05Fe03O | -19.35                     | 87.50                         | 1.622                     | 1.010-1.099 |
| Zn92Cu05Fe05O | -6.12                      | 38.20                         | 0.708                     | 0.577-0.579 |

#### Elastic/Plastic Deformation (EPD) Model

As can be seen from Table 5, values of  $d_e$  are negative for undoped and Cu/Fe doped samples. There is only plastic deformation for all samples. Elastic deformation is not observed. Microhardness values, calculated by EPD model, are far from plateau region where microhardness is not changed with applied load. As a result, it is clearly observed that the EPD model is not useful to determine the microhardness of the samples [12,13].



**Figure 5**. Plots of square root applied loads versus diagonal length for the samples **Table 5**. Best-fit results of experimental data according to *EPD* model

| Samples       | A2 <sup>1/2</sup> | de (µm) | HEDP  | Hv          |
|---------------|-------------------|---------|-------|-------------|
|               | (GPa)             |         | (GPa) | (GPa)       |
| Undoped ZnO   | 0.0317            | -0.29   | 1.863 | 1.290-1.320 |
| Zn94Cu05Fe01O | 0.0292            | -0.31   | 1.581 | 1.085-1.102 |
| Zn93Cu05Fe03O | 0.0313            | -0.47   | 1.816 | 1.010-1.099 |
| Zn92Cu05Fe05O | 0.0198            | -0.19   | 0.727 | 0.577-0.579 |

# Hays-Kendall (HK) Approach

In Table 6, the values of  $W_{HK}$ ,  $A_{1HK}$  and  $H_{HK}$  values are given. The values of  $W_{HK}$  are negative for all samples. Microhardness values, that are calculated according to the HK model, are outside the plateau region. Therefore, this model is not sufficient for determination of the hardness [14].



Figure 6. Applied load versus the square of the impression semi-diagonal length for the samples

| Samples       | Анкх10-5 | Wнк   | Ннк   | Hv          |
|---------------|----------|-------|-------|-------------|
|               | (GPa)    | (N)   | (GPa) | (GPa)       |
| Undoped ZnO   | 78.13    | -0.26 | 1.448 | 1.290-1.320 |
| Zn94Cu05Fe01O | 65.76    | -0.28 | 1.219 | 1.085-1.102 |
| Zn93Cu05Fe03O | 67.93    | -0.43 | 1.259 | 1.010-1.099 |
| Zn92Cu05Fe05O | 33.17    | -0.16 | 0.615 | 0.577-0.579 |

Table 6. Best-fit results of experimental data according to HK model

# Indentation-Induced Cracking (IIC) Model

Fig. 7 shows the results of the application of the indentation-induced cracking model to the data from the four different samples shown in Fig. 2. Despite the significant differences in microhardness values, as well as the varying trends of applied load, it is seen that all of the results fall on a single curve. This confirms that the *RISE* is directly associated with the indentation-induced specimen cracking, it is confirmed that the *RISE* occurs because of indentation-induced cracking [15].



**Figure 7**. Variation of  $In(H_v)$  with  $In(F^{5/3}/d^3)$  according to IIC model for all samples **Table 7**. Best-fit results of experimental data according to *IIC* model

| Samples       | m    | InK<br>(N <sup>(3-5m)/3</sup> /µm <sup>(2-3m)</sup> ) | HIIC<br>(GPa) | Hv<br>(GPa) |
|---------------|------|---|---------------|-------------|
| Undoped ZnO   | 0.48 | 5.48  | 1.302         | 1.290-1.320 |
| Zn94Cu05Fe01O | 0.49 | 5.61  | 1.096         | 1.085-1.102 |
| Zn93Cu05Fe03O | 0.49 | 5.53  | 1.100         | 1.010-1.099 |
| Zn92Cu05Fe05O | 0.52 | 5.81  | 0.575         | 0.577-0.579 |

#### Discussion

When the obtained results are investigated, all samples have hegzagonal structure. With rising doping ratio of Fe, *a* and *c* lattice parameters are partially decreased. Vickers microhardness values ( $H_v$ ) decrease with *Cu* and *Fe* doping. The microhardness values increase with increasing applied load. This result indicates that all materials exhibit *RISE*. If a plastic material is requested for industrial applications, %0.05 *Cu and Fe* doped *ZnO* bulk material is the most appropriate material among all samples (microhardness value is between 0.577-0.579 GPa).

## **4. CONCLUSION**

In this study, the effects of Cu and Fe doping on the micromechanical and structural properties of ZnO system are investigated. Obtained results are listed below.

- The hegzagonal ZnO stuctures have been observed to form for all doping ratios as well as no Cu and Fe phases have been detected. This is an indication that the additive materials (Cu and Fe) have entered the structure.
- Also the Vickers microhardness values computed are noted to increase with increasing the applied load. The dependence of the load showed that *RISE* has an influence on the samples.
- Vickers microhardness values (H<sub>v</sub>) decrease with *Cu and Fe* doping when compared with undoped *ZnO* sample while microhardness values increase with applied load (F).
- As a result, Indentation-Induced Cracking (IIC) model is more suitable to determine the micromechanical properties and *RISE* behavior of *Cu/Fe* doped-ZnO semiconductors.

## REFERENCES

- Serin N, Serin T, Horzum Ş and Çelik Y (2005). Annealing effects on the properties of copper oxide thin films prepared by chemical deposition. Semiconductor Science and Technology, 20:5
- [2] Sato K and Katayama-Yoshida H (2001). Stabilization of Ferromagnetic States by Electron Doping in Fe-, Co- or Ni-Doped ZnO. Japanese Journal of Applied Physica, 40:334-336
- [3] Peng Y, Huo D, He H, Li Y, Li L, Wang H, Oian Z (2012). Characterization of ZnO:Co particles prepared by hydrothermal method for room temperature magnetism. Journal of Magn. Magn. Mater., 324:690-694
- [4] Koralay H, Hicyilmaz O, Cavdar S, Asikuzun E, Tasci T, Ozturk O (2014). Effect of Zn content on microstructure and mechanical performance in Bi<sub>1.8</sub>Sr<sub>2</sub>Ca<sub>2</sub>Cu<sub>3.22-x</sub>Zn<sub>x</sub>O<sub>10+δ</sub> glass ceramic. J. Mat. Sci. Mater. in Elect., 25:3116–3126
- [5] Asikuzun E, Donmez A, Arda L, Cakiroglu O, Ozturk O, Akcan D, Terzioglu C (2015). Structural and mechanical properties of (Co/Mg) co-doped nano ZnO. Cer. Inter., 4:6326-6334
- [6] Arda L, Açıkgöz M, Güngör A (2012). Magnetic and Microstructure Properties of Ni-Doped ZnO Films and Powder by Sol–Gel Process. J. Supercond. Nov. Magn., 25:2701–2705
- [7] Ozturk O, Yildirim G, Asikuzun E, Coskunyurek M, Yilmazlar M, Kilic A. (2013). Change of formation velocity of Bi-2212 superconducting phase with annealing ambient. J. Mat. Sci. Mater. in Elect., 24:4643-54.
- [8] Gong J, Wu J, Guan Z (1999). Examination of the indentation size effect in low-load Vickers hardness testing of ceramics. J. Europ. Cer. Soc., 19:2625-31
- [9] Elmustafa A, Stone D (2003). Nanoindentation and the indentation size effect: Kinetics of deformation and strain gradient plasticity. J. Mech. and Phys. Solids, 51:357-81
- [10] Yilmazlar M, Cetinkara H, Nursoy M, Ozturk O, Terzioglu C (2006). Thermal expansion and Vickers hardness measurements on Bi1.6Pb0.4Sr2Ca2-xSmxCu3Oy superconductors. Physica C: Superconductivity, 442:101-7
- [11] Aydın H, Babanli A, Altintas S P, Asikuzun E, Soylu N, Ozturk O, Dogruer M, Terzioglu C, Yildirim G (2013). Breaking point of the harmony between Gd diffused Bi-2223 slabs with diffusion annealing temperature. J. Mat. Sci. Mater. in Elect., 24:4566-4573
- [12] Upit G, Varchenya S (1966). Microhardness of alkali halide crystals. Phys. Status Solidi (b), 17:831-5
- [13] Bull S, Page T F, Yoffe E (1989). An explanation of the indentation size effect in ceramics. Philosophical Magazine Letters, 59:281-8
- [14] Hays C, Kendall E (1973). An analysis of Knoop microhardness. Metallography, 6:275-82
- [15] Hong L, Bradt R C (1996). The effect of indentation-induced cracking on the apparent microhardness. J. Mater. Sci., 31:1065-1070



# Analytical Studies on Structure-Activity Relationships of Antidiabetic Drugs

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Abstract: Diabetes mellitus (diabetes) is a chronic metabolic disease that requires continuous medical care because the organism does not sufficiently benefit from carbohydrates, fats and proteins due to lack of insulin or defects in its action. The reason for the occurrence of asthenia is that the hormone secretion mechanism in the individual is impaired or the hormone cannot affect the cells. The hormones secreted from the pancreas, a secretory gland with important functions for our body, are involved in regulating blood sugar. The insulin hormone secreted from the beta cells of the pancreas reduces the blood glucose level. Insulin as a hormone allows glucose to be taken into tissues, increases the glycogen level in liver, increases the synthesis of fatty acids and inhibits the breakdown of fatty acids into ketones. Glucose, which cannot be taken by the tissues in case of insufficient insulin, causes water and energy loss by being excreted urine. Diabetes disease should be treated well in the acute or chronic period due to complications. If untreated, hyperlipidemia and cardiovascular system diseases will develop in later periods. The aim of the treatment of diabetes is to keep the blood glucose level between 70-100 mg/dL at fasting time and 140 mg/dL in toughness. For this purpose, the use of oral antidiabetics to replace insulin hormone that cannot be produced in an individual for treatment or to increase the sensitivity of the cells to insulin produced is among the treatment options. The drugs used in the treatment are as Metformin, Glipizide, Glylazide, Rapeglinid, Acarbose, Miglitol. In addition, combined drug therapies are also used. In this study, some physical and chemical properties of the metformin-active drugs used in the treatment of diabetes (closed formulas, molecular weights, colors, melting points, conductivity, pH measurements, solubility tests in polar and apolar solvents, their properties, hardness, microscopic controls, etc.) Elemental analysis using spectroscopic techniques, IR, UV-Vis, magnetic susceptibility, thermogravimetric analysis, such as methods were examined and analyzed in standard solutions and pharmaceutical preparations were carried out.

Keywords: Diabetes, glucose, insulin, spectroscopic techniques, urine


# Synthesis, Structural Characterization and Biological Studies of A New Schiff Base

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**Abstract:** Schiff bases (imines) represent an important class of organic compounds commonly used as synthetic intermediates for covalent organic frameworks and as ligands for metal ions. The development of new organic molecules based on azomethine bond became an important research direction toward novel cytotoxic and cytostatic compounds with a different mechanism of action in comparison with the well-known chemotherapeutic agents. The main issue of this class of compounds is the reversible nature of the C- N bond formation. A special attention was also paid to the Schiff base compounds with antimicrobial, antibacterial, analgesic, antioxidant, antituberclotic, anti-inflammatory, antifungal, antimalarial, antitubercular and anti-urease or antiviral activity. These compounds have often been used as chelating ligands in the field of coordination chemistry for obtaining metal-based supramolecular assemblies, with a large number of potential applications such as catalysis, photochemistry, luminescence, sensing, magnetism, gas storage, gas purification and medicine.

In this study, the ligand of 5-(trifluoromethoxy)salicylidene-4-chloro-*o*-aminophenol was synthesized by the reaction of 5-(trifluoromethoxy)salicylaldehyde and 4-chloro-*o*-aminophenol in the absolute ethanol at 60 °C by the catalyzed of p-toluenesulfonic acid. Later, the complexes of this ligand were prepared with Co(II), Ni(II), Cu(II) and Zn(II) in acetate forms in pure EtOH. Than compounds characterized by spectroscopic techniques. All of the Schiff bases were found to be bidentate ligands involving the imino nitrogen and phenolic oxygen atoms in the complexes and M:L ratio were found to be 1:2 for all the complexes. The structures of ligands and complexes were identified using Elemental Analysis, FT-IR, <sup>1</sup>H-NMR, <sup>13</sup>C-NMR, UV-Vis, Magnetic Susceptibility, SEM, X-Ray and Thermogravimetric Analysis as techniques. The anticancer activities of the prepared complexes were investigated in vitro by XTT method on human colon cancer (HT-29) and breast cancer (MCF-7) cells. The data showed that the complexes exhibited dose-dependent and cell selective anti-proliferative activity on the tested cell lines.

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Keywords: Ligand, o-Aminophenol, Spectroscopic Techniques, Schiff base, XTT Method.



# From Circulated to Liquid Worm Fertilizer

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**Abstract:** The aim of this study is to provide worm manure which is renewed every day by worm manure and exposed to its own water. 30 liter oil bins were cut in half. Holes were drilled in all the covers of the oil drums. Holes were drilled with the help of a drill on the ground part of the three oil containers cut in two. 3 oil bins were drilled in the middle. Basically, the device is formed by joining 2 oil containers to the top of the bottom covered with holes. One hole was drilled on the side of the holes without holes. A pump connected to these holes and taps was started. In order to adjust the pressures of the pipes from the pump, all the thin transparent pipes are equipped with opening and closing taps. 1-2 rows of marble were placed on the top of the top bins. These marbles were covered with a thin tulle and cut to the end of the top container.

Worm fertilizers and foods were mixed in a large container for about 30 minutes using a large mixer. Mixture of tea, corn flour and wheat flour was used. After the mixture became homogeneous, it was poured into oil bins. At the beginning of the experiment, 7.5-8 kg vermicompost and mama were used. In the following days, reinforcement was applied. During the period of 3 months, the ground water accumulated in the lower part of this system was re-empted into the system every day. At the end of 3 months, the last liquid collected on the ground was sent to the analysis and the organic matter contents were examined. According to the results of Selcuk University; Organic matter percentages such as 0,47-0.62.0.46 have been reached. According to the results of this analysis, the worm fertilizer fluid did not reach the desired 5% values of the regulation of the agriculture ministry. Many liquids in this experiment and in the market only have 2 or fewer days of activity. The method of obtaining liquid worm manure should be developed and more original studies should be tried.

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Keywords: Vermicompost tea, Vermiwash, Vermicompost Lachate, Liquit Vermicompost Fertiliezer, Worm wash



# Clustered Regularly Interspaced Short Palindromic Repeats/Crispr Associated Nuclease 9 (Crispr/Cas9) Gene Editing Technology And Clinical Applications

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Abstract: The idea of the genome editing became a current issue when it was understood that DNA double strand breakages trigger DNA repair mechanisms in the past. In order to induce DNA repair mechanism, researchers developed a method that creates targeted DNA double strand breaks by using tools such as; meganucleases (MNs), Zinc Finger Nucleases (ZFNs), Transcription Activator-Like Effector Nucleases (TALENs) and recently Clustered Regularly Interspaced Short Palindromic Repeats/CRISPR Associated Nuclease 9 (CRISPR/Cas9) system. After creation of targeted DNA double strand breaks, cells generally response this DNA damage with one of two DNA repair mechanisms; homology directed repair (HDR) and nonhomologous end-joining (NHEJ). This results in repairment of the targeted sequence, but its efficiency and precision varies with gene editing tool and preferred DNA repair mechanism used in the process. In this context, CRISPR-Cas9 system has advantages over other techniques with its high efficiency, simple usage, low cost and multiplexability that enables to study complex polygenic disorders. The first studies about CRISPR dates back to 1981. A group of scientist discovered 29 base pairs (bps) tandem DNA repeats interspaced with 32 bps spacer sequences in Escherichia coli genome. Similar sequences and Cas gene clusters close to these sequences were determined in different bacteria and archaea in the following years and these sequences were called as CRISPR in 2002. In 2007, it was understood that CRISPR was a response mechanism of the adaptive immune system in prokaryotes that defends bacterial genome against phage and plasmid infections. Afterwards, CRISPR/Cas9 system has been adapted by researchers to create targeted gene editing tool in different organisms. This adapted CRISPR/Cas9 system consists of two major components; the guide RNA (gRNA) and Cas9 nuclease. The gRNA contains a crispr RNA (crRNA, ~20 bp) that can be designed to be complementary to the target DNA site and transactivating crispr RNA (tracrRNA) for Cas9 binding. Target DNA site must be followed by "protospacer-adjacent motif" (PAM) which is necessary for binding of Cas9 nuclease. After the PAM sequence is recognized by Cas9 and complementary base pairing between gRNA and the target DNA site is completed, Cas9 cleaves the DNA to generate a double strand break. Thus by inducing NHEJ or HDR by CRISPR/Cas9 gene editing tool, target DNA sequence can be deleted, replaced or desired sequences can be added. CRISPR/Cas9 technology has been extensively used to generate model organisms in order to elucidate disease etiology or develop new drugs. Although there is no approved clinical application, many clinical trials especially for cancer immunotherapy, hemoglobinopathies and some other genetic diseases are ongoing. These clinical trials are very exciting and their current results hold promise. But many technical hurdles must be overcome to benefit from full potential of CRISPR/Cas9 gene editing tool.

Keywords: CRISPR/Cas9, Gene editing tool, Clinical application, DNA repair mechanisms, DNA double strand breaks.

## **1. INTRODUCTION**

The idea of the genome editing became a current issue when it was understood that DNA double strand breakages trigger DNA repair mechanisms in the past (Hussain et al., 2019). In order to induce DNA repair mechanism, researchers developed a method that creates targeted DNA double strand breaks by using tools such as; meganucleases (MNs), Zinc Finger Nucleases (ZFNs), Transcription Activator-Like Effector Nucleases (TALENs) and recently using Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)/CRISPR Associated Nuclease 9 (CRISPR/Cas9) system. After creation of targeted DNA double strand breaks, cells generally response this DNA damage with one of two DNA repair mechanisms; homology directed repair (HDR) that is error free and nonhomologous end-joining (NHEJ) that is error prone. This results in repairment of the targeted sequence, but its efficiency and precision varies with gene editing tool and preferred DNA repair mechanism used in the process (Rodríguez-Rodríguez et al., 2019; Hussain et al., 2019)

The first studies about CRISPR dates back to 1981. A group of scientist discovered 29 base pairs (bps) tandem DNA repeats interspaced with 32 bps spacer sequences in Escherichia coli genome (Ishino et al., 1987) Similar sequences and Cas gene clusters close to these sequences were determined in different bacteria and archaea in the following years (Mojica et al., 2000) and these sequences were called as CRISPR in 2002 (Jansen et al., 2002; Hussain et al., 2019). In 2007, it was understood that CRISPR was a response mechanism of the adaptive immune system in prokaryotes that defends bacterial genome, against phage and plasmid infections (Barrangou et al., 2007; Hussain et al., 2019). CRISPR/Cas systems generally consist of Cas genes and a CRISPR locus (repetitive sequences (~21–37 bp) interspaced by variable unique spacer sequences). Almost all archaea and about 40% of bacteria have CRIPSR/Cas systems, but they are not found in eukaryotes or viruses (Tang and Fu, 2018; Karimian et al., 2019). CRISPR/Cas systems have been divided into two classes based on the constitution of efector protein and each class has a number of types and subtypes that were classified according to some other characteristics such as, conservation of protein sequence and presence or absence of signature genes (Koonin et al., 2017; Tang and Fu, 2018; Karimian et al., 2019).

In recent years, scientist repurposed these systems as a powerful RNA guided genome editing tools (Jiang and Dudna, 2017). Among these the first and most commonly used one was CRISPR/Cas9 for gene editing. It belongs to type II CRISPR systems (Nishimasu et al., 2014; Tang and Fu, 2018). Nowadays CRISPR/Cas9 also known as 'molecular scissors' is seen as a revolution in molecular biology because of many advantages over other techniques with its high efficiency, simple usage, low cost and multiplex ability that enables to study complex polygenic disorders (Bozok et al., 2017; Memi et al., 2018). Researchers can target almost any genomic sequence by a guide RNA, a component of CRISPR/Cas9 system, and make desired changes in that site. Thus disease-causing mutations can be corrected, function of genes involved in disease development and prognosis can be elucidated or model organism can be generated by targeted mutagenesis (Jiang and Dudna, 2017; Hussain et al., 2019 Zarei et al., 2019). In addition multiplexing ability of this technology allows modification of multiple loci in a single experiment which makes possible understanding of pathological processes developed with the contribution of many genes such as tumor development or polygenic disorders (Jiang and Dudna, 2017). Therefore genome engineering via CRISPR-Cas9 holds enormous promise to treat or cure genetic disorders, including cancer, neurodegenerative diseases, cardiovascular diseases, viral infections and monogenic diseases such as cystic fibrosis, sickle cell anemia, Beta thalassemia and Duchenne muscular dystrophy (Mahmoudiansani et al., 2018; Memi et al., 2018; Foss et al., 2019). In spite of its advantages and potential, there are also a number of challenges including off-target effect and efficient delivery that must be overcome to enable the use of this technology in the clinic (Martinez-Lage et al., 2018; Memi et al., 2018; Karimian et al., 2019).

#### Mechanism of CRISPR/Cas9 System

In prokaryotes, the immune response by CRISPR / Cas system occurs in three steps; adaptation, expression and interference. During the adaptation process, a short DNA fragment from exposed phage or plasmid is inserted into the CRISPR array as a new spacer sequence, thereby a memory is created for this invader that enables the host to prevent future invasion of the same invader. For the selection of the spacer sequence to be taken from the invader, it is understood that some sequences called protospacer adjacent motif (PAM) are mediated. These sequences are critical for the host to distinguish between the self and the non-self. In the expression stage, the formation of pre-crRNA by RNA synthesis from the CRISPR array followed by the expression of the Cas genes and the formation of mature crRNA from pre-crRNA occurs. The interference stage is defined as destruction of the target sequence. In this stage, crRNA is bound to the Cas proteins and this complex interrogate the foreign DNA to find PAM and target sequence. After hybridization between crRNA spacer and a complementary foreign target sequence (protospacer), this complex finally triggers the degradation of the target sequence. Degradation is performed by specific Cas nucleases (Barrangou et al., 2007; Horvarth and Barrangou, 2010; Bozok et al., 2017; Hille and Charpentier, 2017; Hynes et al., 2017).

The adapted CRISPR/Cas9 system as a gene editing tool consists of two major components; the guide RNA (gRNA) and Cas9 nuclease. The gRNA contains a crRNA (~20 bp) that can be designed to be complementary to the target DNA site and transactivating crispr RNA (tracrRNA) for Cas9 binding. Target DNA site must be followed by "protospacer-adjacent motif" (PAM) which is necessary for binding of Cas9 nuclease. After the PAM sequence is recognized by Cas9 and complementary base pairing between gRNA and the target DNA site is completed, Cas9 cleaves the DNA to generate a double strand break (Bozok et al., 2017; Hussain et al., 2019). This generated break is repaired by NHEJ or HDR cellular DNA repair mechanisms. The dominant mechanism used in the cells is NHEJ. NHEJ is a error-prone mechanism, resulting in insertion and / or deletion in the repaired sequence. On the other hand, HDR is activated when a homologous

sequence is present that can be used as a template. In HDR, the accuracy of repair is high as it is carried out by homologous recombination using endogenous or exogenous donor DNA template. Briefly, by inducing NHEJ or HDR by CRISPR/Cas9 gene editing tool, target DNA sequence can be deleted, replaced or desired sequences can be added (Conboy et al., 2018; Tang and Fu, 2018; Rodríguez-Rodríguez et al., 2019).

### **Clinical Applications of CRISPR/Cas9**

Gene editing via CRISPR / Cas9 can be performed ex vivo or in vivo. In the ex vivo approach, the cells taken from the patient are edited in the laboratory and these cells are given to the patient again. On the other hand, gene editing is performed in the patient's body when in vivo approach is used. In this method, CRISPR / Cas9 components or RNA or DNA fragments encoding them are injected into a local body region or circulation by a delivery system (Misra, 2013; Zhan et al., 2019). In vivo delivery and transfection of CRISPR/Cas9 components are generally performed with viral vectors (Mout et al., 2017), while physical or chemical transfection systems can be used in ex vivo approach as well as viral vectors (https://www.synthego.com/guide/how-to-use-crispr/transfection protocols).

In the literature, there are a number of preclinical studies in which gene editing has been performed in various genetic diseases using CRISPR / Cas9. Although there is no currently approved clinical application of CRISPR/Cas9, numerous clinical trials has been initiated due to the promising results of the preclinical studies. Most of the clinical trials aim to develop chimeric antigen receptor T (CAR-T) cells with ex vivo CRISPR / Cas9 applications for use in cancer (Zych immunotherapy al.. 2018; et https://clinicaltrials.gov/ct2/results?cond=&term=CRISPR&cntry=&state=&city=&dist=). However, there are also clinical trials conducted for the treatment of some other genetic diseases such as sickle cell anemia, beta thalassemia, HIV, leber congenital amorosis and malignant hyperthermia (https://clinicaltrials.gov/ct2/results?cond=&term=CRISPR&cntry=&state=&city=&dist=).

One of the first approved clinical trials using CRISPR technique in the US carried out by University of Pennsylvania. In this clinical trial, treatment efficacy of CAR-T cells modified by CRISPR / Cas9 is investigated in the patients with melanoma, sarcoma and multiple myeloma (https://clinicaltrials.gov/ct2/results?cond=&term=CRISPR&cntry=&state=&city=&dist=). While the receptor for tumor specific peptide NY-ESO1 is expressed in these modified CAR-T cells, expression of Programmed Cell Death Protein-1 (PD-1), T cell receptor of  $\alpha$  and  $\beta$  are suppressed (Baylis and Mcleod, 2017). Inhibition of PD-1 increases anti tumor activity of CAR-T cells (Rupp et al., 2017), on the other hand, TCR inhibition decreases auto reactivity (Memi et al., 2018). In a study conducted in China by a group of researchers, PD-1 and TCR genes were silinced by using a similar approach and a clinical trial was started to investigate the effectiveness of mezoteline targeted CAR-T cells on mezoteline positive solid tumors (https://clinicaltrials.gov/ct2/show/results/NCT03545815). It was reported that CD-19 targeted CAR-T cells give promising results in the treatment of relapse or refractor B cell malignancies. But usage of the treatment in certain patients can not be possible. Infants and babies with low blood volume and individuals with insufficient blood volume for CAR-T cell production caused from previous aggressive treatments can be counted among these patients. Since production of CAR-T cells are performed by using otolog T cells specific for an individual, they are not ready to use as universal. Therefore, a group of researcher from China have modified allogenic T cells obtained from healthy unrelated individuals as universal CAR-T cells by silencing some genes via CRISPR technique. Clinical trials are in process on around 80 patients with B cell lenphoma and B cell leukemia for the treatment called UCART019 (Lau and Suh, 2017; https://clinicaltrials.gov/ct2/show/NCT03166878?term=CRISPR&draw=3&rank=13).

Another clinical trial which aims to develop universal CAR-T cells that express CD-20 or CD-22 has been started in China in 2018. Also clinical trials for treatment of different cancers such as esophagus cancer, T cell malignancy and nasopharyngeal carcinoma by using CAR-T cells modified by CRISPR technique are in progress (https://clinicaltrials.gov/ct2/show/NCT03166878?term=CRISPR&draw=3&rank=13).

Human papilloma virus (HPV) induced cervical tumors usually contain integrated copies from the HPV genome. These fragments usually include E6 and E7 HPV oncogenes. E6 induces destruction of cellular tumor suppressor p53, while E7 destabilizes retinoblastoma protein (Rb). Therefore, removal of these fragments from the genome of the tumor cells using the CRISPR / Cas9 technique causes increased expression of p53 and Rb in tumor cells, leading to apoptosis or cell division in the cells (Zhen and Li, 2017). The effectiveness of this treatment is investigated by Chinese researchers with a clinical trial initiated in 2018 (https://clinicaltrials.gov/ct2/show/NCT03057912?term=CRISPR&draw=2&rank=1).

Both beta thalassemia and sickle cell anemia are caused by beta globin mutations. In the treatment method called CTX001, developed by CRISPR Therapeutics, B-globin gene is edited in order to initiate the synthesis of fetal hemoglobin in the bone marrow cells from patients, using CRISPR / Cas9, and is reintroduced to patients. With this approach, serious improvement was achieved in mice. The clinical trials of CTX001 are ongoing and investigational new drug application has been accepted by the FDA (Memi et al., 2018; https://clinicaltrials.gov/ct2/show/NCT03655678?term=CRISPR&draw=3&rank=7).

#### 2. CONCLUSION

The results of the preclinical studies using CRISPR / Cas9 to treat different diseases are very promising and many clinical trials are ongoing. In particular, the studies developing CAR-T cells for cancer immunotherapy have been progressed. CRISPR-Cas9 system has many advantages compared to the other nucleases, such as high efficiency, simple usage, low cost and multiplexability. On the other hand, off -target effect and the necessity to increase the effectiveness of delivery are among the disadvantages of this technique to be overcome. It is foreseen that these disadvantages can be overcome with alternative approaches and in the near future CRISPR / Cas9 mediated gene editing can be used in clinic.

#### REFERENCES

- Barrangou R, Fremaux C, Deveau H, Richards M, Boyaval P, Moineau S, Romero DA, Horvath P (2007). CRISPR provides acquired resistance against viruses in prokaryotes. Science, 315: 1709–1712.
- Baylis F, McLeod M (2017). First-in-human Phase 1 CRISPR gene editing cancer trials: are we ready. Curr Gene Ther, 17(4): 309–319.
- Bozok Çetintaş V, Kotmakçı M, Tezcanlı Kaymaz B (2017). BağışıklıkYanıtındanGenomTasarımına; CRISPR-Cas9Sistemi. Turkiye Klinikleri J Med Sci, 37(1): 27-42.
- Conboy I, Murthy N, Etienne J, Robinson Z (2018). Making gene editing a therapeutic reality. F1000Res, 7. pii: F1000.
- Foss DV, Hochstrasser ML, Wilson RC (2019). Clinical applications of CRISPR-based genome editing and diagnostics. Transfusion, 59(4): 1389-1399.
- Hille F, Charpentier E (2017). CRISPR-Cas: biology, mechanisms and relevance. Philos Trans R Soc Lond B Biol Sci, 371(1707): 20150496.
- Horvath P, Barrangou R (2010). CRISPR/Cas, the immune system of bacteria and archaea. Science, 327(5962):167-170.
- Hussain W, Mahmood T, Hussain J, Ali N, Shah T, Qayyum S, Khan I (2019). CRISPR/Cas system: A game changing genome editing technology, to treat human genetic diseases. Gene, 685:70-75.
- Hynes AP, Labrie SJ, Moineau S (2017). Programming native CRISPR arrays for the generation of targeted immunity. MBio, 7(3): e00202–16.
- Ishino Y, Shinagawa H, Makino K, Amemura M, Nakata A (1987). Nucleotide sequence of the iap gene, responsible for alkaline phosphatase isozyme conversion in Escherichia coli, and identification of the gene product. J. Bacteriol, 169: 5429–5433
- Jansen R, Embden J, Gaastra W, Schouls L (2002). Identification of genes that are associated with DNA repeats in prokaryotes. Mol. Microbio, 43: 1565–1575
- Jiang F, Doudna JA (2017). CRISPR-Cas9 Structures and Mechanisms. Annu Rev Biophys, 46: 505-529.
- Karimian A, Azizian K, Parsian H, Rafieian S, Shafiei-Irannejad V, Kheyrollah M, Yousefi M, Majidinia M, Yousefi B (2019). CRISPR/Cas9 technology as a potent molecular tool for gene therapy. J Cell Physiol, 234(8): 12267-12277.
- Koonin EV, Makarova KS, Zhang F (2017). Diversity, classification and evolution of CRISPR-Cas systems. Curr Opin Microbiol, 37: 67-78.
- Lau CH, Suh Y (2017). In vivo genome editing in animals using AAV-CRISPR system: applications to translational research of human disease. F1000Res, 6: 2153.
- Mahmoudian-sani MR, Farnoosh G, Mahdavinezhad A, Saidijam M (2018). CRISPR genome editing and its medical applications. Biotechnology & Biotechnological Equipment, 32(2): 286-292.
- Martinez-Lage M, Puig-Serra P, Menendez P, Torres-Ruiz R, Rodriguez-Perales S (2018). CRISPR/Cas9 for Cancer Therapy: Hopes and Challenges. Biomedicines, 6(4). pii: E105.
- Memi F, Ntokou A, Papangeli I (2018). CRISPR/Cas9 gene-editing: Research technologies, clinical applications and ethical considerations. Semin Perinatol, 42(8): 487-500.

Misra S (2013). Human gene therapy: a brief overview of the genetic revolution. J Assoc Physicians India, 61(2): 127-133.

- Mojica FJ, Díez-Villaseñor C, Soria E, Juez G (2000). Biological significance of a family of regularly spaced repeats in the genomes of Archaea, Bacteria and mitochondria. Mol. Microbiol, 36: 244–246.
- Mout R, Ray M, Lee YW, Scaletti F, Rotello VM (2017). In Vivo Delivery of CRISPR/Cas9 for Therapeutic Gene Editing: Progress and Challenges. Bioconjug Chem, 28(4): 880-884.
- Nishimasu H, Ran FA, Hsu PD, Konermann S, Shehata SI, Dohmae N, Ishitani R, Zhang F, Nureki O (2014). Crystal structure of Cas9 in complex with guide RNA and target DNA. Cell, 156(5): 935–49.

Retrived in May, 25, 2019 from https://clinicaltrials.gov/ct2/show/NCT03057912?term=CRISPR&draw=2&rank=1

Retrived in May, 25, 2019 from https://clinicaltrials.gov/ct2/show/NCT03655678?term=CRISPR&draw=3&rank=7

- Retrived in May, 25, 2019 from https://clinicaltrials.gov/ct2/show/results/NCT03545815
- Retrived in May, 25, 2019 from https://clinicaltrials.gov/ct2/show/NCT03166878?term=CRISPR&draw=3&rank=13
- Retrived in May, 25, 2019 from https://clinicaltrials.gov/ct2/results?cond=&term=CRISPR&cntry=&state=&city=&dist=
- Retrived in May, 25, 2019 from https://www.synthego.com/guide/how-to-use-crispr/transfection protocols
- Rodríguez-Rodríguez DR, Ramírez-Solís R, Garza-Elizondo MA, Garza-Rodríguez ML, Barrera-Saldaña HA (2019). Genome editing: A perspective on the application of CRISPR/Cas9 to study human diseases (Review). Int J Mol Med, 43(4):1559-1574.
- Rupp LJ, Schumann K, Roybal KT, Gate RE, Ye CJ, Lim WA, Marson A (2017). CRISPR/Cas9-mediated PD-1 disruption enhances anti-tumor efficacy of human chimeric antigen receptor T cells. Sci Rep,7(1): 737.
- Tang Y, Fu Y (2018). Class 2 CRISPR/Cas: an expanding biotechnology toolbox for and beyond genome editing. Cell Biosci, 12: 8:59.
- Zarei A, Razban V, Hosseini SE, Tabei SMB (2019). Creating cell and animal models of human disease by genome editing using CRISPR/Cas9. J Gene Med,21(4): e3082.
- Zhan T, Rindtorff N, Betge J, Ebert MP, Boutros M (2019). CRISPR/Cas9 for cancer research and therapy. Semin Cancer Biol, 55: 106-119.
- Zhen S, Li X (2017). Oncogenic Human Papillomavirus: Application of CRISPR/Cas9 Therapeutic Strategies for Cervical Cancer. Cell Physiol Biochem, 44(6):2455-2466.
- Zhu QM, Ko KA, Ture S, et al. Novel Thrombotic Function of a Human SNP in STXBP5 Revealed by CRISPR/Cas9 Gene Editing in Mice. Arterioscl Thrombosis Vascular Biol. 2017;37(2):264–270.
- Zych AO, Bajor M, Zagozdzon R (2018). Application of Genome Editing Techniques in Immunology. Arch Immunol Ther Exp (Warsz), 66(4): 289–298.



## Comparison on Basic Electronic Properties of Schottky Diodes with Different Rectifier Contacts

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**Abstract:** In this work, experimentally, metal-semiconductor structures based on AuPd/n-GaAs and Ag/n-GaAs Schottky diodes (SDs) without interfacial layer were produced by utilizing DC sputter and dropping Ag paste to analize their electronic characteristics. Then, the current-voltage (I-V) characteristics of the SDs at room temperature were obtained from the region in forward and reverse biases (at +/-3V). The characteristics of SDs, evaluated by using thermionic emission (TE) theory, Ohm's law, Cheung and Cheung's function and modified Norde's function, were compared to different rectifier contact metals.

Keywords: Schottky diodes, Rectifier contact, n-GaAs, I-V characteristics, Basic electronic properties,

#### **1. INTRODUCTION**

Schottky diodes (SDs) have become indispensable elements of semiconductor based electronic technology. SDs has attracted a great deal of attention in the last few years due to its potential applications in electronics such as microwave circuit components, switching elements, opto-electronic devices, memory elements, sensors, photodetectors, solar cells, rectifier devices. SDs in order to achieve maximum efficiency, especially in the fabrication process resulting from the need to affect the electronic characteristics of the diodes, as well as used in all effects caused by metals and semiconductors has been an essential feature a separate research. [1-6] As a result of the potential barrier between the metal (M) and the semiconductor (S) connection structure, rectifier devices are formed. Surface preparation process, barrier height, dislocations, impurities, applied voltage and series resistance ( $R_s$ ) are important in many rectifying equipment such as rectifier. [7,8] There are many techniques to calculate basic electronic properties from the I-V measurement, such as TE theory, Ohm's law, Cheung and Cheung's function, and modified Norde's functions.[2,9,10,11]

As a semiconductor, GaAs is a III-V direct bandgap semiconductor which has direct forbidden bandgap, high electron mobility, a high break-down voltage and mechanical stability. AuPd and Ag are used for the preparation of rectifier contacts. Au and Pd ( $\sim$ 5.1 eV) have higher work function than Ag ( $\sim$ 4.3 eV). At the same time, GaAs's work function equals to 4.69 eV [2,3,12].

In regard of this research; metal-semiconductor structures based on AuPd/n-GaAs and Ag/n-GaAs Schottky diodes(SDs) without interfacial layer were produced by utilizing DC sputter and dropping Ag paste. So, the effect of different rectifier contacts on the performance of SDS were investigated in detail.

#### 2. MATERIAL AND METHODS

Experimentally, metal-semiconductor structures based on Au-Pd/n-GaAs and Ag/n-GaAs SDs as without interfacial layers were prepared to investigate performance of SDs. Firstly, n-type GaAs (100) wafers were cleaned with acetone and isopropyl alcohol to remove any organic. Then, the wafers were immersed in HF+10H<sub>2</sub>O solution and allowed to stand for 1 min. which was used to remove native oxides and then quenched in de-ionized water resistivity of 18 M $\Omega$ .cm. Finally, the wafers were dried with Ar atmosphere.

Approximately, 500 nm thick AuPd (40%-60%) metal as an ohmic contacts were deposited by magnetron DC sputtering on the unpolished side of the crystal. Then, the substrates were annealed at 600 °C in the Argon atmosphere for 30 min.

The AuPd rectifier contacts were deposited by DC sputtering, and their shape was a circular dot with a diameter of 2 mm and a thickness of around 500 nm. Ag contact was obtained by dropping. Due to different size area, each contact area was calculated one by one. The thickness of Ag was between 10 µm and 80 µm. Two series SDs were fabricated and classified as Group AuPd and Group Ag. In each group, there were two SDs.

#### 3. RESULTS AND DISCUSSION

On the prepared samples, the forward and reverse I-V measurement was performed by the use of a Keithley 2400 Sourcemeter. The I-V measurements carried out at  $\pm 3V$  by 20mV steps in room temperature (295 K) under dark conditions (Fig. 1). The I-V characteristics of different rectifier contacts investigated at both regions of forward and reverse biases are presented as semi-logarithmic scaled (InI-V) in Fig. 2.



Figure 1. Schematic and actual cross section of measurement system.

When the rectification rates of diodes are examined in Table 1, the rectifier ratio  $(RR=I_F/I_R)$  of the AuPd group are better than the Ag group. The reason for this is the difference between the work function. Because in order to be a good rectifer, AuPd's work function must be greater than GaAs's work function.

The I-V characteristics of a SDs under TE theory can be expressed as [12].

$$I = I_o \left[ \exp\left(\frac{q(V - IR_s)}{nkT}\right) - 1 \right]$$
<sup>(1)</sup>

Here, the parameters indicated by n, q, V,  $IR_s$ , T,  $I_o$  and k are the ideality factor, the charge of electron, applied bias voltage on the structure, the voltage drop on the series resistance ( $R_s$ ), temperature, saturation current and Boltzmann's constant, respectively. Here, the saturation current ( $I_o$ ) can be extracted from the straight-line intercept of ln(I) at zero biases as given in Eq. (2).

$$I_o = AA^*T^2 \exp\left(-\frac{q\Phi_{B0}}{kT}\right)$$
<sup>(2)</sup>

The intercept and slope of the ln(I)-V plot give the  $\Phi_{B0}$  and n values. The  $\Phi_{B0}$  and n values are expressed as:

$$\Phi_{B0} = \frac{kT}{q} \exp\left(\frac{AA^*T^2}{I_o}\right)$$
(3)  
$$n = \frac{q}{kT} \left(\frac{dV}{d(\ln I)}\right)$$
(3)

(4)

Where the rectifier contact area and the effective Richardson constant are indicated by A and A\* (8,16 A/cm<sup>2</sup>K<sup>2</sup> for ntype GaAs), respectively [14]. The I<sub>o</sub>, n and  $\Phi_{B0}$  values for AuPd and Ag groups were tabulated as below in Table I. As seen in Table I, the n factor values of Ag group are higher than AuPd group. The n factor of group AuPd are between 2.456 and 4.002. Others are between 10.410 and 12.576. Ag (~4.3 eV) have lower metal work function than AuPd (~5.1 eV). To be a good rectifier, work function of used metal must be higher than work function of GaAs (4.69 eV) [12].



Figure 2. The I-V characteristics of the devices; the effects of different rectifier contacts on SDs are shown at a log scale.

The R<sub>s</sub> and R<sub>sh</sub> are an important parameter which affect device's performance. Not practically but theoretically the R<sub>s</sub> value is supposed to be "0", and the R<sub>sh</sub> value emerges to " $\infty$ ". The R<sub>s</sub> and R<sub>sh</sub> values of SDs can be calculated with different methods [2,9,10,11,16]. Ohm's law (R<sub>i</sub>=dV<sub>i</sub>/dI<sub>i</sub>) was used to calculate R<sub>s</sub> (at +3 V) and R<sub>sh</sub> (at -3V) from the I-V curve which tabulated in Table I, respectively. For AuPd group, these values are changed from 0.655 k $\Omega$  to 454.084 k $\Omega$ . For Ag group, these values are changed from 103.460 k $\Omega$  to 3888.740 k $\Omega$ .

| Samples | from Th                | from Ohm's Law |                      |               |                       |   |                      |
|---------|------------------------|----------------|----------------------|---------------|-----------------------|---|----------------------|
|         | I <sub>0</sub> (A)     | n              | Ф <sub>в0</sub> (eV) | $RR(I_F/I_R)$ | <b>R</b> <sup>2</sup> | $\mathbf{R}_{s}\left(\mathbf{K}\Omega\right)$ | R <sub>sh</sub> (KΩ) |
| AuPd_D1 | 2.74x10 <sup>-10</sup> | 2.456          | 0.814                | 287.976       | 0.999                 | 0.655   | 188.684              |
| AuPd_D2 | 1.61x10 <sup>-09</sup> | 4.002          | 0.828                | 478.154       | 0.997                 | 0.949   | 454.084              |
| Ag_D1   | 8.81x10 <sup>-10</sup> | 10.410         | 0.779                | 2.85188       | 0.996                 | 717.837                                       | 2047.180             |
| Ag_D2   | 2.23x10 <sup>-09</sup> | 12.576         | 0.756                | 3.75869       | 0.999                 | 103.460                                       | 3888.740             |

Table I. Results of the SDs by using thermionic emission (TE) theory and Ohm's law.

The other method to calculate the  $R_s$  values of SDs is the Cheung and Cheung's function which has been acquired from the forward-biases of I-V plot [2]. The Eq. 6 and 7 are referred to as the Cheung and Cheung's function.

$$\frac{dV}{d(\ln I)} = IR_s + \left(\frac{nkT}{q}\right) \tag{6}$$

$$H(I) = V - \frac{nkT}{q} \ln\left(\frac{I}{AA^*T^2}\right) = IR_s + n\Phi_{B0}$$
<sup>(7)</sup>

According to these equations, the dV/dln(I) and H(I) versus I plots of AuPd and Ag groups are presented in Figure 3. According to the dV/dln(I)-I graphs, the R<sub>s</sub> for group AuPd and Ag groups range from 0.429 k $\Omega$  to 0.819 k $\Omega$  and 12.706

 $k\Omega$  to 32.812 k $\Omega$ , respectively. At the graphs of H(I)-I, the Rs of group AuPd are between 0.300 k $\Omega$  and 0.430 k $\Omega$ . The Rs of group Ag are between 61.534 k $\Omega$  and 188.904 k $\Omega$ , respectively.



**Figure 3.** The dV/d (ln I) and H(I) versus I curves obtained from Cheung's functions for SDs When the values of n from TE (Table I) are greater than 2, the modified Norde's function is expressed by Bohlin as below [11]:

$$F(V) = \frac{V}{\gamma} - \frac{kT}{q} \ln\left(\frac{I(V)}{AA^*T^2}\right)$$
(8)

here is grater than n values of SJSs. F(V)-V plots have minimum points of  $V_{min}$  and  $I_{min}$  which use to calculate  $\Phi_{B0}$  and  $R_s$  values from Eq. 8a and 8b.

$$\Phi_{B0} = F(V_{\min}) + \frac{V_{\min}}{\gamma} - \frac{kT}{q}$$
(8a)

$$R_s = \frac{(\gamma - n)kT}{qI_{\min}} \tag{8b}$$

The F(V)-V plots of the AuPd/n-GaAs and Ag/n-GaAs SDs are shown in Fig. 4. The modified Norde's functions calculated the value of  $R_s$  as below. Analyzing the values given in Table II, the  $R_s$  values of AuPd group are lower than Ag group.  $\Phi_{B0}$  values range from 0.782 eV to 0.924 eV.

Table II. Results of SDs by using Cheung's function and modified Norde's function.

| Samples | f  | function | From m               | odifie         | d Norde's            | function           |                 |  |          |
|---------|--|----------|----------------------|----------------|----------------------|--------------------|-----------------|--|----------|
|         | $dV/dlnI(\Omega)$  |          | $H(I)$ - $I(\Omega)$ |                |                      |                    |                 |  |          |
|         | $\mathbf{R}_{s}(\mathbf{k}\Omega)$ $\mathbf{R}^{2}$ $\mathbf{R}_{s}(\mathbf{k}\Omega)$ |          | $\Phi_{B0}(eV)$      | $\mathbb{R}^2$ | F(V <sub>min</sub> ) | $\mathbf{V}_{min}$ | $\Phi_{Bo}(eV)$ | $\mathbf{R}_{\mathrm{s}}\left(\mathbf{k}\Omega\right)$ |          |
| AuPd_D1 | 0.429  | 0.976    | 0.300                | 1.129          | 0.999                | 0.7765             | 0.52            | 0.924  | 12.472   |
| AuPd_D2 | 0.819  | 0.970    | 0.429                | 0.867          | 0.997                | 0.7791             | 0.80            | 0.782  | 772.351  |
| Ag_D1   | 12.705   | 0.436    | 61.534               | 0.828          | 0.990                | 0.7762             | 0.40            | 0.787  | 3615.120 |
| Ag_D2   | 32.818   | 0.665    | 188.904              | 0.772          | 0.994                | 0.754              | 0.88            | 0.796  | 309.646  |



Figure 4. The F(V) versus V curves obtained from modified Norde's functions for SDs

#### 4. CONCLUSION

The effects of different rectifier contact on the I-V characteristics of AuPd/n-GaAs and Ag/n-GaAs Schottky diodes have been investigated. The values of ideality factor,  $R_s$  and and  $R_{sh}$  are smaller for the AuPd group than that for the Ag group. In light of these informations, when we compare the results of the AuPd and Ag SDs, AuPd SD shows good results. AuPd contact is suitable for producing electronical applications.

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#### REFERENCES

- [1] Bardeen, J., Surface states and rectification at a metal semi-conductor contact, Phys. Rev., 71, 717-727, 1947.
- [2] S. M. Sze, Semiconductor Devices: Physics and Technology, New Tork: Wiley, 1985.
- [3] Ziel, A. V., Solid State Physical Electronics, 2nd Ed.Prentice-Hall, Inc., 1968.
- [4] Aboelfotoh, M.O., Temperature dependence of the Schottky-barrier height of tungsten on n-type and p-type silicon, Solid State Electronics, 34, 1, 51-55, 1991.
- [5] S. O. Tan, H. Uslu Tecimer, O. Çiçek, H. Tecimer, İ. Orak ve Ş. Altındal, «Electrical characterizations of Au/ZnO/n-GaAs Schottky diodes under distinct illumination intensities,» J Mater Sci: Mater Electron, cilt 27, pp. 8340-8347, 2016.
- [6] O. Çiçek, H. Uslu Tecimer, S. Tan, H. Tecimer, Ş. Altındal ve I. Uslu, «Evaluation of electrical and photovoltaic behaviours as comparative of Au/n-GaAs (MS) diodes with and without pure and graphene (Gr)-doped polyvinyl alcohol (PVA) interfacial layer under dark and illuminated conditions,» *Composites Part B*, cilt 98, pp. 260-268, 2016.
- [7] Ü. Özgür, Y. I. Alivov, A. Teke, M. A. Reshchikov, S. Doğan, V. Avruti, S. J. Cho ve H. Morkoç, «A comprehensive review of ZnO materials and devices,» *Journal of Applied Physics*, cilt 98, p. 041301, 2005.

- [8] F. Z. Pür ve A. Tataroğlu, «Analysis of the series resistance and interfacial states of Au/Si3N4/n-Si (metal-insulatorsemiconductor) Schottky diodes using I–V characteristics in a wide temperature range,» *Physica Scripta*, cilt 86, p. 035802, 2012.
- [9] S. K. Cheung ve N. W. Cheung, «Extraction of Schottky diode parameters from forward current-voltage characteristics,» *Appl. Phys. Lett.*, cilt 49, no. 2, pp. p. 85,, 1986.
- [10] H. Norde, «A Modified Forward I-V Plot for Schottky Diodes with High Series Resistance,» J. Appl. Phys., cilt 50, no. 7, pp. 5052-5053, 1979.
- [11] K. E. Bohlin, «Generalized Norde plot including determination of the ideaUty factor,» *Journal of Applied Physics*, cilt 60, p. 1223, 1986.
- [12] B. L. Sharma, Metal-Semiconductor Schottky Barrier Junctions and Their Applications, New York: Plenum Press, 1984.
- [13] C. J. Stephen, Zinc Oxide Bulk, Thin Films and Nanostructures, Elsevier Science, 2006.
- [14] E. Rhoderick ve R. Williams, Metal-Semiconductor Contacts, Oxford: Oxford University Press, 1978.
- [15] I. Orak, A. Koçyiğit ve A. Turut, «The surface morphology properties and respond illumination impact of ZnO/n-Si photodiode by prepared atomic layer deposition technique,» *Journal of Alloys and Compounds*, cilt 691, pp. 873-879, 2017.
- [16] M. Gökçen, Ş. Altındal, M. Karaman ve U. Aydemir, «Forward and reverse bias current–voltage characteristics of Au/n-Si Schottky barrier diodes with and without SnO<sub>2</sub> insulator layer,» *Physica B Condens. Matter.*, cilt 406, no. 21, p. 4119–4123, 2011.



## ORAL PRESENTATION

# LabVIEW Based Program for Analyzing Photovoltaic Cells

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Abstract: In this study, it was aimed to calculate the datas obtained from the photovoltaic measurement system quickly. For this purpose, AuPd/ZnO/n-GaAs Schottky Junction Structure (SJS) was produced by using RF and DC sputtering techniques to investigate on electronic properties on SJSs. The forward and reverse current-voltage (I-V) characteristics of SJS at  $\pm 3V$  were measured at room tempereture. The electronics properties such as the series resistance, the shunt resistance, the ideality factor and the barrier height were calculated by using thermionic emission (TE) theory, Ohm's law, Cheung and Cheung's function and Norde's function. Based on Labview<sup>®</sup> software to design an easy handling Graphical User Interface (GUI) was developed. In this way, the results were obtained and graphed from tool when input values (temperature, contact area, Richardson constant etc.).

Keywords: Labview, Schottky Junction Structure (SJS), ZnO nano thin films, I-V characteristics, basic electronic properties.



## **Quasi-Static Indentation of Transition Metal Doped ZnO Nanoparticles**

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**Abstract:** Sodium and Yttrium doped zinc oxide nanoparticles were synthesized via sol-gel process. The phase, structural and mechanical properties were investigated using X-ray diffraction, scanning electron microscopy, energy dispersive X-ray spectroscopy and micro hardness based on quasi-static indentation technique. Load-depth curves were obtained by applying indentation loads at room temperature. As the doping concentration increased, a significant increase was observed in the hardness values computed from loading-unloading curves using the Oliver and Pharr method.

Keywords: ZnO, mechanical properties, quasi-static indentation

#### **1. INTRODUCTION**

In recent years, semiconductor structures play an important role in both technological and scientific research. The vast majority of the most important parts of electronic devices such as computers, mobile phones and digital storage devices are fabricated from semiconducting materials. Among various metal oxide semiconductors, considerable research has been focused on ZnO because of its unique physical, chemical, magnetic, electrical, optical, piezoelectric, and mechanical properties. Many devices such as solar cells, power generators, laser diodes, light emitting diodes, transducers, actuators, and sensors have been developed using these properties (Souri et al., Wang et al., Tsukazaki et al.) Among the semiconductors, diluted magnetic semiconductors (DMSs) have attracted significant interest due to their potential applications in spintronics devices, such as optoelectronics, spin field-effect transistors, field emissions devices, gas sensors, non-volatile memory devices and quantum computers. Spintronics devices with smaller size, less energy consumption and faster operation can be obtained by combining existing features of magnetic and semiconducting materials. Zinc oxide (ZnO) is one of the attractive candidates for these applications due to its low toxicity, low cost and potential applications. Theoretical and experimental studies have predicted that doped ZnO with transition metals (TM) and rare earth (RE) elements shows enhanced mechanical properties.

#### 2. MATERIALS AND METHODS

Y doped ZnO nanoparticles were synthesized by sol gel method. Zinc Acetate dihydrate (Zn (CH<sub>3</sub>COO)<sub>2</sub>•2H<sub>2</sub>O, Merck), and Yttrium acetate tetrahydrate (Y(CH<sub>3</sub>CO<sub>2</sub>)<sub>3</sub>•4H<sub>2</sub>O, Alfa Aesar) were used as precursor materials. Methanol used as solvent and Monoethanolamine is used as sol stabilizer. 0.25 M methanolic precursor solution is prepared, stirred and aged overnight. After aging solutions are dried in ambient atmosphere to form the gel. Once all the solvent is evaporated gels were heat treated at 400°C for 10 minutes to burn the organic material and 600°C for 30 minutes for proper crystal orientation. X-ray diffraction (XRD - Bruker D8 Advance) measurements were performed to characterize crystal structure and make phase analysis. Scanning Electron Microscopy (SEM – Quanta FEG 250) was used to investigate surface morphology, as well as indent imprint and cracks. Energy Dispersive Spectroscopy (EDS-EDAX) Mapping analysis was performed to determine elemental constituents and simple quantitative concentrations of all the samples. Instrumented Indentation tests were performed by using Universal Mechanical Tester (UMT- Bruker SYS2). Indentations were carried out, by using Vickers type with the loads of 400, 800, 1200, 1600 and 2000 mN for 10 s at room temperature.

#### 3. RESULTS AND DISCUSSION

#### Results

### Structural analysis

X-ray diffraction spectra of undoped, Na doped and Y doped ZnO are shown in Fig. 1. It is clear from the diffraction peaks that all the samples show typical hexagonal wurtzite structure. No impurity peak or secondary phase is observed in the diffraction pattern confirming that doping ions incorporated into Zn sites. The diffraction peaks correspond to (100), (002), (101), (102), (110), (103), (200), (112), (201) reflections. The intensity of (101) is the highest in all the samples. However, in the case of Na doping, the intensity of this peak decreased, confirming that decrement in the crystal quality of the samples. A shift towards lower angle direction is observed in the doped samples. The tensile compressive strain caused by larger ionic radius of Y and Na ions as compared to the Zn ions could be responsible for this shifting [Cullity et al.].

Fig. 2 shows the SEM images of the samples. Undoped sample shows more voids compared to the rest of the samples. Interestingly, Y doped sample shows a quite dense and compact distribution. Nevertheless, in the Na doped sample, non-uniform particle distribution and deep voids are observed. Thus, it can be concluded that Y doping provides compact microstructure while Na doping causes porosity and intergranular voids in ZnO.

Fig. 3 shows EDS mapping spectra and elemental concentrations of all the samples. The samples contain of zinc, oxygen, yttrium and sodium elements. Depending on the mapping analysis, there is no elemental impurity in the samples. It is clear from the mapping that, all the doping elements are is distributed homogeneously in the ZnO microstructure.



(c)

Figure 1. XRD spectra of (a) undoped, (b) Y and (c) Na doped ZnO nanoparticles.



Figure 2. SEM images of (a) undoped, (b) Y and (c) Na doped ZnO nanoparticles.



Figure 3. EDX mapping images of (a) undoped, (b) Y and (c) Na doped ZnO nanoparticles.

#### Mechanical analysis

Fig. 4 (a)-(c) show the load- contact depth curves of undoped, Y doped and Na doped ZnO respectively. The applied load are 400, 800, 1200, 1600 and 2000 mN in the test. The figures also include the hardness and indentation modulus values calculated from load- unload curves for all the samples. All the mechanical parameters were obtained by applying Oliver and Pharr method. In this method, hardness and indentation modulus are calculated by considering both the load and depth during plastic and elastic deformation.

By using this method, the hardness and elastic modulus can be determined more accurately and precisely compared to static indentations technics. The loading part can be expressed as

$$F = Ch^n \tag{1}$$

where F is the applied load, h contact depth, and C and n are the constant related with geometry of indenter tip. Value of the exponent m for cones is 2 (Oliver et al).

The fiting unloading curve is given by

$$F = a(h - h_0)^m \tag{2}$$

where  $h_0$  final contact depth, *a* and *m* are the fit parameters related with geometry of indenter tip. The contact stiffness is obtained by derivative of fitting unloading curve as expressed below (Zeng et al.)

$$S = \left|\frac{dF}{dh}\right|_{F_{max}} = ma \ (h - h_0)^{m-1} \tag{3}$$

and the contact depth can be written as

$$h_{c=}h_{max}\mathcal{E} \ \frac{F_{max}}{S} = h_{max}\mathcal{E} \ \frac{F_{max}}{dF_{dh}}$$
(4)

where  $F_{max}$  is maximum load and  $\mathcal{E}$  is contact model parameter which is 0.75 for Vickers indenter (Torres et al.). In case of hard material indentation, the indenter deformation can be taken into account by considering reduced modulus determined from the slope of the unloading curve at maximum load as given below

$$E_r = \frac{\sqrt{\pi}}{2\beta\sqrt{A_c}} \tag{5}$$

where  $A_c$  area of indent imprint,  $\beta$  is a contact model parameter (Oliver et al.). By using equation (5) the indentation module can be expressed as

$$\frac{1}{E_r} = \frac{1 - v^2}{E} + \frac{1 - v_i^2}{E_i} \tag{6}$$

where *E* and *v* are Young's Modulus and Poisson's ratio of the sample while  $E_i$  and  $v_i$  are the same parameters for the indenter, and hence, the hardness is

$$H = \frac{F_{max}}{A_c} \tag{7}$$

According to figures, there is a small but systematic increase in the hardness with applied load while there is a proportional increase in the indentation modulus values with increasing load. Thus, it can be concluded that the mechanical properties of this material depend on the applied load. This increase in the hardness values is called the reverse indentation size effect (RISE).

There are different approaches in the literature that are related to origin of RISE: (i) the loss of energy arising from material chipping around the indenter during loading process, (ii) the formation of cracks during the loading in brittle materials which in plastic deformation is dominant, (iii) the existence of a distorted zone near the crystal-medium interface (Kaya et.al). Indentation modulus of the samples were from the load-unload curves, as well. Depending on figure 4, the indentation modulus of the samples increased with increasing applied load, confirming that the indentation modulus is load dependent.



Figure 4. Indentation curves and indentation results of (a) undoped, (b) Y and (c) Na doped ZnO nanoparticles.

#### Discussion

When the hardness values of the samples are examined in general, it can be seen that the hardness of the undoped sample has the lowest value. With Y doping, a significant reduction was observed in the hardness of ZnO. It is also observed that the hardness value of Y doped ZnO is higher than Na doped ZnO. So, it can be concluded that Y doping is more effective than Na in obtaining compact ZnO structure. There is also an increment in the indentation modulus of the samples. Na doped ZnO has the highest indentation modulus values.

#### 4. CONCLUSION

Undoped, Y doped and Na doped ZnO nanoparticles were synthesized successfully via sol-gel process. The structural and mechanical properties were investigated by using x-ray diffraction (XRD), scanning electron microscope (SEM), Energy Dispersive X-ray Spectroscopy (EDS) and Universal Mechanical Tester (UMT). Instrumented indentation method was used to investigate mechanical properties of the samples. Obtained results show that Y and Na doping play an important role in improving mechanical properties of ZnO.

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#### REFERENCES

- A.Tsukazaki, A. Ohtomo, T. Onuma, M. Ohtani, T. Makino, M. Sumiya, K. Ohtani, S. F. Chichibu, S. Fuke, Y. Segawa, H. Ohno, H. Koinuma, M. Kawasaki, Repeated
- B.D. Cullity, Elements of X-Ray Diffraction, second ed., Addison-Wesley Publishing Company Inc., Phillippines, 1978.
- F. Torres, Y. Benino, T. Fujiwara, T. Komatsu. Evaluation of elastic/mechanical properties of some glasses and nanocrystallized glass by cube resonance and nanoindentation methods. Mater. Res. Bull., 39, (2004) 1431-1443.
- H. Souri, I.W. Nam, H.K. Lee, A zinc oxide/polyurethane-based generator composite as a self-powered sensor for traffic flow monitoring, Compos. Struct.134 (2015) 579-586.
- K. Zeng, C.-h Chiu, An analysis of load-penetration curves from instrumented indentation, Acta Mater, 49 (2001) 3539-3551.
- Oliver, W., Pharr, G. An improved technique for determining hardness and elastic modulus using load and displacement sensing indentation experiments. J. Mater. Res., 6 (1992) 1564-1583.
- S. Kaya, D. Akcan, O. Ozturk, L. Arda. Enhanced mechanical properties of yttrium doped ZnO nanoparticles as determined by instrumented indentation technique. Ceram. Int., 44 (9) (2018), pp. 10306-10314
- W. Oliver, G. Pharr. Measurement of hardness and elastic modulus by instrumented indentation: Advances in understanding and refinements to methodology. J. Mater. Res., 19 (2004) 3-20.
- Z.L. Wang, Zinc oxide nanostructures: growth, properties and applications, J.Phys. Condens. Matter, 16 (2004) 829-858.



# Flexible Antenna Applicator Design for Medical Applications and Low Energy Pulsed Radio Frequency Energy Applications

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**Abstract:** Low frequency and low-energy electromagnetic field (EMF)'s research work for soft tissue injuries, skin ulcers, non-degenerative nerve diseases, and bone fractures treatment is an effective method. This method can be divided into different categories according to the technical parameters used in clinical applications. One of these treatments is pulsed radio frequency energy (PRFE) shortwave radio frequency energy in the 13.56-27.12 MHz carrier frequency range. In this paper, due to more effective and better result of use in wearable applications a flexible antenna applicator operating at 27 MHz frequency range is designed for the purpose of accelerates wound healing. The subject of study has diverse technical potential in medical applications. New and feasible treatment modalities can be developed with minor changes for different diseases (tooth problems, bone fractures, chronic pain, etc.).

**Keywords:** Electromagnetic field (EMF), pulsed radio frequency energy (PRFE), flexible antenna, wound healing, wearable applications

### **1. INTRODUCTION**

Nowadays the cellular, molecular and physiological processes of wound healing and resulting positive effects have attracted great attention in the scientific health community. Local wound factors and systematic mediators act as regulators in the healing process. It is estimated that approximately 1-2% of the population in developed countries is complaining of chronic wounds [1]. In addition to the fact issue, chronic wounds are an important health problem affecting the patient's quality of life, they also cause high health costs in the World. Recently, advances have been made in this field with the technological developments in wound treatments. Despite these developments, the specific approaches in wound treatment still remain important due to the complex structure and patient diversity of wound healing [2]. The electromagnetic field (EMF) has an important role in the use of stimulator and therapeutic purpose in medicine and biology. Since the discovery of the electromagnetic field (EMF)s, it has attracted the interest of scientists in treatment and diagnosis. In particular, non-ionizing electromagnetic field (EMF)s are used to induce various biological effects in cells [3]. Previous studies have shown that electromagnetic field (EMF) affects cell proliferation, differentiation, cellular cycle, apoptosis, DNA mapping and expression, cytokine expression and more [4-6]. The utilization of low frequency and lowenergy electromagnetic field(EMF) research work for soft tissue injuries, skin ulcers, non-degenerative nerve diseases, and bone fractures treatment is an effective method [7]. This method can be divided into different categories according to the technical parameters used in clinical applications [8]. One of these treatments is the pulsed radio frequency energy (PRFE) shortwave radio frequency energy in the 13.56-27.12 MHz carrier frequency range. In clinical studies, pulsed radio frequency energy (PRFE) therapy has been shown to shorten recovery time and reduce recurrenced. In addition, wound model and treatment mechanism have been investigated. Much of the research in the literature is related to the investigation of the biological effects of very low-frequency magnetic fields in the laboratory environment. Despite the multitude of researches, the mechanism of electromagnetic fields still remains unknown. In Figure 1, there is the transduction mechanism of the most accepted pulsed radio frequency energy in the world. This suggested mechanism does not mean that it has been directly affects EMF activity in independent cytologic Ca + 2 or in the activities of voltage controlled Ca + 2 channels. Instead of the case as in the original design of the ECM model, voltage-dependent Ca + 2implies an EMF effect in connection with CaM. In this paper, due to more effective and better result of use in wearable applications, a flexible antenna applicator operating with the Industrial Scientific Medical (ISM) band at 27 MHz frequency range is designed by CST program purpose of accelerating wound healing.



Figure 1: Mechanism of pulsed electromagnetic flow for tissue repair [9].

### 2. MATERIAL AND METHODS

#### **Kapton Substrate Material**

The main purpose of this project is the design of spiral flexible PRFE applicator operating at 27 MHz for wound healing treatment system. The selection of the substrate material plays a key role in the design process of the proposed spiral flexible PRFE applicator. As the antenna is supposed to be flexible, the suggested materials must present high malleability and robustness levels, in order to stand for definite deformation scenarios, like bending, crumpling, twisting and etc. Morever, these flexible materials should exhibit favorable electromagnetic properties (loss tangent and relative permittivity) to ensure easy combining with radio frequency circuits and to accomplish the performance demands of modern flexible and wearable technologys. After look at a various of flexible antennas defined on the literature, Kapton polyimide film has been chosened as the candidate for spiral flexible PRFE applicator antenna dielectric substrate material. Dielectric constants of polyimides, in general, are known to increase gradually with decreasing frequency. For instance, Kapton H film (0,0254 mm) has a dielectric constant of approximately 3.5 at 1 kHz and 3.3 at 10 MHz [10].

#### **PDMS Substrate Material**

PDMS has become a widely used substrate material for flexible antennas. This tends, can be extensively defined by PDMS's characteristics: it is chemically inert, thermally stable, permeable to gases, simple to handle and manipulate, and exhibits isotropic and homogenous properties [11]. PDMS is a great water isolator so make possible a long-term, stable device (e.g. antenna) use. PDMS has good Radio Frequency (RF) and mechanical properties, a low and adjustable dielectric constant and quite well chemical stability. The initial fluid state of the PDMS lets the preparation of composite substrates of various density, thus controlling the range of dielectric constants that can be made and in addition, PDMS's dielectric constant have a range of 2.3-2.8. Furthermore, the fluid property of the elastomer also offers control over the substrate thickness in addition to the likelihood of immersion the antenna inside the substrate [12].

#### Stages of the Fabrication of the Kapton flexible antenna

In biomedical applications, polymers rapidly have been taken place among important materials. Multidimensional electronic science has been recently received considerable attention. More and more versatile electronic tools are being produced every day. These include flexible imaging instruments, smart labels, and wearable products. In recent years, polymers such as polydimethylsiloxane (PDMS) have been widely used in the manufacture of microwave circuit systems. The PDMS material is used as a backing in the flexible circuit industry. The long lifespan and the ability to withstand copper pressures make PDMS a very important material for such uses. Such materials are generally used in the electronics market as businesses start to prefer more flexible, lighter and faster parts. The figure below shows the production steps of the PDMS material with adjustable dielectric constant, indicated in liters. Kapton material is used as a substrate in the flexible circuit industry. The long lifespan and the ability to withstand copper pressures make Kapton a very important material for such uses. Such enterprises are generally used in the electronics market as enterprises start to prefer smaller, lighter and faster parts [13].



**Figure 2:** (a) PDMS silicone substance and curing chemical are mixed (10: 1 Vol/ Vol). (b) 30 minutes degasing process. (c) The top and the bottom surfaces on the antenna are covered with PDMS material. (d) Heat has been supplied for 16 hours with a value set to 60 degrees. (e) Produced flexible antenna prototype. (f) Bended form of flexible antenna.

# 3. RESULTS AND DISCUSSION

## **PRFE** Applicator Design

In this study, an antenna formed PRFE applicator with an octagonal spiral resonator at 27 MHz operating frequency is designed. This PRFE applicator consists of four layers, the octagonal spiral resonator is printed on the Kapton surface in order for the PRFE applicator to be flexible to of which are made up of PDMS material to be used on the top and bottom layers. The antenna has 45 spiral counts and the epsilon value entered in the CST program is 7.6 for Kapton and 1.9 for PDMS. Epsilon values and size may change slightly in the fabrication stages of the antenna.



**Figure 3:** (a) Geometry of Kapton flexible antenna at CST (b) Geometry of Kapton flexible antenna after fabrication. **Table 1:** Parameter list of Kapton flexible antenna in CST.

| Width                               | 150 mm   |
|-------------------------------------|----------|
| Length                              | 150 mm   |
| Thickness                           | 3.035 mm |
| Thickness of Kapton and PDMS        | 1 mm     |
| Thickness of copper part of antenna | 0.035 mm |
| Thickness of each part of spiral    | 0.75 mm  |
| Gap of between two spiral           | 0.75 mm  |
| 11                                  | 67.72 mm |
| 12                                  | 47.19 mm |
|                                     |          |

#### **Antenna Simulation Results**



**Figure 4:** (a) Geometry of Kapton flexible antenna at CST (b) Result of S11 parameter at CST before matching circuit is applied (c)E-field distribution on Kapton flexible antenna (d) H-field distribution on Kapton flexible antenna.



Figure 5: (a) Curve of antenna for calculation of E-field and H-field (b) E-field result (c) H-field result.

#### **Antenna Experimental Results**

S parameters results before applying the matching circuit are shown in Figure 6. Among the reasons why these results are different from the CST program, it can be said that there are changes in the size of the antenna and the Epsilon values of Kapton and PDMS in the fabrication stages of the antenna. After these results, it was decided to implement the matching circuit. The required components values for the matching circuit obtained using the CST program are shown in Figure 7 (a). The circuit we created is shown in Figure 7 (b). The results of the S parameters after the matching circuit has been applied shows in Figure 8.



Figure 6: (a) S parameter result on air before matching circuit is applied (b) S parameter result on arm before matching circuit is applied (c) S parameter result on shoulder before matching circuit is applied.



**Figure 7:** (a) Capacitor and inductor values given by CST (b) Used matching circuit; capacitor value is 75 pF, inductor value is 1.5µH.



Figure 8: (a) S parameter result on air after matching cicuit is applied. (b) S parameter result on arm after matching cicuit is applied. (c) S parameter result on shoulder after matching cicuit is applied.

Different signal waveforms at 27 MHz are applied to the designed PRFE device and the desired level of electric and magnetic fields are obtained. The block diagram of this process is shown in Figure 9 (a), applied waveforms, input powers and electric and magnetic field results are shown in Figure 9 (b) and Figure 9 (c). And the results obtained for all input power values applied are shown in Table 2.



Figure 9: (a) The block diagram of experimental setup (b) Input power for Kapton flexible antenna measurement system with Signal Hound EMC E5 E-Field probe. After matching circuit is applied, magnitude of E-field result is -58.07 dBm. (c) Input power for Kapton flexible antenna measurement system with Signal Hound EMC H20 H-Field probe. After matching circuit is applied, magnitude of H-field result is -37.54 dBm.

| Input Power | Magnitude of E-field | Magnitude of H-field |
|-------------|----------------------|----------------------|
| 5 dBm       | -58.07 dBm           | -35.62dBm            |
| 7 dBm       | -55.93 dBm           | -32.54 dBm           |
| 10 dBm      | -52.85 dBm           | -29.48 dBm           |
| 1 Vpp       | -59.17 dBm           | -37.54 dBm           |
| 2 Vpp       | -52.88 dBm           | -31.35 dBm           |

**Table 2:** Relation between input power and magnitude of E-field and H-field, after matching circuit is applied.

#### 4. CONCLUSION

In this paper, design of spiral flexible PRFE applicator operating at 27 MHz for wound healing treatment systems has been illustrated with the numerical calculations and experimental measurement. Experimental results are quite well. It has been concluded that the purposed PRFE applicator is compatible with biomedical standards. It has been pointed out that better performance results are obtained fort he purposes PRFE applicator to be used in combination with the matching circuit. Considering the low risk, low cost, and ease of use, the clinical application of PRFE treatment is likely to increase in coming years.

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#### REFERENCES

[1] J. A. Stratton, Electromagnetic theory: John Wiley & Sons, 2007.

- [2] M. Pesce, A. Patruno, L. Speranza, and M. Reale, "Extremely low frequency electromagnetic field and wound healing: implication of cytokines as biological mediators," European cytokine network, vol. 24, pp. 1-10, 2013.
- [3] L. Y. Sun, D. K. Hsieh, P. C. Lin, H. T. Chiu, and T. W. Chiou, "Pulsed electromagnetic fields accelerate proliferation and osteogenic gene expression in human bone marrow mesenchymal stem cells during osteogenic differentiation," Bioelectromagnetics, vol. 31, pp. 209-219, 2010.
- [4] M. Lupke, J. Frahm, M. Lantow, C. Maercker, D. Remondini, F. Bersani, et al., "Gene expression analysis of ELF-MF exposed human monocytes indicating the involvement of the alternative activation pathway," Biochimica et Biophysica Acta (BBA)-Molecular Cell Research, vol. 1763, pp. 402412, 2006.
- [5] O. Orwar, M. Karlsson, D. Chiu, A. Stromberg, and A. Karlsson, "Method and apparatus for manipulation of cells and cell-like structures focused electric fields in microfludic systems and use thereof," ed: Google Patents, 2006.
- [6] M. Mercandetti and A. J. Cohen, "Wound healing: healing and repair," Emedicine. com. Accessed January, vol. 20, p. 38, 2005.
- [7] K. Hug and M. Röösli, "Therapeutic effects of wholebody devices applying pulsed electromagnetic fields (PEMF): A systematic literature review," Bioelectromagnetics, vol. 33, pp. 95-105, 2012.
- [8] Hug, K. and M. Röösli, Therapeutic effects of whole body devices applying pulsed electromagnetic fields (PEMF): A systematic literature review. Bioelectromagnetics, 2012. 33(2): p. 95-105.
- [9] Mercandetti, M., 2005

[10] Kapton Polyimide Film-Summary of Properties, DuPont Co., Polymer Products Department, Industrial Films Division, Wilmington, DE 1989.

- [11] A. Mata, A. J. Fleischman, and S. Roy, "Characterization of Polydimethylsiloxane (PDMS) Properties for Biomedical Micro/Nanosystems," Biomedical Microdevices, vol. 7, no. 4, pp. 281-293, Dec. 2005
- [12] E. Apaydin, "Microfabrication Techniques for Printing on PDMS Elastomers for Antenna and Biomedical Applications," Ohio State University, 2009
- [13] Yoshikawa T, Tanigawa M, 2000



# Estimation of Radiogenic Heat Production in Clay and Pumice Brick Samples Used as Structural Building Materials in Turkey

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Abstract: The radiogenic decay of the unstable radioisotopes supplies the largest internal source of heat. The kinetic energy of the emitted particles (alpha and beta) and the gamma-ray associated with the different radioactive decay processes is absorbed in the rocks and finally transformed into heat. More than 98% of present-day heat production is the result of the decay series of uranium (238U) and thorium (232Th) and the radioactive potassium (40K). In the study, radiogenic heat contributions of clay and pumice brick samples commonly used for wall masonry and/or internal partition walls in constructions in Turkey were determined based on the brick's K, U and Th concentrations and density. The values of radiogenic heat production (RHP) estimated for clay bricks and pumice bricks varied from 0.3 to 1.3  $\mu$ W/m3 with an average of 0.8  $\mu$ W/m3 and 0.8 to 1.7  $\mu$ W/m3 with an average of 1.4  $\mu$ W/m3, respectively. The results indicate that the RHP of pumice bricks is higher than the RHP of clay bricks.

Keywords: Radiogenic heat production, clay brick, pumice brick, building materials, Turkey.

### **1. INTRODUCTION**

The main interior sources of heat in the Earth are strongly depended on the amount and distribution of the decay of unstable radioactive isotopes in the crust. The thermal (heat) structure of the Earth's crust determines its tectonics and its seismicity. Knowledge of the distribution of radiogenic heat production in the Earth is critical for the determination of crustal thermal and rheological structure. The radiogenic decay of the uranium (238U; 235U), thorium (232Th) and potassium (40K) provides the largest internal source of heat (Ehinola et al., 2005). During radioactive decay, mass is converted into energy. The energy emitted by decay processes, consisting of the kinetic energy of the emitted particles and the  $\gamma$ - radiation associated with the different decay processes is absorbed in the rocks and finally transformed into heat (Philip, 2001). Therefore, temperature is one of the most important physical parameters in the place. Despite this, our knowledge of the thermal structure of the ground is not enough (Albarede, 1975; Beaumont et al., 2001; Brady et al, 2006).

In this study, measurements of activity concentrations of Uranium (238U), Thorium (232Th) and Potassium (40K) were done using EDXRF spectroscopy. The activity concentrations are parts per million (PPM) used in the determination of radiogenic heat production of each of the elements and its distribution pattern.

## 2. MATERIALS AND METHODS

For analyses of the clay and pumice brick samples, 25 different samples were collected from the different parts of Turkey. The samples, each about 1 kg in weight, were ground, homogenized and sieved to about 100 mesh screen. The samples were then placed for drying at 110 °C for 24 h to ensure that moisture is completely removed. The analysis survey was conducted using energy dispersive x-ray fluorescence (Spectro Xepos, Ametek). The target changer, with up to 8 polarization and secondary targets, offers many different excitation conditions ensuring the optimum determination of all elements from 11Na to 92U. A spectral resolution of less than 155 eV at Mn  $K\gamma$  is achieved. The samples to be measured were passed through the crusher and sieved. Approximately 3 gr of powdered samples were used for the measurements. For the analysis of the powder samples, the "powder method" registered on the device was used.

If the density of the samples is  $\rho$  and the concentrations in Uranium (CU), thorium (CTh) and potassium (CK) are known, its radiogenic heat generation rate A can be determined using the values given (Shittu et al., 2016; Bubu and Ononugbo, 2017).

 $A[\mu Wm^{-3}] = 10^{-5*}\rho[kgm^{-3}]^*(9.52*C_U[ppm] + 2.56*C_{Th}[ppm] + 3.48*C_K[\%])$ (1)

#### 3. RESULTS AND DISCUSSION

The concentrations of the radionuclides were presented in Table 1. Radiogenic heat productions for clay and pumice brick samples were calculated from the <sup>238</sup>U, <sup>232</sup>Th and <sup>40</sup>K concentration using the formula (1) and presented in Table 1 and 2, respectively.

|  | Table <sup>†</sup> | ۱. | Concentrations | of <sup>238</sup> U, | <sup>232</sup> Th, | <sup>40</sup> K a | nd radio | ogenic | heat | production | in cla | y bricl | c sam | ples o | f Tı | urkey |
|--|--------------------|----|----------------|----------------------|--------------------|-------------------|----------|--------|------|------------|--------|---------|-------|--------|------|-------|
|--|--------------------|----|----------------|----------------------|--------------------|-------------------|----------|--------|------|------------|--------|---------|-------|--------|------|-------|

| Sample ande | <sup>238</sup> U | <sup>232</sup> Th | $^{40}$ K | Radiogenic heat production |
|-------------|------------------|-------------------|-----------|----------------------------|
| Sample code | ppm              | ppm               | (%)       | $(\mu W/m^3)$              |
| CBRICK-1    | 1,6              | 6,4               | 1,5       | 0,5                        |
| CBRICK-2    | 1,6              | 5,6               | 1,1       | 0,5                        |
| CBRICK-3    | 1,5              | 4,0               | 1,0       | 0,4                        |
| CBRICK-4    | 6,7              | 9,7               | 1,6       | 1,3                        |
| CBRICK-5    | 2,4              | 10,4              | 2,4       | 0,8                        |
| CBRICK-6    | 2,3              | 9,4               | 2,0       | 0,7                        |
| CBRICK-7    | 2,1              | 8,4               | 2,2       | 0,7                        |
| CBRICK-8    | 2,2              | 9,7               | 2,3       | 0,7                        |
| CBRICK-9    | 2,0              | 6,8               | 2,2       | 0,6                        |
| CBRICK-10   | 2,3              | 6,7               | 1,9       | 0,6                        |
| CBRICK-11   | 4,0              | 12,6              | 3,5       | 1,1                        |
| CBRICK-12   | 3,1              | 9,9               | 3,0       | 0,9                        |
| CBRICK-13   | 3,1              | 11,6              | 2,8       | 0,9                        |
| CBRICK-14   | 2,2              | 9,1               | 2,9       | 0,8                        |
| CBRICK-15   | 2,0              | 7,2               | 2,3       | 0,6                        |
| CBRICK-16   | 3,0              | 15,7              | 2,8       | 1,1                        |
| CBRICK-17   | 4,6              | 14,3              | 2,0       | 1,2                        |
| CBRICK-18   | 1,3              | 5,5               | 1,2       | 0,4                        |
| CBRICK-19   | 2,9              | 12,3              | 1,6       | 0,9                        |
| CBRICK-20   | 3,4              | 10,5              | 1,3       | 0,9                        |
| CBRICK-21   | 1,1              | 3,6               | 0,8       | 0,3                        |
| CBRICK-22   | 2,7              | 11,5              | 1,6       | 0,8                        |
| CBRICK-23   | 3,8              | 11,3              | 1,3       | 1,0                        |
| CBRICK-24   | 3,2              | 10,8              | 1,4       | 0,9                        |
| CBRICK-25   | 6,2              | 10,0              | 1,2       | 1,2                        |
| Average     | 2,8              | 9,3               | 1,9       | 0,8                        |
| SD          | 0,3              | 0,6               | 0,1       | 0,1                        |
| Min         | 1,1              | 3,6               | 0,8       | 0,3                        |
| Max         | 6,7              | 15,7              | 3,5       | 1,3                        |

For the clay brick samples, the activity concentrations of <sup>238</sup>U ranged from 1.1 to 6.7 ppm. The lowest <sup>238</sup>U activity concentration was measured in sample 21 and the highest <sup>238</sup>U activity concentration was measured in sample 4. The activity concentrations of <sup>232</sup>Th ranged from 3.6 to 15.7 ppm. The highest <sup>232</sup>Th activity concentration was measured in sample 16 and the lowest <sup>232</sup>Th activity concentration was measured in sample 21. The activity concentrations of <sup>40</sup>K ranged from 0.8 to 3.5 %. The highest <sup>40</sup>K activity concentration was measured in sample 11 and the lowest <sup>40</sup>K activity concentration was measured in sample 21.

Table 2. Concentrations of <sup>238</sup>U, <sup>232</sup>Th, <sup>40</sup>K and radiogenic heat production in pumice brick samples of Turkey

| Sampla anda | <sup>238</sup> U | <sup>238</sup> U <sup>232</sup> Th |     | Radiogenic heat production |
|-------------|------------------|------------------------------------|-----|----------------------------|
| Sample code | ppm              | ppm                                | (%) | $(\mu W/m^3)$              |
| PBRICK1     | 7,8              | 10,6                               | 3,6 | 1,4                        |
| PBRICK2     | 7,8              | 10,9                               | 3,8 | 1,4                        |
| PBRICK3     | 6,6              | 10,3                               | 3,4 | 1,3                        |
| PBRICK4     | 6,9              | 9,2                                | 2,1 | 1,2                        |
| PBRICK5     | 8,1              | 12,2                               | 3,6 | 1,5                        |

| PBRICK6  | 6,0 | 11,3 | 2,4 | 1,2 |
|----------|-----|------|-----|-----|
| PBRICK7  | 7,1 | 13,1 | 3,8 | 1,4 |
| PBRICK8  | 7,4 | 13,7 | 3,8 | 1,5 |
| PBRICK9  | 4,1 | 6,5  | 2,8 | 0,8 |
| PBRICK10 | 7,7 | 10,6 | 3,5 | 1,4 |
| PBRICK11 | 6,1 | 8,5  | 3,0 | 1,1 |
| PBRICK12 | 7,7 | 10,2 | 3,7 | 1,4 |
| PBRICK13 | 7,6 | 9,7  | 3,3 | 1,4 |
| PBRICK14 | 7,5 | 14,7 | 3,8 | 1,5 |
| PBRICK15 | 7,8 | 11,1 | 3,7 | 1,4 |
| PBRICK16 | 5,5 | 20,2 | 3,5 | 1,5 |
| PBRICK17 | 5,5 | 20,0 | 3,7 | 1,5 |
| PBRICK18 | 5,6 | 20,3 | 3,8 | 1,5 |
| PBRICK19 | 5,3 | 19,0 | 3,8 | 1,4 |
| PBRICK20 | 5,9 | 20,6 | 3,7 | 1,5 |
| PBRICK21 | 5,7 | 24,4 | 2,8 | 1,6 |
| PBRICK22 | 6,7 | 23,3 | 3,1 | 1,7 |
| PBRICK23 | 6,2 | 22,9 | 2,8 | 1,6 |
| PBRICK24 | 6,2 | 26,0 | 2,8 | 1,7 |
| PBRICK25 | 6,4 | 23,2 | 2,8 | 1,6 |
| Average  | 6,6 | 15,3 | 3,3 | 1,4 |
| SD       | 0,2 | 1,2  | 0,1 | 0,0 |
| Min      | 4,1 | 6,5  | 2,1 | 0,8 |
| Max      | 8,1 | 26,0 | 3,8 | 1,7 |

For the pumice brick samples, the activity concentrations of <sup>238</sup>U ranged from 4.1 to 8.1 ppm. The lowest <sup>238</sup>U activity concentration was measured in sample 9 and the highest <sup>238</sup>U activity concentration was measured in sample 5. The activity concentrations of <sup>232</sup>Th ranged from 6.5 to 26 ppm. The highest <sup>232</sup>Th activity concentration was measured in sample 24 and the lowest <sup>232</sup>Th activity concentration was measured in sample 9. The activity concentrations of <sup>40</sup>K ranged from 2.1 to 3.8 %. The highest <sup>40</sup>K activity concentration was measured in sample 4.

Estimated radiogenic heat production (RHP) values for clay bricks and pumice bricks are varies between 0.3 to 1.3  $\mu$ W/m<sup>3</sup> and on average 0.8  $\mu$ W/m<sup>3</sup>, 0.8 to 1.7  $\mu$ W/m<sup>3</sup> and on average 1.4  $\mu$ W/m<sup>3</sup>, respectively. The results show that the RHPs of pumice bricks are higher than the RHPs of clay bricks.

#### 4. CONCLUSION

The results indicate that the contribution and rate of heat production of 238U, 232Th and 40K in the samples vary significantly with geological locations. In the samples, we cannot make a comparative analysis since radiological heat production is not calculated before. Our data provide a data infrastructure for geologists in radiogenic heat generation. This is important for tectonic movements, hydrocarbon, and geothermal energy in terms of resource evaluation in the working areas.

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#### REFERENCES

- Albarede F (1975). The heat flow/heat generation relationship: an interaction model of fluids within cooling intrusions. Earth and Planetary Science Letters, 27: 73–78.
- Anonymous (2015). Spectro Xepos Brochure, Ametek, http://www.spectro.com/products/xrf-x-ray-fluorescence-spectrometer/edxrf-spectrometer-xepos.
- Bubu A & Ononugbo C P (2017). Radiogenic Heat Production Due to Natural Radionuclides in the Sediments of Bonny River, Nigeria. Journal of Scientific Research & Reports, 17(6): 1-9.
- Beaumont C, Jamieson R A, Nguyen M H & Lee B (2001). Himalayan tectonics explained by extrusion of a low-viscositycrustal channel coupled to focused surface denudation. Nature, 414: 738–742.

- Brady R J, Ducea M N, Kidder S B & Saleeby J B (2006). The distribution of radiogenic heat production as a function ofdepth in the Sierra Nevada Batholith, California. Lithos, 86: 229–244.
- Ehinola EA, Joshua E O, Opeloye S A & Ademola J A (2005). Radiogenic heat production in the cretaceous sediment of Yola arm of Nigeria Benue. Through: Implications for thermal history and hydrocarbon generation. J. Applied Sciences, 5: 696-701.
- Kurnaz A, Turhan Ş, Gezelge M, Hançerlioğulları A & Çetiner M A (2016). Elemental Composition of Soils Mixed with the Grape Molasses. Turkish Journal of Agriculture Food Science and Technology, 4(9): 748-751.

Philip A O (2001). An introduction to geophysical exploration. McGraw-Hill, New York.

Shittu A, Hankouraou S & Ziyadat H (2016). Determination of radioactivity concentration and annual ommitted effective dose in drinking water collected from Local Borehole in Gombe, Nigeria. Sch. J. Phys. Math. Stat. 3(2): 56-65.



## Pulsed Electromagnetic Field Application with High-Resolution Wireless Sensor Network

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**Abstract:** In this paper, an efficiency improved Pulsed Electromagnetic Field (PEMF) system design is provided for PEMF applications, operating in an extremely low-frequency range compared to current treatment systems. In order to build the PEMF device firstly, computer-aided 3D electromagnetic simulations and the distribution of the magnetic field created by designing coil geometrical structure have examined. The coil that can be capable of generating a homogeneous magnetic field and the resulting magnetic field distribution is computed by numerical computation CST Studio. A homogeneous magnetic field has advantages on wound healing applications. In the designed PEMF device operating frequency and amplitude of excited PEMF signal waveform can be changed.

Keywords: Pulsed electromagnetic field, wound healing, low frequency, coil

### **1. INTRODUCTION**

Wound healing is a physiological reaction of our body against tissue damage. This reaction is not a simple phenomenon, it involves a complex interaction between many cell types, cytokines, mediators and vascular systems [1]. Pulsed magnetic fields (PEMF) can be described as non-ionizing, low frequency electromagnetic field irradiation [2]. In this application, signals ranging from 1 mT to 3 mT magnetic field is used, operating at frequencies up to 75 Hz, containing different waveforms with extremely low frequency. The application of pulsed electromagnetic field radiation to the target tissue causes physiological changes in the cellular region and thus facilitates the wound healing process. [3]. PEMF has a variety of effects on cell proliferation, cell differentiation, cell cycle, apoptosis and DNA division [4]. Studies have shown that PEMF signals with a magnetic field size of less than 3 mT and frequency less than 100 Hz have been found to be more effective [5].PEMF increases oxygen, stimulates blood circulation, stimulates adenosine triphosphate production, increases membrane permeability and cell metabolism, accelerates cellular curing, improves communication between cells, improves ion transfer in cells [6]. PEMF therapy is an electromagnetic field generating electromagnetic field and is usually used as an adjunctive therapy. Different operating frequencies are used in pulsed electromagnetic field technology [8].

The most important problem in PEMF therapies is that the correct frequency and magnitude of the magnetic field cannot be determined accurately. The designed PEMF device in this project is examined in order to determine the best parameters with biological trials and computer simulations covering the frequency range mentioned above.

## 2. MATERIAL AND METHODS

Coil structures are developed by means of a computer-aided 3D electromagnetic simulator CST Studio. After the preparation of the prototype, biological experiments are performed in In Vitro cell culture studies within the scope of laboratory applications. The input signal frequency and amplitude of the device are changeable and there is a waveform generator with selectable waveforms of sine, triangle, and square. The output of this generator is connected to the amplifier. The project has required an amplifier compatible with the changing frequency and amplitudes of the waveform generator. The output of the amplifier is connected to the coil. Thanks to the sensors and wireless module, we get the data from the biological samples.

## 3. RESULTS AND DISCUSSION

### **PEMF** Applicator Design

A magnetic coil applicator is designed with CST Studio program.



Figure 1: Geometry of Magnetic coil on CST Studio





Figure 2: (a) Geometry of Magnetic coil on CST Studio, (b) B-Field Result of the coil on CST Studio

## **Coil Experimental Results**

In this study, a coil is generated by the winding of copper wire 150 times. After generating the coil, experimental results are obtained by measuring current and voltage values from the output of the coil by using a multimeter and by measuring the magnetic flux density generated by the coil on the computer by using the PASCO Capstone program. Other parameters of the generated coil are shown in Table 1 and the experimental results of the coil for different input AC voltage values are shown in Table 2.

| Parameter              | Symbol      | Value |
|------------------------|-------------|-------|
| Number of<br>Turns     | Ν           | 150   |
| Resistance             | $R(\Omega)$ | 10.3  |
| Inductance             | mH          | 10,45 |
| Operating<br>Frequency | f(Hz)       | 75    |

Table 1: Parameter list of the generated coil



Figure 3: Measurement Setup

 Table 2: Measurement Results of the Generated Coil

| Input Voltage            | Frequency | Coil Voltage | Coil Current | Magnetic flux density |  |
|--------------------------|-----------|--------------|--------------|-----------------------|--|
| (Volts- <sub>RMS</sub> ) | (Hertz)   | (Volts)      | (Amperes)    | (Millitesla)          |  |
|                          |           |              |              |                       |  |
| 3                        | 75        | 47,00        | 0,34         | 0.95                  |  |
| 5                        | 75        | 69,00        | 0,51         | 1.50                  |  |
| 7                        | 75        | 89,00        | 0,66         | 1.95                  |  |

## Experimental Results of Exposure of Magnetic Field on a Cell Culture

In this study, the magnetic field generated by the coil is exposed to a cell culture that includes tissue samples. Changes versus time on magnetic flux density that absorbed by the cell culture and temperature values are observed by means of a magnetic Hall Effect sensor and a temperature sensor. Sensor data is taken with a wireless module and shown on the computer screen. Experimental results for different input AC voltage values are shown in table 3.

 Table 3: Magnetic Hall Effect sensor and temperature sensor data results

| Input Voltage<br>(Volts-RMS) | Time<br>(Seconds) | Temperature<br>(Celsius) | Magnetic Flux<br>Density<br>( Millitesla) |
|------------------------------|-------------------|--------------------------|---|
| 3                            | 15                | 29.10                    | 0.82                                      |
| 3                            | 30                | 29.15                    | 0.85                                      |
| 3                            | 45                | 29.18                    | 0.88                                      |
| 3                            | 60                | 29.26                    | 0.88                                      |
| 5                            | 15                | 29.10                    | 1.00                                      |
| 5                            | 30                | 29.20                    | 1.05                                      |
| 5                            | 45                | 29.23                    | 1.07                                      |
| 5 | 60 | 29.30 | 1.12 |
|---|----|-------|------|
| 7 | 15 | 29.23 | 1.40 |
| 7 | 30 | 29.30 | 1.44 |
| 7 | 45 | 29.52 | 1.50 |
| 7 | 60 | 29.63 | 1.53 |

### 4. CONCLUSION

It has been proven that when the electromagnetic wave is emitted at low frequency, there are some electrical changes at the tissue level. This provides the possibility of being used as an adjunctive treatment in medical applications such as wound healing, bone healing, and pain relief. In that designed project it's observed that simulation and measurement results are close to each other. Sensor data showed the temperature is nearly constant by the time, and magnetic flux density absorbed by the cell culture by the time is nearly homogeneous as needed.

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### REFERENCES

- Bowden LG, Byrne HM, Maini PK, Moulton DE. A morphoelastic model for dermal wound closure. Biomech Model Mechanobiol. 2016 Jun;15(3):663-81
- [2] Crocetti, S., et al., Low intensity and frequency pulsed electromagnetic fields selectively impair breast cancer cell viability. PloS one, 2013. 8(9): p. e72944.
- [3] B. F. Sisken, J. Walker, "Therapeutic aspect of electromagnetic fields for soft tissue healing. Electromagnetic Fields: Biological interactions and mechanisms", *Journal of Advance in Chemistry*
- [4] Wang, Q., et al., [Effect of pulsed electromagnetic field with different frequencies on the proliferation, apoptosis and migration of human ovarian cancer cells]. Sheng wu yi xue gong cheng xue za zhi= Journal of biomedical engineering= Shengwu yixue gongchengxue zazhi, 2012. 29(2): p. 291-295.
- [5] Hug, K. and M. Röösli, Therapeutic effects of whole-body devices applying pulsed electromagnetic fields (PEMF): A systematic literature review. Bioelectromagnetics, 2012. 33(2): p. 95-105.
- [6] Markov, M.S., *Expanding use of pulsed electromagnetic field therapies*. Electromagnetic Biology and Medicine, 2007. **26**(3): p. 257-274.
- [7] Dr. Garry F. Gordon, The Science of PEMF, Gordon Research Institute, october 27, 2012, The Woodland of Van Buren Wayne, Michigan
- [8] McCarthy, C.J., M.J. Callaghan, and J.A. Oldham, *Pulsed electromagnetic energy treatment offers no clinical benefit in reducing the pain of knee osteoarthritis: a systematic review.* BMC Musculoskeletal Disorders, 2006. **7**(1): p. 51.
- [9] Costin, G.-E., S. A Birlea, and D. A Norris, *Trends in wound repair: cellular and molecular basis of regenerative therapy using electromagnetic fields*. Current molecular medicine, 2012. **12**(1): p. 14-26.



# Evaluation of Different Alternatives for the Intersection of Erzincan Province Training and Research Hospital with Microsimulation Modeling

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**Abstract:** While the number of registered vehicles in Erzincan is approximately 49,000 in 2015, this figure has increased to 59.000 by 2018. The city, which has a population of 139,000, has been one of the cities with the highest population / vehicle . In the current traffic systems, the delay times increased, the densities increased and the blockages started to show. As a result of increasing number of vehicles and transportation demands, there has been a need to improve the intersection and existing access roads throughout the city.

In the light of this information; Especially, the intersection of Erzincan Mengücek Gazi Training and Research Hospital with 500 bed capacity was taken into consideration. During the peak periods, 676 vehicle roadside parking spaces were used in the hospital, which is the center of attraction of the city. Expectations and needs of the users were put forward and the intersections were simulated with the Microscopic Simulation Method using AIMSUN program. The most appropriate intersection type has been determined by taking into consideration the factors such as delay, travel time, and stop time and applicability.

Keywords: Traffic, Intersection, AIMSUN, Microsimulation, Erzincan



# Interaction of 2-hydorxy-juglone with Different Aldehydes in the Presence of Hantzsch Ester

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Abstract: Juglone - a naphthoquinone compound present in high amounts in the plants of the Fam. Juglandaceae. Especially high amounts can be detected in the eastern black walnut (Juglans nigra) and common walnut (Júglans régia). Juglone is an allelochemical produced by the plants to inhibit the growth of other species. Ancient Greek and Roman civilizations used walnuts as herbicides. It is well known that juglone has a wide spectrum of biological activity, including antibacterial and antifungal properties. Juglone is widely used in traditional medicine in the form of tincture, balsam, syrup, as general tonic, immunomodulatory, blood sugar regulatory, hipotensive and antiparasitic remedy. Juglone containing ointments are used as anti-inflammatory, antibacterial and antifungal remedies. Anticancer properties have been also detected for juglone by some researchers. The main objective of the present research was investigation of the reaction between 2-hydroxy-juglone with different aldehydes with obtaining of the new derivatives of 1,4naphthoquinone. Reaction has been performed in the presence of the Hantzsch ester as a reducing agent and L-proline as catalyst. In the case of the aromatic aldehydes, the coupling with 2-hydroxy-juglone took place with formation of the products with the yields between 70-85%. The structures of the obtained compounds have been confirmed with application of different physico-chemical methods of analysis. In the case of the citronellal, the reaction results in the formation of two compounds. The compounds have been individually separated with application of the column chromatography. The yield of the main product with opened chain was > 20%. The identity of the compounds has been established by various 1H-, 13C-NMR experiments including those bi-dimensional (COSY, HSQC, HMBC, NOE).

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**Keywords:** Organic chemistry, organic synthesis, juglone, 5-hydroxy-1,4-naphthoquinone, 2,5-dihydroxy-1,4-naphthoquinone, Hantzsch ester



# Supercritical Fluids: A Sustainable Energy for Novel and Safe Dried Fruits and Vegetables

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**Abstract:** In food technology, researches work hard to find new technologies which could substitute the conventional industrial processes and could achieve their various purposes. Mainly, these researches look forward to reduce the environmental threatening, to alleviate the usually encountered negative effects during and after processing of foodstuffs, and for the obtainment of novel product to meet the needs of expanding food markets. Supercritical fluid technology is one of the new researched subjects for the development or even the replacement of existing technologies. Since the used substances (CO<sub>2</sub>) are inexpensive and not toxic, the supercritical fluid technology is considered as an economical and green technology which is ready to invade several applications. As it is known, drying is among the thermal processes applied for the preservation of food and increasing its shelf life. Because of the habitual thermal drawbacks of drying, many studies have tried to apply the supercritical fluids for the dehydration of foodstuffs. In order to know the latest studies done on the dehydration using this non-thermal technology, it was reviewed here in this paper the main supercritical drying processes and the main obtained results for fruits and vegetables. It was concluded that this technology till now reached an important development, due to its positive effects in reducing the microbial charge, to preserve the nutritional value and the bioactivity of dried food. Despite all of that, the investigations on supercritical fluid technology should continue to make clear all its advantages, even disadvantages, and conditions for more and large scale of drying applications in the food industry.

Keywords: Novel technology, CO2, drying, fruits and vegetables.

### **1. INTRODUCTION**

The recovery of gases causing global warming is one of the most important issues. Nowadays, ongoing technologies are working hard to reduce gas emissions.  $CO_2$  is one of the many gases emitted from many sources, such as industrial and domestic emissions.  $CO_2$  from emission gases can be separated, and stored in underground or oceans (Benson and Orr, 2008).  $CO_2$  is used in various industrial applications. For instance, in food industry, it is added in beverages and employed for modified packaging applications (Mazzotti et al., 2005). The compressed  $CO_2$  is also used as supercritical fluids (SCFs) (Chapel et al., 1999).

Investigations of the last decades showed that there have been many applications of SCF which might be used in food industry, oil processing (extraction, fractionation, refining), extraction of aromas, extraction of bioactive compounds, decaffeination of coffee beans, and removal of some compounds and contaminants from food materials (alcohols, pesticides) or for the encapsulation of liquids and for analytical purposes (chromatography) (Palmer & Ting, 1995; Lehotay, 1997, Brunner, 2005). In order to reduce the negative effects of some technologies in the food industry, SCF has been used for different purposes in recent years. SCFs can be an alternative technique that could decrease the thermal drawbacks, for example during the drying process of some food materials (fruits, vegetables, meats, gels etc.).

Drying or dehydration is an indispensable process in food industry to increase the shelf life of perishable food, to keep the maximal quality, and to lower the cost of shipping and packaging. As mentioned before, the usual thermal treatments used in drying negatively influence the sensitive foodstuffs. The changes during drying are chemical (browning, color loss, lipid oxidation etc.), physical (texture, rehydration, solubility etc.) and nutritional properties (microbial survival, protein and vitamin loss etc.) (Chou & Chua, 2001). The non-thermal emerging technologies, like SCFs, become increasingly important in producing novel foods by minimizing the influencing factors to keep the original composition of food and to increase its quality (decreasing its microbial load) (Smigic et al., 2019). The aim of this review is to mention all the recent studies done on the dehydration of fruits and vegetables using SCF drying process.

### What Is a Supercritical Fluid?

A supercritical fluid is a substance where its temperature and pressure are above its thermodynamic critical point. After the point, the fluids are in a state in which no more liquid-vapor equilibrium would exist. Generally, supercritical fluids have high pressure and different levels of temperatures depending on the type of substance (Fukushima, 1999; Savage. et al., 1995; Marta Vazquez, 2010). Figure 1 displays the critical point of CO<sub>2</sub> fluid. Also, Table 1 shows critical point of some fluids.



Figure 1. Phase (Pressure-Temperature) diagram for CO2 (Khalloufi et al., 2010)

| Table 1. Critical | points of some | fluids ( | Savage et al  | 1995) |
|-------------------|----------------|----------|---------------|-------|
| LUDIC L. CITTICU  | points of some | manab (  | ouvage et an, | 1)))) |

| Fluid          | T °C   | P atm | Fluid     | T ℃    | P atm |
|----------------|--------|-------|-----------|--------|-------|
| Ethylene       | 8.85   | 49.7  | l-hexene  | 230.85 | 31.3  |
| Xenon          | 15.85  | 57.6  | t-Butanol | 232.85 | 39.2  |
| Carbon dioxide | 30.85  | 72.8  | n- Hexane | 233.85 | 29.3  |
| Ethane         | 31.85  | 48.2  | Acetone   | 234.85 | 46.4  |
| Nitrous oxide  | 35.85  | 71.5  | Methanol  | 238.85 | 79.9  |
| Propane        | 95.85  | 41.9  | Ethanol   | 242.85 | 63.0  |
| Ammonia        | 131.85 | 111.3 | Toluene   | 317.85 | 40.6  |
| Methyl amine   | 156.85 | 73.6  | Water     | 373.85 | 217.6 |

\*T: Critical temperature, P: Critical pressure

SCFs exhibit intermediate physical and chemical properties between liquids and gases. Since they have gas-like diffusivity and viscosity properties, they present a high density comparable to that of liquids. These properties enable to use the SCFs in divergent applications and industries (i.e as solvent for extraction of specific compounds). Table 2 shows the characteristics of the three different state (Fukushima, 1999; Marta Vazquez, 2010).

|                                     | Liquid | SCF       | Gas  |
|-------------------------------------|--------|-----------|------|
| <b>Density</b> (g/cm <sup>3</sup> ) | 1      | 0.1 -0.5  | 10-3 |
| Viscosity(Pa·s)                     | 10-3   | 10-3 10-5 | 10-5 |
| Diffusivity(cm <sup>2</sup> /s)     | 10-5   | 10-3      | 10-1 |

Thanks to their properties, solubilization and transport, and low surface tension, which classify SCFs as a good alternative against the traditional solvents used for the extraction and separation of organic and non-organic compounds. Since the 60th and 70th of the last century, researches and industries have integrated these fluids in various technical processes like extraction, chromatography, chemical reaction, drying, cleaning, waste treatment (Fukushima, et al 1999; Marta Vazquez, 2010).

The drying is considered to be a way of extraction of water from different type of matrixes (food, tissue, gels etc.). Regardless the purpose of drying; preparing sample for electron microscopy scanning, production of aerogels, foods conservation, etc., SCF could be one of the novel techniques to reach these targets (Figure 2) (Williams et al., 2002; Knez et al., 2014).

CO2 is widely used as a supercritical fluid, due to its advantages following:

- it has a critical point (31.3 °C, 72.9 atm) at low temperature, and is appropriate for the biological treatments with a limited thermal effect.

- CO<sub>2</sub> is non-toxic, non-flammable, non-mutagenic, non-corrosive, readily available and also inexpensive.

- It is considered as a "green technology" for being environmentally friendly and sustainable technology (Savage et al., 1995; Marta Vazquez, 2010; Knez et al., 2014).

The rising of interest in applying  $SCCO_2$  in drying methods and removing liquids, might be resulted from the aforementioned advantages. After treatment only at atmospheric conditions,  $SCCO_2$  can be easily removed from the dryed food without damaging the initial structure (Williams et al., 2002; Knez et al., 2014).



Figure 2. Basic flow sheet of supercritical fluid extraction plant (Hrnčič et al., 2018)

# **Regeneration of Scfs in Drying Systems**

The continuous driving of water out of solid food matrices, leads to saturate the SCF by water. In order to regenerate and recover the fluid for more extraction and drying process, the water has to separate. The reduction of fluids density, by the decreasing of pressure and/ or the increasing of temperature, results simply in the separation of extract (water) from the fluid (Figure 2) (Palmer & Ting, 1995). Recently, there are some studies about the regenerating of water-extracting agent by means of dense polymeric membranes separating water from  $CO_2$ . In this process, as a theory, the humidified SCF after leaving the water-extraction unit enters the membrane compartment (parallelly arranged flat membranes), the SCF gets dehydrated and can be reused for another cycle of drying process (Shamu et al., 2019a; Shamu et al., 2019b). The Figure 3 summarizes the principle steps of water-extraction, dehydration and regeneration of SCFs.



**Figure 3.** Schematic view of the principle steps of water-extraction, dehydration and regeneration of SCFs (Shamu et al., 2019b)

### **Supercritical Drying Studies**

The uses of supercritical fluids are being expanded in many industries to fulfill the various purposes of the application. The drying processes for the production of dried foodstuffs have been tested for the last years. Table 3 summarizes some of the studies done on the drying fruit and vegetables.

| Material  | Condition  | Purposes  | Results  | Reference  |
|---|--|---|--|--|
|   | of drying  |   |  |  |
| Wet Okara<br>(Soybean<br>pulp)<br>Fresh<br>apples<br>(Golden<br>Delicious<br>Variety) | T (40°C),<br>P (103 bar),<br>t (10-60 h),<br>CO2 Flow<br>rate<br>(20ml/min)<br>T (35°C),<br>P (10 MPa),<br>Ethanol | To investigate the drying kinetics<br>and water extraction efficiency of<br>SCF at the used conditions.<br>To determine the effect of sample<br>loading on the drying (<500mg<br>and >1000mg).<br>To investigate the potential of<br>SCCO <sub>2</sub> drying of apples using both<br>pure and ethanol-modified SCCO <sub>2</sub> .<br>To analyze drying profiles, color<br>and microstructure of the final<br>product. | The feasibility of drying high<br>moisture particulates such as Okara<br>using SCCO <sub>2</sub> even at low<br>temperatures.<br>The water uptake efficiency of<br>SCCO <sub>2</sub> is higher at high mass loading.<br>The drying profiles of pure SCCO <sub>2</sub><br>showed lower degree of dehydration.<br>The changes in color $\Delta E$ did not<br>shown significant differences<br>between the two dried samples. | Sengupta<br>et al., 2012<br>Vetralla et<br>al., 2018 |
| Red bell<br>peppers,<br>coriander,<br>strawberry<br>, apple                           | T (40-<br>60°C),<br>P (100-140),<br>t (16 h),<br>CO2 F<br>(150kg /h)   | To explore the use of SCCO <sub>2</sub><br>drying as alternative technique<br>for the obtainment of pasteurized<br>and high-quality dried product.  | An unaltered nutritional value,<br>flavonoids contents are in the range of<br>fresh food.<br>Also, the SCCO <sub>2</sub> is able to preserve<br>the ascorbic acid.<br>SCCO <sub>2</sub> is able to complete inactivate<br>yeasts and molds.<br>Bacteria could be inactivated up to 8<br>log.   | Zambon et<br>al., 2018b                              |
| 'Elstar'<br>apple   | T (50 °C),<br>P (12.5<br>MPa),<br>t (16 h)   | To investigate the effects of the SCCO <sub>2</sub> -drying technique on changes in sensory properties and some secondary metabolites of dried apple cuts/snacks during twelve months of storage at 'room' temperature.   | The results for the acceptance level of<br>freeze-dryed apple sample were<br>compatible with those of SCCO <sub>2</sub><br>dryed sample during storage<br>SCCO <sub>2</sub> drying has shown less lost<br>than air-drying for secondary<br>metabolites   | Tomic et<br>al., 2019                                |

Table 3. The use of supercritical drying for fruit and vegetables

\**T*= Temperature, *P*=Pressure, *t*= time *F*= Flow rate

# **Microbial Inactivation**

The SCCO<sub>2</sub> drying is not only to obtain a product with less water content, but also reducing the probability of microbial spoilage. Such fluids may provide microbial inactivation and significantly reduce microorganisms. In a study, Boudeaux et al. (2018) have investigated about the effects of SCCO<sub>2</sub> and freeze-drying methods on the microbial load of coriander inoculated with *Salmonella*, *Listeria monocytogenes* and *Escherichia coli O175: H7* before drying. They found that the microbiological reductions in coriander dried by SCCO<sub>2</sub> were 4 log better more than those of freeze-drying, and the yeasts and molds were almost absent.

Also, Zambon et al. (2018a) reported that the reduction could be up to 4 log for mesophilic bacteria and high inactivation of yeasts and molds. In another study about coriander, Michelino et al. (2018) had demonstrated that the combination of high-power ultrasound (HPU) with SCCO<sub>2</sub> (Figure 4) could be more efficient than SCCO<sub>2</sub> treatment alone at the inactivation of microorganisms.

Koubaa et al. (2018) have reviewed the studies in the agro-food industry that use ultrasound-assisted SCF technology. The studies have generally demonstrated that the technology was efficient for microbial inactivation and pasteurization of many foodstuffs (juice, fresh cut fruits, meats) in a short time of treatment.



**Figure 4.** HPU+SCCO2 drying reactor. The drying chamber is below, while the HPU sonotrode is above. (Michelino et al., 2018)

# Phytochemicals and Antioxidant Activity

Braeuer et al. (2017), who studied the  $SCCO_2$  assisting drying of persimmon and mango slices under Raman sensors, observed that the carotenoids intensity were seen only a negligible variation during the process time. The phytochemicals (flavonoids, chlorogenic and gallic acid derivatives) of the coriander dried by  $SCCO_2$  and air-drying techniques was compared. As a result, there were no significant differences between both techniques in point of view of polyphenols retention (Zambon et al., 2018a).

Bušić et al.(2014), studied the bioactive components and antioxidant properties of basil samples dried bysupercritical drying, air drying and freeze-drying techniques. Due to the prolonged drying time (4h) of the supercritical drying, phenolic compounds and antioxidant capacity were negatively affected. Furthermore, they have indicated that ascorbic acid for some basil samples under suitable conditions (time, pressure) could be preserved similarly to freeze-dried ones.

However, antioxidant activity for freeze-drying were undergone the least decreasing comparing to sypercritical drying. Also, the application of this non-thermal technology for decreasing microorganisms, has minimized the loss of nutrients and flavors in the foodstuffs during the process (Koubaa et al., 2018).

### **Microstructure and Texture**

Lee et al. (2011) studied apple slices pretreated with  $SCCO_2$  and dried at 70°C, then dried samples have visualized in SEM. They observed more pore distribution and shrinking in the samples comparing with the control samples. Djekic et al. (2018) reported that  $SCCO_2$  drying could have an effect on apple slices, have show a partial deformation of shape and appearance of cracks on the surface. However, in a study, the air-dried carrots had less porous structure and more density than the samples treated  $SCCO_2$  giving more original structure and volume retention (Brown et al., 2008).

### Color

Djekic et al. (2018) have dried apple samples by three types of drying (freeze -drying, air and supercritical drying). The highest color change ( $\Delta E$ ) were encountered in air-dried apples slices after treatment and during storage. Brown et al. (2008) determined that the carrot samples dried by SCCO<sub>2</sub> and freeze- drying had lower color change and browning index. On the other hand, the color and chlorophylls content of dried basil samples by freeze-drying were better than the supercritical and conventional drying techniques (Bušić et al., 2014). Vetralla et al. (2018) suggested that the treatment of apple slices by SCCO<sub>2</sub> had inactivated polyphenol oxidase, which significantly gave treated slices higher lightness compared to non-treated ones (Figure 5).



Figure 5. Images of apple pieces untreated (A), EtOH- SCCO2 (B), and SCCO2 (C) (Vetralla et al., 2018)

### **Dehydration rate**

 $SCCO_2$  as a pretreatment for drying of apple slabs has shown fast dehydration rate comparing to the non-treated slabs. Furthermore, for an optimum rate, the increase in pressure demonstrated to be more functional than temperature increasing (Lee et al., 2011). However, Brown et al. (2008), demonstrated that the increase of temperature from 40 to 60 °C has increased the drying rates for the both supercritical and air drying. The drying kinetics obtained by Raman sensors were initially slow, after that fast and again slow towards the end of dehydration processes (Braeuer et al., 2017). In general, the drying with supercritical fluid is found to be longer than the air drying. This situation was primarily due to the water concentration difference and secondly to the convection phenomenon (Khalloufi et al., 2010).

Moreover, the inclusion of ethanol as a co-solvent has proved its action to increase considerably the drying rate of  $SCCO_2$  and to shorten the time of treatment (Figure 6) (Brown et al., 2008; Brown et al., 2010). In which, a flow rate of 11/min with 6 mol % ethanol reduced the drying time from 300 (in pure  $SCCO_2$ ) to 210 min (Brown et al., 2010). Additionally, a combination of supercritical fluids with HPU could make drying time shorter and enhance the removal of water (Michelino et al., 2018).



**Figure 6.**-Drying profiles for raw carrot pieces dried in air ( $\blacktriangle$ ), pure SCCO2( $\blacksquare$ ) and modified SCCO2 with ethanol ( $\circ$ ). All experiments were carried out at 20 MPa and 50 °C (Brown et al., 2008)

These enhancements can be realized as aforementioned by the new creation of new pores and channels, and basically due to the supercritical properties (diffusion coefficient). However, in contrary to air drying, before the supercritical drying, the application of cooking for carrots, which decreases the tortuosity and increases the diffusivity through the destroyed

cellular structure, has no effects on the rate of water removal (Brown et al., 2008). Despite the controversial action of temperature and relatively long time of treament, the supercritical drying could be an effective process of drying and much faster if it will be modified and/ or hybrided with other systems (co-solvents, HPU etc.).

### 2. CONCLUSION

The dehydration processes of some fruits and vegetables by supercritical fluids are very promising for this green technology. Frequently, supercritical fluids are presented by  $CO_2$ , which is considered as non-toxic, inexpensive and environment- friendly substance. Generally, the application of  $SCCO_2$  in drying has shown important microbial inactivation (bacteria, yeast, molds). The nutrients and bioactive compounds are more preserved comparing with the conventional process (thermal process). Also, the microstructure and color of dried fruits are more retained. However, the dehydration rate is relatively slow if it is not combined or modified with other technologies. Finally, more investigations are needed to unveil all the positive contributions and the negative actions that supercritical fluid application could bring in processing and drying of these novel foods.

#### REFERENCES

Benson, S. M., & Orr, F. M. (2008). Carbon dioxide capture and storage. MRS bulletin, 33(4), 303-305.

- Braeuer, A. S., Schuster, J. J., Gebrekidan, M. T., Bahr, L., Michelino, F., Zambon, A., &Spilimbergo, S. (2017). In Situ Raman Analysis of CO2-Assisted Drying of Fruit-Slices. Foods, 6(5), 37.
- Brown, Z. K., Fryer, P. J., Norton, I. T., Bakalis, S., &Bridson, R. H. (2008). Drying of foods using supercritical carbon dioxide— Investigations with carrot. Innovative food science & emerging technologies, 9(3), 280-289.
- Brown, Z. K., Fryer, P. J., Norton, I. T., &Bridson, R. H. (2010). Drying of agar gels using supercritical carbon dioxide. *The Journal of Supercritical Fluids*, 54(1), 89-95.
- Brunner, G. (2005). Supercritical fluids: technology and application to food processing. Journal of food engineering, 67(1-2), 21-33.
- Bušić, A., Vojvodić, A., Komes, D., Akkermans, C., Belščak-Cvitanović, A., Stolk, M., &Hofland, G. (2014). Comparative evaluation of CO2 drying as an alternative drying technique of basil (Ocimumbasilicum L.)—The effect on bioactive and sensory properties. *Food research international*, 64, 34-42.
- Chapel, D. G., Mariz, C. L., & Ernest, J. (1999, October). Recovery of CO2 from flue gases: commercial trends. In *Canadian society* of chemical engineers annual meeting (Vol. 4). Saskatchewan Canada.
- Chou, S. K., & Chua, K. J. (2001). New hybrid drying technologies for heat sensitive foodstuffs. *Trends in Food Science & Technology*, 12(10), 359-369.
- Djekic, I., Tomic, N., Bourdoux, S., Spilimbergo, S., Smigic, N., Udovicki, B., ... & Rajkovic, A. (2018). Comparison of three types of drying (supercritical CO2, air and freeze) on the quality of dried apple–Quality index approach. *LWT*, 94, 64-72.
- Fukushima, Y. (1999). Application of supercritical fluids. R&D Review of Toyota CRDL, 35(1), 1-9.
- Hrnčič, M. K., Cör, D., Verboten, M. T., &Knez, Ž. (2018). Application of supercritical and subcritical fluids in food processing. Food Quality and Safety, 2(2), 59-67.
- Khalloufi, S., Almeida-Rivera, C., &Bongers, P. (2010). Supercritical-CO2 drying of foodstuffs in packed beds: Experimental validation of a mathematical model and sensitive analysis. *Journal of food engineering*, 96(1), 141-150.
- Knez, Ž., Markočič, E., Leitgeb, M., Primožič, M., Hrnčič, M. K., &Škerget, M. (2014). Industrial applications of supercritical fluids: A review. *Energy*, 77, 235-243.
- Koubaa, M., Mhemdi, H., &Fages, J. (2018). Recovery of valuable components and inactivating microorganisms in the agro-food industry with ultrasound-assisted supercritical fluid technology. *The Journal of Supercritical Fluids*, 134, 71-79.
- Lee, B. S., Choi, Y. H., & Lee, W. Y. (2011). Drying Characteristics of Apple Slabs after Pretreatment with Supercritical CO 2. *Preventive Nutrition and Food Science*, *16*(3), 261-266.
- Lehotay, S. J. (1997). Supercritical fluid extraction of pesticides in foods. Journal of chromatography A, 785(1-2), 289-312.
- Marta Vazquez da Silva. (2010). Supercritical fluids and its applications. *Recent progress in chemical engineering*, 293-312. https://paginas.fe.up.pt/ceft/pdfs/MVazquez RPCE2010.pdf. Visited in 27/7/2018.
- Mazzotti, M., Abanades, J. C., Allam, R., Lackner, K. S., Meunier, F., Rubin, E., ... & Zevenhoven, R. (2005). Mineral carbonation and industrial uses of carbon dioxide. *IPCC special report on carbon dioxide capture and storage*, 319-338.
- Meterc, D., Petermann, M., & Weidner, E. (2008). Drying of aqueous green tea extracts using a supercritical fluid spray process. *The journal of supercritical fluids*, 45(2), 253-259.
- Michelino, F., Zambon, A., Vizzotto, M. T., Cozzi, S., &Spilimbergo, S. (2018). High power ultrasound combined with supercritical carbon dioxide for the drying and microbial inactivation of coriander. Journal of CO2 Utilization, 24, 516-521.

- Palmer, M. V., & Ting, S. S. T. (1995). Applications for supercritical fluid technology in food processing. *Food chemistry*, 52(4), 345-352.
- Savage, P. E., Gopalan, S., Mizan, T. I., Martino, C. J., & Brock, E. E. (1995). Reactions at supercritical conditions: applications and fundamentals. *AIChE Journal*, *41*(7), 1723-1778.
- Sengupta, S., Chakraborty, M., Bhowal, J., & K Bhattacharya, D. (2012). Study on the effects of drying process on the composition and quality of wet okara. *In: 15th International Conference on Sustainable Energy Technologies*, 2016, Singapore, pp.1-7
- Shamu, A., Miedema, H., Metz, S. J., Borneman, Z., &Nijmeijer, K. (2019b). Mass transfer studies on the dehydration of supercritical carbon dioxide using dense polymeric membranes. Separation and Purification Technology, 209, 229-237.
- Shamu, A., Dunnewold, M., Miedema, H., Borneman, Z., &Nijmeijer, K. (2019a). Permeation of supercritical CO2 through dense polymeric membranes. *The Journal of Supercritical Fluids*, 144, 63-70.
- Smigic, N., Djekic, I., Tomic, N., Udovicki, B., & Rajkovic, A. (2019). The potential of foods treated with supercritical carbon dioxide (sc-CO2) as novel foods. *British Food Journal*.
- Tomic, N., Djekic, I., Zambon, A., Spilimbergo, S., Bourdoux, S., Holtze, E., ... &Udovicki, B. (2019). Challenging chemical and quality changes of supercritical Co2 dried apple during long-term storage. *LWT*.
- Vetralla, M., Ferrentino, G., Zambon, A., & Spilimbergo, S. (2018). A Study about the Effects of Supercritical Carbon Dioxide Drying on Apple Pieces. *International Journal of Food Engineering*, 4(3).
- Williams, J. R., Clifford, A. A., & Al-Saidi, S. H. (2002). Supercritical fluids and their applications in biotechnology and related areas. *Molecular biotechnology*, 22(3), 263.
- Zambon, A., Michelino, F., Bourdoux, S., Devlieghere, F., Sut, S., Dall'Acqua, S., ... & Spilimbergo, S. (2018b). Microbial in activation efficiency of supercritical CO2 drying process. Drying Technology, 1-6.
- Zambon, A., M. Vizzotto, T., Morbiato, G., Toffoletto, M., Poloniato, G., Dall'Acqua, S., De Bernard, M... Spilimbergo, S. (2018a). Supercritical CO2 drying of food matrices. In: 21st International Drying Symposium. 11-14 Sept, València, Spain, pp. 17-23



# **Epigenetic Approach to Nutrigenomics and Cancer**

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Abstract: Nutrigenomics is a multidisciplinary science consisting the information about how food and bioactive compounds inside them affect expression of genes. Epidemiologic and clinical studies revealed that dietary factors are related to cancer formation and development. Whereas dietary mutagens cause mutations in genes taking place many biological pathways; many nutrients and their bioactive compounds demonstrate protecting effects on DNA metabolism and repairing. There are many bioactive food compounds with proved cancer preventive effects that have role in epigenetic regulation, even protecting against oxidative stress damage. Epigenetics term represents the permanent alterations in DNA structure which has ability to change gene expression without differentiating the sequence of DNA. Histone modifications and DNA methylation to be the most frequent, also non coding RNA (ncRNA) regulations are epigenetic changes that have effects on expression of genes. Epigenetic changes are affecting formation and pathogenesis of chronic disorders such as cancer. In studies, it was demonstrated that genes that have roles in regulation of cell cycle, DNA repair, angiogenesis, apoptosis were altered epigenetically via hypermethylating their CpG islands which led to their supression and inactivation. These regulatory genes are tumor supressor genes such as, cyclin dependent kinases, phosphatases, insulin-like growth factor-II, E-cadherin, glutathione-S-transferase. Patterns of DNA methylation are reported to be affected through comsuming of multiple and/or combined food ingredients such as as zinc, vitamin A and some other dietary ingredients. Reversible histon modifications including acetylation, methylation, phosphorylation, dephosphorylation, ubiquitination, biotinylation are other epigenetic mechanisms. Of these alterations, histone deacetylase modification was reported to be important in cancer. Many nutrients consisting butyrate, diallyl disulfide, sulforaphane were demonstrated to change enzyme activity of histone deacetylase. Local and global histone acetylation were increased in promoters of genes such as P21 and Bax that have roles in cell senescence and apoptosis. In cancers, variable miRNA expression levels were detected and evidences revealed that miRNA and other ncRNA regulations, may lead to a mechanism that protects from cancer via nutrients and dietary compounds. In recent years, many studies focused on investigating roles of dietary compounds in cancer epigenetics including ncRNAs. Natural dietary agents determined to affect epigenetic mechanisms and ncRNA expression profiles are reported to be curcumin, epigallocatechin-3-gallate (EGCG), resveratrol. In the studies, it was shown that many bio-active compounds of nutrients have effects on epigenetic processes like methylation of DNA, histone modifications, and ncRNA regulations. If it is though epigenetic alterations frequently occur in cancers, transition of these changes via nutrition may be a very useful prevention and auxiliary treatment tool for cancer.

Keywords: Nutrigenomics, Cancer, Epigenetics, DNA methylation, Histone modification, ncRNA

# **1. INTRODUCTION**

Genetic advances give new insights into various health problems, consisting diet's role in prevention of cancer. After the completion of Human Genome Project in 2003, significant progress has occured in understanding the impact of the human genome on health and different disorders. Genomic investigations have heralded the development of novel therapies targeting the molecular mechanisms of diseases. In this context, nutrigenomics that is a combination of molecular nutrition and genomics has emerged as a science that demonstrates how nutrients affect gene expression and in conclusion affect metabolism of the cell (Nicastro et al., 2012).

Nutrition provides different substances that can act as nutrients, energizers (carbohydrates and fats), cell structure sources (proteins), metabolism controllers (vitamins and minerals), and thus the protection of homeostasis. The nutritional status of an individual occurs as a conclusion of the interaction of many factors like genetic structure, physical structure of the body, emotional and social situation (Sales et al., 2014).

Nutrients and other bioactive compounds can be useful as well as initiating various disorders. Disorders affected by consumption of food include Celiac, Phenylketonuria, and chronic diseases such as Cancer, Diabetes and Dyslipidemies. Accordingly, together with other omics, nutrigenomic aims to explain the interaction of bioactive compounds of food sources and genes (Liu and Qian, 2011).

Nutrigenomics is a multidisciplinary field that investigates the effect of nutrients and bioactive food components on expression of genes (Lundstrom, 2013).

Epigenetics term has emerged in order to explain to environmental and gene interactions during development that are effective in the emergence of phenotypes. This term was then used to identify cellular processes, that were generated in response to environmental effects, which could be inherited without any change in the DNA sequence (a change in the phenotype without alteration in the genotype), and had a long-term effect on gene expression (Joseph et al., 2016).

The most common epigenetic mechanisms include DNA methylation and chromatin remodeling. Epigenetic modifications may play an important role in the formation and pathogenesis of inherited diseases, such as cancer.

Epigenetics includes modifications of protein material and DNA, due to the connection of DNA and histones, changes happen in the chromatin structure; but the sequence of nucleotides does not change. Epigenetic changes are slow and persistent, but they can also be reversed (Nasir et al., 2019).

Epigenetic changes involve three different regulation types.1) Histone modifications 2) DNA methylation 3) Regulation of gene expression by nc-RNAs (Russo et al., 2017)

# **DNA Methylation**

DNA methylation is characterized by the binding of CpG dinucleotides to the 5th carbon of the cytosine base. DNA methyl-transferases (DNMTs) that are found in 3 major forms, DNMT1, DNMT3a, DNMT3b; catalyze the modification. DNA methylation leads to activation or suppression of transcription, according to the location of the methylation site (gene promoters or other regions) (Dates and Tollefsbol, 2018). Abnormal DNA methylation patterns in cancer, like hypermethylation or hypomethylation, have been correlated with the progression and metastasis of cancer in different types of tumors (Ehrlich, 2019; Majumdar et al., 2011).

Patterns of DNA methylation can be changed globally or in particular regions all over the whole genome. The DNA methylation mechanism depends on the presence of cofactors that regulate the enzymatic activity of the DNA-methyltransferases and methyl donors such as the ones coming from one-carbon cycle (Andreescu et al., 2018).

S-Adenosyl Methionine (SAM) which is the only known physiological methyl group donor, is synthesized via methionine, using different dietary procursors (folate, choline, betaine) and using homocysteine as methylation substrate (Feil and Fraga, 2012; Zeisel, 2009). Reducing the presence of methyl donors decreases SAM synthesis that leads to DNA methylation changes (hypermethylation or hypomethylation), whereas the increase in the presence of methyl donors generally correlates with DNA hypermethylation (Cheng and Blumenthal, 2008). DNA hypermethylation leads to the transcriptional silencing of tumor suppressor genes so that leading to malignant transformation and forming many types of cancer, while DNA hypomethylation leads to the activation of proto-oncogenes (Wajed et al., 2001).

Methionine, folate, betaine and choline are sources of methyl groups in food (Obeid, 2013). The metabolism of choline, methionine and folate interacts at the place where homocysteine is changed into methionine. Homocysteine S-methyltransferase catalyzes the methylation of homocysteine by using betaine as a methyl donor. In an alternative route, 5-methyltetrahydrofolate-homocysteine-S-methyltransferase (methionine synthase or MTR) forms methionine from de novo methyl group coming from the one-carbon pool. SAM is converted from methionine by Methionine adenosyltransferase (Niculescuand Zeisel, 2002).

The lack of nutrients involved in one-carbon metabolism, such as folate, methionine, cobalamin, pyridoxine, riboflavin, causes cancer-related changes in DNA methylation. Riboflavin deficiency leads to inhibition of methylenetetrahydrofolate reductase (MTHFR), whereas folate and pyridoxine deficiency cause inhibition of DNA methyltransferases (DNMTs), thus resulting in hypomethylation of DNA that leads to increased cancer risk (Niculescuand Zeisel, 2002).

### **Histone Modifications**

Modifications of histones have a crucial role in chromatin structure regulation and function, thus affecting processes such as transcription, repair of DNA, replication (Bannister and Kouzarides, 2011). Unlike DNA methylation, histone modifications have variations. Various modifications, such as methylation, phosphorylation, ubiquitination, acetylation, and sumolyation, form at the amino terminus of the histones. These modifications can activate or suppress transcription. There is a correlation between different cancers types and the alteration in certain posttranslational histone modifications (Dates and Tollefsbol, 2018).

### miRNAs

miRNAs which are about 22 nucleotides in length, are a class of non-coding RNAs, involved in post-transcriptional gene silencing to influence the translation and stability of mRNAs (Bartel, 2004). miRNAs play roles in many biological processes including cell growth, differentiation, apoptosis and cell cycle regulation (Jovanovic and Hengartner, 2006). In cancer, a variety of study data is available to demonstrate that miRNA expressions are variable. By the effects of nutrients and bioactive compounds, protective anti cancer mechanisms can be triggered via miRNA and other non-coding RNA regulations (Ross and Davis, 2011).

### **Bioactive Compounds and DNA Methylation**

Resveratrol and genistein are phytoestrogens that interact with estrogen receptors and estrogen-responsive genes. Genistein has been reported to inhibit epigenetic mechanisms responsible for proliferation in esophageal squamous carcinoma and prostate cancer cells (Majid et al., 2008). Phytoestrogens have been reported to stimulate p21 promoter, suppress the transcriptional activation of AP-1, as well as inducing PTEN expression (Chung et al., 2006). Bioflovonoid quercetin, tea catechins [epigallocatechin-3-gallate (EGCG)], coffee polyphenols (caffeic acid, chlorogenic acid) have effects on inhibition of DNA methylation and inhibition of DNMT activity (Liao et al., 2014). The polyphenol curcumin is abundant in turmeric and in a small amount in ginger. It has been shown that by inhibiting the activity of DNMT, curcumin demonstrated anticancer effect in leukemia cells (Liu et al., 2009; Andreescu et al., 2018). Lycopene is present in tomatoes and red fruits such as red carrots, watermelons and papayas. Lycopene was reported to have duty in hypomethylation of Glutathione-S-transferase P1 in breast neoplasia and has been shown to induce the expression of RARβ2 and hairpin induced 1 genes in non-cancerous cells. However, the mechanism of action of lycopene in DNA methylation is not fully understood and further studies are needed (Bishop and Ferguson, 2015; Andreescu et al., 2018).

### **Bioactive Compounds and Histon Modifications**

Since the acetylation-deacetylation balance in histones is caused by the interaction between histone acetyltransferases (HAT) and histone deacetylases (HDAC), changes in the activity of these enzymes have an effect on cancer development by directly affecting the epigenetic status of histones (Herceg, 2009). HDAC activity is inhibited by organosulfur compounds. Diallyl disulfide in the garlic and sulfarophane in crucifer are such examples of compounds that demonstrate potential tumor suppression effect by inhibiting HDACs, Dialil disulfide has been shown to induce histone hyperacetylation and increase expression of p21 in colon cancer cells (Dashwood and Ho, 2007). Sodium buturate which is a derivative of short-chain fatty acids; and luteoline flavonoid which is present in parsley, thyme, peppermint, basil celery, artichoke in high amounts, prevent the development and invasion of cancer cells by inhibiting HDAC activity and therefore increase histone acetylation (Attoub et al., 2011). It is demonstrated that, in order to inhibit cell proliferation and induce apoptosis in cancer cells, genistein increases histone acetylation and HAT activity whereas curcumin has been reported to inhibit the activity of HDACs (Andreescu et al., 2018). The effect of curcumin increases with the presence of reactive oxygen species because it has been reported that the effect of curcumin decreases when antioxidant enzymes decrease (Kang et al., 2005). Resveratrol and quercetin have been shown to lead to the activation of sirtuin 1 (SIRT1) which is a protein deacetylase (Chung et al., 2010). EGCG has been shown to change histone acetylation and methylation status and to be a HDAC inhibitor in cancer cells. In skin cancers, EGCG has been reported to cause histone acetylation, methylation and ubiquitination, while it has been demonstrated to cause histone phosphorylation in lung cancers (Andreescu et al., 2018).

### **Bioactive Compounds and miRNAs**

As well as folic acid affects DNA methylation, it affects miRNA expression. It has been shown that some miRNA changes can be prevented when sufficient folat is taken (eg, miRNA-122 is increased in order to demonstrate tumor suppressor effect by folate intake in hepatocellular carcinoma). Folate has also been shown to inhibit alcohol-induced expression of miRNA-10a (Andreescu et al., 2018). Curcumin increases tumor suppressor miRNA-22 expression and reduces oncogenic miRNA-199a expression and thus protects from cancer (Parasramka et al., 2010; Saini et al., 2010;

Sun et al., 2008) (91-93) In breast and prostate cancer cells, curcumin has been shown to affect tumor suppressor miR-15a and miR-16 expressions (Saini et al., 2010; Yang et al., 2010). In melanoma cells, genistein has been reported to induce oncogenic miRNA-27a downregulation and thus suppress cell proliferation (Sun Q et al., 2009). Genistein was demonstrated to upregulate tumor suppressor miRNA-1296, as well; which reduces the expression of MCM2, a gene responsible for carcinogenesis in prostate cancer (Parasramka et al., 2010).

### **2. CONCLUSION**

Epigenetic processes are regulated by bioactive food components that have anticancer potentials. Molecules taken via diet are components of many chemical classes consisting folate, polyphenols, retinoids (Vitamin A), fatty acids, allyl compounds. They can affect mechanisms such as signal transduction pathways, cell growth, differentiation, and apoptosis which are effective in protecting against cancer; as well as affecting epigenetic processes such as global DNA hypomethylation, tumor suppressor gene promoter hypermethylation, onco-modifications that are affecting initiation of cancers (Ong et al., 2011).

### **3. REFERENCES**

- Andreescu N, Puiu M, Niculescu M., 2018. Effects of Dietary Nutrients on Epigenetic Changes in Cancer. Methods in Molecular Biology 1856: 121-139.
- Attoub S, Hassan AH, Vanhoecke B, Iratni R, Takahashi T, Gaben A-M, Bracke M, Awad S, John A, Kamalboor HA, Al Sultan MA, Arafat K, Gespach C, Petroianu G., 2011. Inhibition of cell survival, invasion, tumor growth and histone deacetylase activity by the dietary flavonoid luteolin in human epithelioid cancer cells. European Journal of Pharmacology 651 (1–3): 18-25.

Bannister AJ, Kouzarides T., 2011. Regulation of chromatin by histone modifications. Cell Research 21(3): 381-95.

- Bartel DP., 2004. MicroRNAs: genomics, biogenesis, mechanism, and function. Cell 116(2): 281-97.
- Bishop KS, Ferguson LR., 2015. The interaction between epigenetics, nutrition and the development of cancer. Nutrients 7 (2): 922-947.
- Cheng X, Blumenthal RM., 2008. Mammalian DNA methyltransferases: a structural perspective. Structure 16(3): 341-350.
- Chung JH, Ostrowski MC, Romigh T, Minaguchi T, Waite KA, Eng C., 2006. The ERK1/2 pathway modulates nuclear PTENmediated cell cycle arrest by cyclin D1 transcriptional regulation. Human Molecular Genetics 15 (17): 2553-2559.
- Chung S, Yao H, Caito S, Hwang J-W, Arunachalam G, Rahman I., 2010. Regulation of SIRT1 in cellular functions: role of polyphenols. Archives of Biochemistry and Biophysics 501 (1): 79-90.
- Dashwood RH, Ho E., 2007. Dietary histone deacetylase inhibitors: from cells to mice to manSeminars in Cancer Biology 17(5): 363-369.
- Dates CR, Tollefsbol TO., 2018. Transforming Cancer Epigenetics Using Nutritive Approaches and Noncoding RNAs. Current Cancer Drug Targets 18(1): 32-38.
- Ehrlich M., 2009. DNA hypomethylation in cancer cells. Epigenomics 1(2):239-59.
- Feil R, Fraga MF., 2012. Epigenetics and the environment: emerging patterns and implications. Nature Reviews Genetics 13(2): 97-109.
- Herceg Z., 2009. Epigenetics and cancer: towards an evaluation of the impact of environmental and dietary factors. Mutagenesis 22(2): 91-103.
- Joseph PV, Abey SK, Henderson WA., 2016. Emerging Role of Nutri-Epigenetics in Inflammation and Cancer. Oncology Nursing Forum 43(6): 784-788.
- Jovanovic M, Hengartner MO., 2006. miRNAs and apoptosis: RNAs to die for. Oncogene 25(46): 6176-87.
- Kang J, Chen J, Shi Y, Jia J, Zhang Y., 2005. Curcumin-induced histone hypoacetylation: the role of reactive oxygen species. Biochemical Pharmacology 69(8): 1205-1213.
- Liao Y-P, Chen L-Y, Huang R-L, Su P-H, Chan MWY, Chang C-C, Yu MH, Wang PH, Yen MS, Nephew KP, Lai HC., 2014. Hypomethylation signature of tumorinitiating cells predicts poor prognosis of ovarian cancer patients. Human Molecular Genetics 23 (7): 1894-1906.
- Liu B. and Qian S. B., 2011.Translational regulation in nutrigenomics. American Society for Nutrition. Advances in Nutrition 2: 511–519.
- Liu Z, Xie Z, Jones W, Pavlovicz RE, Liu S, Yu J, Li PK, Lin J, Fuchs JR, Marcucci G, Li C, Chan KK, 2009. Curcumin is a potentDNA hypomethylation agent. Bioorganic & Medicinal Chemistry Letters 19(3): 706-709.
- Lundstrom K., 2013. Past, present and future of nutrigenomics and its influence on drug development. Current Drug Discovery Technologies 10(1): 35-46.

- Majid S, Kikuno N, Nelles J, Noonan E, Tanaka Y, Kawamoto K, Hirata H, Li LC, Zhao H, Okino ST, Place RF, Pookot D, Dahiya R., 2008. Genistein induces the p21WAF1/CIP1 and p16INK4a tumor suppressor genes in prostate cancer cells by epigenetic mechanisms involving active chromatin modification. Cancer Research 68(8): 2736-2734.
- Majumdar S, Buckles E, Estrada J, Koochekpour S., 2011. Aberrant DNA methylation and prostate cancer. Current Genomics 12(7): 486-505.
- Nasir A, Bullo MMH, Ahmed Z, Imtiaz A, Yaqoob E, Jadoon M, Ahmed H, Afreen A, Yaqoob S., 2019. Nutrigenomics: Epigenetics and cancer prevention: A comprehensive review. Critical Reviews in Food Science and Nutrition 7: 1-13.
- Nicastro HL, Trujillo EB, Milner JA., 2012. Nutrigenomics and Cancer Prevention. Current Nutrition Reports 1(1): 37-43.
- Niculescu MD, Zeisel SH., 2002. Diet, methyl donors and DNA methylation: interactions between dietary folate, methionine and choline. The Journal of Nutrition 132(8): 2333S-2335S.
- Obeid R., 2013. The metabolic burden of methyl donor deficiency with focus on the betaine homocysteine methyltransferase pathway. Nutrients 5(9): 3481–3495.
- Ong TP, Moreno FS, Ross SA., 2011. Targeting the epigenome with bioactive food components for cancer prevention. Journal of Nutrigenetics and Nutrigenomics 4(5): 275-92.
- Parasramka MA, Ho E, Williams DE, Dashwood RH., 2012. MicroRNAs, diet, and cancer: new mechanistic insights on the epigenetic actions of phytochemicals. Molecular Carcinogenesis 51(3): 213-230.
- Ross SA, Davis CD., 2011. MicroRNA, nutrition, and cancer prevention. Advances in Nutrition 2(6): 472-85.
- Russo, G. L., V. Vastolo, M. Ciccarelli, L. Albano, P. E. Macchia, and P. Ungaro., 2017. Dietary polyphenols and chromatin remodeling. Critical Reviews in Food Science and Nutrition 57 (12): 2589–99.
- Saini S, Majid S, Dahiya R., 2010. Diet, microRNAs and prostate cancer. Pharmaceutical Research 27(6): 1014-1026.
- Sales NM, Pelegrini PB, Goersch MC., 2014. Nutrigenomics: definitions and advances of this new science. Journal of Nutrition And Metabolism 2014: 202759.
- Sun M, Estrov Z, Ji Y, Coombes KR, Harris DH, Kurzrock R., 2008. Curcumin (diferuloylmethane) alters the expression profiles of microRNAs in human pancreatic cancer cells. Molecular Cancer Therapeutics 7: 464-473.
- Sun Q, Cong R, Yan H, Gu H, Zeng Y, Liu N., 2009. Genistein inhibits growth of human uveal melanoma cells and affects microRNA-27a and target gene expression. Oncology Reports 22: 563-567.
- Wajed SA, Laird PW, DeMeester TR., 2001. DNA methylation: an alternative pathway to cancer. Annals of Surgery 234(1): 10-20.
- Yang J, Cao Y, Sun J, Zhang Y., 2010. Curcumin reduces the expression of Bcl-2 by upregulating miR-15a and miR-16 in MCF-7 cells. Medical Oncology 27(4): 1114-1118.
- Zeisel SH., 2009. Epigenetic mechanisms for nutrition determinants of later health outcomes. The American Journal of Clinical Nutrition 89(5): 1488S–1493S.



# Effect of Reduction Time on the Electrical Properties of PAN/AgNO3 Nanofibers

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**Abstract:** In this study, conductive polyacrylonitrile (PAN) nanofibers with the addition of silver nanoparticles were produced by electrospinning. PAN was chosen as a template polymer due to its easy spinnability and availability, while silver was preferred due to its superior electrical, optical, mechanical and catalytic properties. Production of nanofibers with silver nanoparticle addition is a challenge, since the silver nanoparticles tend to agglomerate in the spinning solution. Therefore, instead of silver nanoparticles, silver nitrate (AgNO3) was added into the spinning solution. PAN/AgNO3 nanofibers were successfully produced by electrospinning. In order to obtain Ag nanoparticles within the fiber structure, PAN/AgNO3 nanofiber mats were reduced by sodium borohydrate (NaBH4) with different times (5, 10, 30 and 60 minutes). The effect of reduction time on the electrical conductivities of the PAN/AgNO3 nanofibers were investigated. With the increasing reduction time, the electrical resistance values were decreased indicating that the surfaces became more conductive. The lowest electrical resistance was (55.176 m $\Omega$ ) obtained with the samples reduced for 60 minutes, while the highest electrical resistance value was (99.947 m $\Omega$ ) obtained with the samples reduced for 5 minutes. It was concluded that, reduction time has a significant effect on the electrical properties of PAN/AgNO3 nanofibers, and the electrical conductivity of the PAN/AgNO3 nanofibers increases with the increase in reduction time.

Keywords: Conductive nanofibers, silver nitrate, polyacrylonitrile, reduction, sodium borohydrate

### **1. INTRODUCTION**

Fibers with nano-scaled diameters can be produced by electrospinning. Electrospinning is a simple, low-cost, and effective process to produce nanofibers. The method involves using electrostatic forces to fabricate nanofiber mats. In this method, a polymer solution is subjected to electric field. Under the applied electrostatic force, a charged polymer jet is ejected from the nozzle and travels towards to a grounded collector. Under the influence of the electrical field, the solvent evaporates, the diameter of the jet reduces and fibers with nano-scaled diameters are collected on the collector. Many polymers can be electrospun with this method (Thandavamoorthy et al., 2005; Grapenson et al., 2016).

Due to their small diameters, electrospun nanofibers have unique properties, such as a high surface area-to-volume ratio, small pore sizes, high porosity, high mechanical strength, flexibility, etc. Therefore, these structures are especially preferred for application areas such as filters, tissue scaffolds, protective clothing, and sensors (Özdoğan et al., 2006; Rajkishore et al., 2011).

For sensor applications, nanofibers should be electrically conductive. There are many methods reported in the literature to produce conductive nanofibers. Among all of these methods, adding a conductive compound into the electrospinning solution is the most common one (Duzyer 2019; Demirsoy N et al., 2014).

The aim of this study is to produce PAN/AgNO3 nanofibers by electrospinning, to achieve conductive nanofibers by reducing AgNO3 with sodium borohydrate (NaBH4) and to investigate the effect of reduction time on the electrical properties of PAN/AgNO3 nanofibers. For this purpose, PAN/AgNO3 nanofibers were produced by electrospinning. Afterwards, the nanofibers were reduced with NaBH4 with different times (5, 10, 30 and 60 minutes). Finally, the surface and the electrical properties of the nanofibers were evaluated.

# 2. MATERIALS AND METHODS

In this study commercially available PAN (Mn: 200-240 kDa) polymer was used to produce the nanofibers. AgNO3, the conductive component was purchased from Tekkim Laboratory Chemicals. DMF was used as the solvent to prepare polymer solutions while NaBH4 was used as the reducing agent. These chemicals were purchased from Sigma-Aldrich.

In this study, PAN was chosen as the carrier polymer. In the first step, 7% wt. of PAN was dissolved in DMF and stirred at 75 °C on a magnetic stirrer until it was completely dissolved. Then, AgNO3 was added into the solution with a weight of 3% of the polymer and sonicated for 2 minutes by using a homogenizer (OV5 Homogenizer, Velp Scientifica).

PAN/AgNO3 nanofibers were produced on a rotating disc (100 rpm) by an electrospinning device (Nanospinner24, Inovenso) with a flow rate of 1 mL/h. The voltage was 18 kV and the distance between the needle and the collector was 13.5 cm.

After electrospinning, the nanofiber mats were cut with a size of 2x2 cm and were reduced with NaBH4 (1 % wt. in water) for 5, 10, 30 and 60 minutes. After reduction, the samples were rinsed with distilled water and then were dried at 40°C for 2 hours in an incubator (Nüve-EN 025). Afterwards, the samples were kept in room conditions for 2 days.

Visual changes after the reduction process were examined by camera images. The morphologies of the samples were investigated with an Olympus BX51 light microscope. Resistance of the samples were measured by a Four-Point Probe System (FPP 470-Entek Elektronik).

# **3.RESULTS AND DISCUSSION**

In this study, AgNO3 was chemically reduced using NaBH4, and Ag nanoparticles were formed within the fiber mat.

Figure 1 shows the camera images of the  $PAN/AgNO_3$  nanofiber mats before and after reduction. The yellowish color can be considered as an indicator for the reduction of  $AgNO_3$ . The samples treated with  $NaBH_4$  for 60 minutes showed the most significant color change.



Figure 1. Camera images of the nanofiber mats before and after the reduction process

In order to see the effect of  $NaBH_4$  on the surface properties of the nanofibers, optical microscope images were captured (Figure 2). The images showed that randomly aligned, bead-free, and uniform nanofibers were produced and the  $NaBH_4$  reduction did not damage the nanofibers.



Figure 2. Light microscope images of the PAN/AgNO3 nanofibers before and after the reduction process

Table 1 shows the resistance values of the nanofiber mats depending on the reduction time. The results proved that conductive nanofibers were successfully produced. According to these values, the resistance was decreased as the reduction time was increased (Figure 3).

Table 1. Properties of the Samples with Increasing Reduction Time



Figure 3. Electrical Properties of the Samples with Increasing Reduction Time

### 4. CONCLUSION

In this study the PAN/AgNO3 nanofibers were produced by electrospinning. In order to make them conductive, the produced nanofibers were reduced with NaBH4. The effect of reduction time on the electrical properties of PAN/AgNO3 nanofibers were investigated. It was seen that, NaBH4 did not damage the nanofiber structure and the resistance values of the samples were decreased with the increasing reduction time. The lowest electrical resistance was achieved with the samples reduced for 60 minutes. It can be concluded that electrospinning is an effective method to produce PAN/AgNO3 nanofibers, and NaBH4 reduction is an effective reduction method. Moreover, the electrical properties of the PAN/AgNO3 nanofibers can be improved by changing the reduction time.

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### REFERENCES

Demirsoy N, Uçar N, Önen A, Karacan I, Kızıldağ I, Eren O & Borazan I (2014). The effect Dispersion Technique, Silver Particle loading and Reduction Method on the Properties of Polyacrylonitrile-Silver Composite Nanofiber. Journal of Industrial Textiles, 45(6), 1173-1187

Duzyer S (2019). Different Methods of Fabricating Conductive Nanofibers. Journal of Textile & Apparel, 9(1), 78-85.

- Grapenson S & Jakob A (2016). Conductive Polypyrole Nanofibers via Electrospinning Electrical and Morphological Properties. Polymer, 47(5), 1597-1603
- Özdoğan E, Demir A & Seventekin N (2006). Nanotechnology and its Applications in Textile Industry. Textile and Apparel. 16(4), 225-229
- Rajkishore N, Padhye R, Kyratzis I L, Truong Y B & Arnold L (2011) Recent Advances in Nanofibre Fabrication Techniques, Textile Research Journal, 82(2), 129-147.
- Thandavamoorthy S, Bhat G S, Tock R W, Parameswaran S & Ramkumar S S (2005). Electrospinning of Nanofibers. Applied Polymer Science, 96(2), 557-569



# Thermal Diffusivity Measurement with the Combined Use of Quantitative IRT and Hot Box Method - A Promising Method for In-Situ Assessment

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Abstract: Thermal diffusivity is one of the major thermal characteristics that can be used to interpret the thermal performance of an insulated or not-insulated building walls and to identify thermal defects in those walls. Thermal diffusivity ( $\alpha$ , m<sup>2</sup>/s) is determined with an equation ( $\alpha = \lambda/\rho.c$ ) in which the thermal parameters, specifically thermal conductivity ( $\lambda$ , W/mK), bulk density ( $\rho$ , kg/m<sup>3</sup>) and specific heat capacity (c, KgJ/mK) are used. These properties are determined with laboratory analyses and require sample preparation at certain dimensions. Among these properties, thermal conductivity is determined by means of direct measurements using expensive equipment. Therefore, there is necessity to develop new measurement setups allowing practical assessment of thermal diffusivity, if possible using non-destructive methods for in-situ assessment. The main interest of the study is development of an experimental setup by using InfraRed Thermography (IRT) for measuring the real thermal diffusivity performance of building walls on site. For that purpose, two types of wall samples (W1 and W2) made of fired brick and autoclaved aerated concrete masonry units were prepared to examine their thermal diffusivity performance by quantitative IRT and calibrated hot box method at laboratory. Thermal diffusivity of those sample walls were determined by means of three different methods as explained below in detail

- The theoretical thermal diffusivity values were calculated with the use of thermal conductivity values of masonry units given in the standard TSE 825:2008.
- The reference data on thermal diffusivity of the wall samples were achieved by using thermal conductivity value which was measured by means of calibrated hot box method as defined in the standard TS EN ISO 8990:2002, and bulk density and specific heat values measured with laboratory tests. The calculated data and the measured reference data were found to be in good agreement with each other.
- The new measurement method was developed by the combined use of IRT and hot box method in which the testing methods defined in the standards BS EN ISO 8990:1996, ASTM STP 1320:1997 were adapted to shape the experimental setup. The warm side of the hot box was used as the heat source and the IR camera was positioned at the cold side of the sample wall. The changes in surface temperature of the sample wall were monitored with sequential IR imaging. The surface temperature data recorded during the warming up period of the hot box till it reached to steady state condition were used for the calculation of thermal diffusivity of the sample wall. The thermal diffusivity values measured with the new experimental setup using IRT were found to be almost the same with the reference thermal diffusivity values measured with the calibrated hot box method.

The joint interpretation of the results show that the experimental setup using IRT is successful in direct measurement of thermal diffusivity. Considering that the reliable data collected with this experimental setup, it is a promising method for the in-situ assessment of thermal diffusivity for building walls.

Keywords: Thermal diffusivity, infrared thermography, calibrated hot box, brick wall, autoclaved aerated wall



# Effect of Waste Tire Rubber and Fly Ash on Lightweight Geopolymer Concrete Production

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Abstract: Geopolymer materials will be the alternative material to be used in the construction sector in the future after concrete. In order to reduce  $CO_2$  emissions from cement production, it is necessary to investigate different methods or use alternative materials to cement by using a binder with less  $CO_2$  emission. The geopolymer material has the potential to be used as an alternative material to cement due to its low  $CO_2$  emissions, salts and acids, high resistance to heat and fire. In this study, in addition to aggregate, a certain amount of rubber production wastes and fly ash additive geopolymer lightweight concretes were produced. Samples prepared with sodium hydroxide (SH) and sodium silicate (SS) solutions (as 16 moles) as a geopolymer binder were cured in the curing room at 80% humidity until the experiment day after curing for 7 days in 70 ° C oven in the curing room. The effects of fly ash (FA) and waste tire crumb rubber (TRC) additives on some mechanical properties of lightweight geopolymer concrete were investigated. As a result, it was determined that although there is a decrease in the strength according to the increasing tire ratio, it can be used as a light construction material.

Keywords: Fly ash, waste tire ruber, slug, geoplymer mortar, compressive strength

### **1. INTRODUCTION**

Researchers, scientists and engineers to search for alternative construction materials that is be essentially sustainable, durable, user and eco-friendly. A solution to all these is with innovative geopolymeric construction materials. Geopolymers are an inorganic materials produced at low temperature in alkali medium through the process of geopolymerization – a synthesis which is as analogous as geo-synthesis of natural rocks, whereby Aluminium and Silicates rich precursors react in an exothermic way with alkali activators to give rise to geopolymers (Walah and Rangan 2006; Luhar et al. 2016; Luhar et al. 2018). Geopolymer is known material for its durability and environmental sustainability. Sustainable practice of industrial ecology demands the use of by-product of one industry as source materials for other industrial application. This will help in complying with environmental parameters for both the industry. Environmental concerns compelled us to reduce the material consumption by enhancing the durability and resource efficiency using advanced technology (Jaydeep and Chakravarthy, 2013; Bhowmick and Ghost, 2015). Geopolymer concrete is being studied extensively and shows promise as a substitute to Portland cement concrete (Parveen and Sharmaa, 2013).

The products obtained from scrap tyres rubber, which have an important place as waste, are classified (figure 1) as whole scrap tyres, slit tyres, shredded/chipped, ground and rubber crumb. Rubber crumb particle sizes range between 4.75mm to less than 0.075 mm (Rischardson et al., 2011) and are irregularly shaped, torn particles due to the micro-mill process, they are subjected to during the manufacturing process (Topçu, 1995; Maraghechi et al 2012; Kotresh and Balechev, 2014). The recycling will reduce the waste and contribute to a greener environment. Over the past few years, a number of researchers have studied on incorporated the waste tires into cement based materials (Tountanji, 1996; Bennazzouk et al. 2007; Sgobba et al 2010; Azmi et al 2016; Su et al. 2015; Pacheco-Torgal et al. 2015). In this study aim, some properties of geopolymer concrete which can be produced by using waste tire rubber were determined and effect of waste tire rubber usage was investigated.



Figure 1. Tire rubber

# 2. MATERIALS AND METHODS

Calcite aggregate was used in the preparation of the mixtures. The properties determined for ground blast furnace slag (GBFS) and flay ash (FA) and tire curmb rubber (TCR), which are used as mineral additives in mortar mixtures, are given in Table 1.

Table 1. Chemical and physical properties of FA and GBFS

| Chemical Composition           | FA    | GBFS  |
|--------------------------------|-------|-------|
| CaO                            | 1,73  | 56,10 |
| SiO <sub>2</sub>               | 55,73 | 21,00 |
| Al <sub>2</sub> 0 <sub>3</sub> | 29,76 | 17,00 |
| Fe <sub>2</sub> O <sub>3</sub> | 5,41  | 0,62  |
| Na <sub>2</sub> O              | 1,96  | 0,056 |
| K <sub>2</sub> 0               | 3,11  | 0,28  |
| MgO                            | 3,3   | 5,75  |
| SO <sub>3</sub>                | 0,3   | 0,19  |

Sodium silicate (SS) and sodium hydroxide (SH) were commercially available for the activation of GBFS and GP with the materials used in the mixtures. SH is in granulated form with a purity of 97%. SH was prepared in a laboratory environment of 16M (moles). SS and SH 's mixing ratio of 50/50 obtained by the activation solution obtained by the addition of GP and FA instead of GBFS mixture rates and codes are given in Table 2. Kastamonu mains water was used as mixture and curing water.

Table 2. Mix design for a mold (g/dm<sup>3</sup>)

| Mix | Slag         | Fly     | Fine          | Coarse        | Glass      | Silikat      | Hidroksite   | Water        |
|-----|--------------|---------|---------------|---------------|------------|--------------|--------------|--------------|
| No  | ( <b>g</b> ) | Ash (g) | Aggregate (g) | Aggregate (g) | Powder (g) | ( <b>g</b> ) | ( <b>g</b> ) | ( <b>g</b> ) |
| A   | 700          | 0       |               |               | 0          |              |              |              |
| B   | 700          | 0       |               |               | 75         |              |              |              |
| C   | 700          | 0       | 400           | 150           | 150        | 125          | 125          | 150          |
| D   | 350          | 350     | 400           | 130           | 0          | 123          | 123          | 150          |
| E   | 350          | 350     |               |               | 75         |              |              |              |
| F   | 350          | 350     |               |               | 150        |              |              |              |

In the experimental study, three batches were produced, one for control and three for each batch. Sand and substitute materials were mixed dry in the concrete, then the mixing alkaline activator was added. The process followed in the preparation of the mixtures is shown in the Figure 2. The last step, remaining mixture water added and was mixed to the concrete for 3 min. Then the mixing was terminated.  $10 \times 10 \times 10 \times 10$  cm cube specimens were placed in the sample containers for 3<sup>th</sup>, 7<sup>th</sup> and 28<sup>th</sup> days compressive and flexural strength. The specimens were covered with an impermeable cover for 24 hours at  $23 \pm 2$  ° C and the relative humidity of 55-60%, then the samples taken from the sample cup were kept in lime

saturated water at  $20 \pm 2$  ° C until the test day. Compressive strenght according to TS 12390-3 and flexural strenght according to TS 12390-6 were applied to the samples.



Figure 2. Geoplimer mix procedure

### **3. RESULTS**

Compressive strengt and tensile splitting strenght are the average of at least three standard cured strength specimens made from the same concrete sample and tested at the same age. The vast majorities of cases strength requirements for concrete are at an age of 28<sup>th</sup> days of curing. After 28<sup>th</sup> days, the concrete cubes were examined for the purpose of their compressive strengths and tensile splittingh strenght. The results of the mixtures are given in Figure 3 and Figure 4.



Figure 3 Compressive strenghts for geoplimer mortars



Figure 4 Tensile splitting strenghts for geoplimer mortars

When the Figure 3 is examined, it is seen that the compressive strength of geopolymer concrete prepared 16 molarity is above 40 MPa. However, with the addition of 75 g TCR, a 30% strength loss was experienced in compressive strength and a 42% strength loss with 150 g TCR addition. In FA added groups, this ratio was 75% for 75 g and 81% for 150 g, respectively. These strength losses were similar in the results of the tensile splitting strength with FA addition and 40% loss was experienced with the increase in the amount of TCR. This can be explained by the insufficiency of the decreasing amount of GBFS. However, when the groups without FA (Figure 4) were examined, it was found that the TCR additive increased the strength up to 45% depending on the increasing rate.

### 4. CONCLUSION

The main purpose of this research is to investigate the effect of using TCR on geopolymer concrete. For this purpose, compressive strength and tensile splitting strength of geoplimer samples were examined. It has been determined that the addition of TCR additive in geoplimer concretes causes significant decreases in strength and FA additive has a similar effect due to decreasing GBFS. In addition, it has been observed that there is an increase in tensile splitting strength with the positive effect of increasing TCR additive. However, it has been observed that FA contribution due to decreasing GBFS is negatively affected by decreasing strength values. As a result, it was determined that although there is a decrease in the strength according to the increasing TCR ratio, it can be used as a lightweight construction material.

### REFERENCES

- Azmi, A. A., Abdullah, M. M. A. B., Ghazali, C. M. R., Sandu, A. V., & Hussin, K. (2016). Effect Of Crumb Rubber On Compressive Strength Of Fly Ash Based Geopolymer Concrete. In MATEC Web of Conferences (Vol. 78, p. 01063). EDP Sciences.
- Benazzouk, A., Douzane, O., Langlet, T., Mezreb, K., Roucoult, J. M., & Quéneudec, M. (2007). Physico-mechanical properties and water absorption of cement composite containing shredded rubber wastes. Cement and Concrete Composites, 29(10), 732-740.)
- Bhowmick, A., & Ghosh, S. (2012). Effect of synthesizing parameters on workability and compressive strength of fly ash based geopolymer mortar. Int. J. Civ. Struct. Eng, 3(1), 168 n177. J.T. Gourley, Journal of the Australian Ceramics Society, 50(1), 102 (2014)
- Jaydeep, S., & Chakravarthy, B. J. (2013). Study On Fly Ash Based Geo-Polymer Concrete Using Admixtures. International Journal of Engineering Trends and Technology, 4(10).
- Kotresh, K. M., & Belachew, M. G. (2014). Study on waste tyre rubber as concrete aggregates. International Journal of Scientific Engineering and Technology, 3(4), 433-436.
- Luhar, S., & Khandelwal, U. (2015). A study on water absorption and sorptivity of geopolymer concrete. SSRG International Journal of Civil Engineering, 1-10.
- Luhar, S., Chaudhary, S., & Dave, U. (2016). Effect of different parameters on the compressive strength of rubberized geopolymer concrete. Multi-disciplinary Sustainable Engineering: Current and Future Trends.
- Luhar, S., Chaudhary, S., & Luhar, I. (2018). Thermal resistance of fly ash based rubberized geopolymer concrete. Journal of Building Engineering, 19, 420-428.

- Maraghechi, H., Fotovat Ahmadi, I., & Motahari, S. (2012). Effect of adding crumb tire rubber particles on the mechanical properties of DCPD-modified sulfur polymer mortars. Journal of Mechanics of Materials and Structures, 6(9), 1283-1294.
- Pacheco-Torgal, F., Ding, Y., & Jalali, S. (2012). Properties and durability of concrete containing polymeric wastes (tyre rubber and polyethylene terephthalate bottles): An overview. Construction and Building Materials, 30, 714-724.
- Parveen, S. D., & Sharma, A. (2013). Rubberized concrete: Needs of good environment (overview). Int. J. Emerg. Technol. Adv. Eng, 3, 192-196.
- Richardson, A., Coventry, K., Dave, U., & Pienaar, J. (2011). Freeze/thaw performance of concrete using granulated rubber crumb. Journal of Green Building, 6(1), 83-92.
- Sgobba, S., Marano, G. C., Borsa, M., & Molfetta, M. (2010, June). Use of rubber particles from recycled tires as concrete aggregate for engineering applications. In 2nd International conference on sustainable construction materials and technologies.
- Su, H., Yang, J., Ling, T. C., Ghataora, G. S., & Dirar, S. (2015). Properties of concrete prepared with waste tyre rubber particles of uniform and varying sizes. Journal of Cleaner Production, 91, 288-296.
- Topcu, I. B. (1995). The properties of rubberized concretes. Cement and concrete research, 25(2), 304-310.
- Toutanji, H. A. (1996). The use of rubber tire particles in concrete to replace mineral aggregates. Cement and Concrete Composites, 18(2), 135-139.

Wallah, S., & Rangan, B. V. (2006). Low-calcium fly ash-based geopolymer concrete: long-term properties.



# Effect of Adding Glass Powder in Geopolymer Concrete

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**Abstract:** Cement is one of the most important building materials that human beings have been using for the last decades. In addition, high energy requirements of cement production and the harmful effects of CO<sub>2</sub> gas on the atmosphere are one of the most important subject studies in recent years. For this reason, a number of studies are carried out for the production of alternative binding material for the cement, too. Geopolymer materials, because of the excellent mechanical properties of the building material as a very large potential materials. Metakaolin makes it possible to make a building material with cement-like binding properties as a result of the activation of materials such as blast furnace slag and fly ash by mixing with various chemical activators. In this study, the effect of fly ash and glass powder in the production of geopolymer mortar obtained by the activation of fly ash with sodium hydroxide (NaOH) and sodium silicate (Na<sub>2</sub>SiO<sub>3</sub>) was investigated. Fly ash (FA) and glass powder (GP) were replaced with ground blast furnace slag (GBFS), and the geopathic mortar was poured into 10x10x10 cm molds. These specimens were determined by bending and compressive strengths. As a result, the effect of slag additive is more effective than fly ash and glass powder, and the use of glass powder and fly ash has been seen to have a negative effect on these mortars.

Keywords: Fly ash, marble powder, slug, geoplymer mortar, compressive strength

### **1. INTRODUCTION**

Solid wastes are substances and masses resulted by the various human activities that have to be dumped. Solid waste materials usually include industrial waste, medical waste, and domestic waste. In particular, construction waste is the output result of construction and destruction, rehabilitation, repair, removal of existing structures, and installations. This waste is composed of sand, stone, gravel, tiles, ceramic, marble, glass, aluminum, wood, plastic, paper, paints, plumbing pipes, electric parts and asbestos, and other materials.For centuries, glass has been serving as a universal packaging container, holding commodities and today, manufacturers use glass to hold everything from soda to perfume (EPA,2012). With time, the increasing use of glass products is resulting in large amounts of waste glass (IEA,2007). Reuse/recycle and waste reduction are very important elements in a framework of waste management. It helps to conserve natural resources, reduce demand for valuable landfill space, diminish the need of raw materials to make new product, reduce air and water pollution, reduce energy and create new jobs (Ling et al., 2013). However, not all used glass can be recycled into new glass because of impurities, cost, or mixed colors (Shi, 2009). It is necessary to establish new options to utilize recycled waste glass. One important option is to use waste glass as construction materials. Glass is basically a product of the super cooling of a melted liquid mixture consisting primarily of sand (silicon dioxide) and soda ash (sodium carbonate) to a rigid condition, in which the super cooled material, does not crystallize and retains the organization and internal structure of the melted liquid (Chesner et al. 2002). With more than 10 billion tons of concrete produced annually, it is considered to be the most important building material (Meyer, 2009). Also, the demand for concrete is expected to grow to approximately 18 billion tons a year by 2050 [Roskovic and Bjegovic, 2006; Mehta and Monteiro, 2006). Consequently, the concrete industry is going to use a considerable amount of natural resources to produce cement and concrete. For this reason, research works are being made on alternative building materials for last years and a significant space is covered by the potential use of waste or recycled materials in concrete (Shayan 2002; Ledererova and Grünner 2006; Jevtic et al. 2012).

Geopolymer binders are a class of inorganic polymers synthesized by the reaction of an aluminosilicate material with a concentrated alkali hydroxide and/or alkali-silicate solution, having an amorphous three-dimensional structure (Nazari and Sanjavan, 2015). The use of waste materials in the production of geopolymer binder is another potential solution to overcoming the aforementioned waste management problems, which recently has been successfully introduced (Tchadjie

et al. 2016; Ahmari et al. 2012; Vasquez et al. 2012;Paya et al. 2015). Ecological or environmental benefits of use of geopolymer technology for recycling of waste-glass include (1) the increased diversion of waste material from landfills for useful applications, (2) the reduction in the use of energy and  $CO_2$  emission attributed to Portland cement production, (3) the conservation of natural resources (Nazari and Sanjavan, 2015). The aim of this study is to investigate the effect of use of marble dust and fly ash, an industrial waste in geoplimer mortars.

# 2. MATERIALS AND METHODS

Calcite aggregate was used in the preparation of the mixtures. The properties determined for ground blast furnace slag (GBFS), flay ash (FA) and glass powder GP, which are used as mineral additives in mortar mixtures, are given in Table 1.

| <b>Chemical Composition</b>    | FA    | GBFS  | GP    |
|--------------------------------|-------|-------|-------|
| CaO                            | 1,73  | 56,10 | 11,41 |
| SiO <sub>2</sub>               | 55,73 | 21,00 | 71,0  |
| Al <sub>2</sub> 0 <sub>3</sub> | 29,76 | 17,00 | 1,57  |
| Fe <sub>2</sub> O <sub>3</sub> | 5,41  | 0,62  | 0,39  |
| Na <sub>2</sub> O              | 1,96  | 0,056 | 12,89 |
| K <sub>2</sub> 0               | 3,11  | 0,28  | 0,54  |
| MgO                            | 3,3   | 5,75  | 1,24  |
| SO <sub>3</sub>                | 0,3   | 0,19  | 0,07  |

Table 1. Chemical and physical properties of materials

Sodium silicate (SS) and sodium hydroxide (SH) were commercially available for the activation of GBFS and GP with the materials used in the mixtures. SH is in granulated form with a purity of 97%. SH was prepared in a laboratory environment of 16M (moles). SS and SH 's mixing ratio of 50/50 obtained by the activation solution obtained by the addition of GP and FA instead of GBFS mixture rates and codes are given in Table 2. Kastamonu mains water was used as mixture and curing water.

| Mix | Slag         | Fly Ash      | Fine          | Coarse        | Glass      | Silikat     | Hidroksite   | Water        |
|-----|--------------|--------------|---------------|---------------|------------|-------------|--------------|--------------|
| No  | ( <b>g</b> ) | ( <b>g</b> ) | Aggregate (g) | Aggregate (g) | Powder (g) | <b>(g</b> ) | ( <b>g</b> ) | ( <b>g</b> ) |
| A   | 600          | 0            |               |               | 200        |             |              |              |
| B   | 500          | 0            |               |               | 300        |             |              |              |
| C   | 400          | 0            | 200           | 700           | 400        | 150         | 150          | 200          |
| D   | 300          | 300          | 800           | /00           | 200        | 150         | 150          | 200          |
| E   | 250          | 250          |               |               | 300        |             |              |              |
| F   | 200          | 200          |               |               | 400        |             |              |              |

**Table 2.** Mix design for a mold  $(g/dm^3)$ 

In the experimental study, three batches were produced, one for control and three for each batch. Sand and substitute materials were mixed dry in the concrete, then the mixing alkaline activator was added. The process followed in the preparation of the mixtures is shown in the Figure 1. The last step, remaining mixture water added and was mixed to the concrete for 3 min. Then the mixing was terminated.  $10 \times 10 \times 10$  cm cube specimens were placed in the sample containers for 3<sup>th</sup>, 7<sup>th</sup> and 28<sup>th</sup> days compressive strength. The specimens were covered with an impermeable cover for 24 hours at  $23 \pm 2$  ° C and the relative humidity of 55-60%, then the samples taken from the sample cup were kept in lime saturated water at  $20 \pm 2$  ° C until the test day. Compressive strength according to TS 12390-3 was applied to the samples.



Figure 1. Geoplimer mix procedure

# **3. RESULTS**

Compressive strength is the average of at least three standard cured strength specimens made from the same concrete sample and tested at the same age. The vast majorities of cases strength requirements for concrete are at an age of 28<sup>th</sup> days of curing. After 28<sup>th</sup> days, the concrete cubes were examined for the purpose of their compressive strengths. The results of the mixtures are given in Figure 2.



Figure 2. Compressive strenghts for geoplimer mortars

When the figure 2 is examined, it is seen that despite the increasing GP and FA ratio, the compressive strength decreases due to the decrease of GBFS. With the increase of glass powder (GP) a strength loss of about 20% was observed. However, this negative effect of glass powder (GP) was found to have some increase effect with increasing fly ash (FA).



Figure 3. Absorbsion rate for geoplimer mortars

When the water absorption rates were examined (figure 3), it was seen that there was an increase in the gap structure compared to the increasing GP and FA ratio, and this increase resulted in less gap formation in the GP addition of FA.

### 4. CONCLUSION

The main aim of this research was to study the effect of adding glass powder (GP) to geopolymer concrete. The tests which were used in this study were compressive strength and total absorption. The basic materials for mixing Concrete was fly ash, fine aggregate, coarse aggregate, silikat and hidroksite. It was conclude that gass powder in geoplimer concrete affected glass powder in concrete affected positively on compressive Strength during curing days of glass powder. In addition to that, as the glass powder increases, the compressive strength decreases. Also we can note that when adding fly ash to sample, the compressive strength decreases. Moreover, we can say that the best results was obtained samples the high GBFC contain. It also conclude that the absorption percentage in mix increased glass dust and fly ash.

### REFERENCES

- Ahmari, S., Ren, X., Toufigh, V., & Zhang, L. (2012). Production of geopolymeric binder from blended waste concrete powder and fly ash. Construction and Building Materials, 35, 718-729.
- Chesner, W. H., Collins, R. J., MacKay, M. H., & Emery, J. (2002). User guidelines for waste and by-product materials in pavement construction (No. FHWA-RD-97-148, Guideline Manual, Rept No. 480017). Recycled Materials Resource Center..
- IEA, 2007. Tracking Industrial Energy Efficiency and CO2 Emissions. OECD/IEA, pp: 1-321.
- Jevtic, D., Zakic, D., & Savic, A. (2012). Achieving sustainability of concrete by recycling of solid waste materials. Mechanical Testing and Diagnosis, 2(1), 22.
- Ledererová, M., & Grünner, K. (2006). Optimization of the Technology for Recycling Concrete Materials. Slovak Journal of Civil Engineering.
- Ling, T. C., Poon, C. S., & Wong, H. W. (2013). Management and recycling of waste glass in concrete products: Current situations in Hong Kong. Resources, Conservation and Recycling, 70, 25-31.
- Mehta, P. K., & Monteiro, P. J. (2006).. Concrete: Microstructure, Properties and Materials. 3rd Edn., McGraw-Hill, New York.
- Meyer, C. (2009). The greening of the concrete industry. Cement and concrete composites, 31(8), 601-605.
- Nazari, A., & Sanjayan, J. G. (2015). Synthesis of geopolymer from industrial wastes. Journal of Cleaner Production, 99, 297-304.
- Payá, J., Monzó, J., Borrachero, M. V., & Tashima, M. M. (2015). Reuse of aluminosilicate industrial waste materials in the production of alkali-activated concrete binders. In Handbook of alkali-activated cements, mortars and concretes (pp. 487-518). Woodhead Publishing.
- Rosković, R., & Bjegović, D. (2005). Role of mineral additions in reducing CO2 emission. Cement and Concrete Research, 35(5), 974-978.
- Shayan, A. (2002, January). Value-added utilisation of waste glass in concrete. In IABSE Symposium Report (Vol. 86, No. 6, pp. 12-21). International Association for Bridge and Structural Engineering.

- Shi, C. (2009). Corrosion of glasses and expansion mechanism of concrete containing waste glasses as aggregates. Journal of materials in civil engineering, 21(10), 529-534.
- Tchadjié, L. N., Djobo, J. N. Y., Ranjbar, N., Tchakouté, H. K., Kenne, B. B. D., Elimbi, A., & Njopwouo, D. (2016). Potential of using granite waste as raw material for geopolymer synthesis. Ceramics International, 42(2), 3046-3055.
- US EPA, 2012. Glass, Common Wastes and Materials. Wastes Resources Conservation-common Wastes and Materials. Retrieved from: http://www.epa.
- Vásquez, A., Cárdenas, V., Robayo, R. A., & de Gutiérrez, R. M. (2016). Geopolymer based on concrete demolition waste. Advanced Powder Technology, 27(4), 1173-1179.



# Use of Marble Powder and Fly Ash in Self Compacting Concrete

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Abstract: Recently, Self-Compacting Concrete (SCC) has gained an extraordinary usage for placement in congested reinforcement concrete structures where casting conditions are un-easy and in high rise buildings where pump ability properties are mandatory. SCC utilized where, Fresh property of concrete required high fluidity and good cohesiveness. The project can lead to use of different percentages of marble powder between 0 %, 5%, 10% and 20% as an industrial waste and fly ash as a pozzolana material with similar percentages as marble powder. The established benefits substitute marble powder and fly ash by cement to make concrete more affordable, save the landfill and minimize CO<sub>2</sub> emission when use of less cement. The amount of marble powder as a waste material is significantly in the rise. Thereby, the utilization of marble powder in self compacting concrete is the main substance considered as a filler material. The main aim of these study is the behavior of SCC with marble powder and fly ash and gaining knowledge of the effect on fresh property as well as durability and also investigate the compatibility of marble powders in SCC along with chemical admixture such as super plasticizers and substitution of cement by marble powder and fly ash. The marble sector, which is in use in the construction sector, is a sector that increases its usage area in many countries and is developing day by day thanks to its rich roll bearings. Wastes in this sector are developing marble powder is becoming an environmental problem. The aim of this study is to reduce the effect of environmental pollution caused by marble dust and fly ash in SCC and to examine whether they are a material that can be used in SCC. For this purpose, different mixtures of marble powders and fly ash were formed by replacing the cement with different ratios. 100 mm cube samples were prepared with the prepared mixtures, and some physical properties of these samples were determined by pressure and tensile strength at 3, 7 and 28 days. Samples were compared with SCC concrete values with conventionally generated reference. As a result, it was observed that the contribution of fly ash in SCC was more effective than the contribution of waste marble dust and could be used as powder material.

Keywords: Fly ash, chemical admixture, marble powder, SCC, fresh concrete, compressive strength.

### **1. INTRODUCTION**

SCC is relatively a recent innovation and it has been considered as one of the great developments in construction. The high fluidity is main property of SCC and it can be placed under its Self-Weight without vibration (Domone, 2006a, 2006b; Gaywala et al. 2011; Okamura and Ozowa 1995; Okomura and Ouchi, 2003). In order to get SCC of high flowability without segregation during the transportation, the use of high powder content, super plasticizers and admixtures seems a legitimate solution (Domone, 2007; Murthy et al. 2012). However, the cost of such concrete is high-rise buildings. The use of mineral additives such as silica fumes (SFs), Fly ash (FA) could decrease the cost of the materials and boost the self-compatibility. Different studies have indicated that natural pozzolana have been widely used as a substitute for Portland cement in many applications due to its advantageous properties that include cost-reduction, reduction in heat evolution, decreased permeability and increased chemical resistance (ramanathan et al 2013; EFNARC,2002; Shetty 2012). Marble powder (MP is described as an inert material obtained as an industrial by-product during sawing, shaping, and polishing of marble which has also successfully been utilized as an addition in SCC.) However, sometimes when marble powder mixed with other materials such as fly ash and slag will not give a good result because of the phenomena of the segregation and bleeding that occurred during the slump test results. Furthermore, marble powder that used as a mineral addition of cement is reported to enhance some properties of fresh and hardened self-compacting concrete (SCC) (Belaidi et al. 2012; Tayep et al. 2011).

# 2. MATERIALS AND METHODS

### Materials

In this study, CEM-I 42.5 R Portland cement which is in compliance with EN 197-1 standard was used. Sieve analysis as aggregate was used as aggregate of crushed stone and silica sand of limestone origin in accordance with TS 3530 EN 933-1. 5, 10 and 20% fly ash and marble powder were used as substitutes for cement in the mixture.

Fly ash (FA) was used as mineral additive in the concrete mixture and fly ash was obtained from Çatalağzı Thermal Power Plant. Physical and chemical properties of cement and fly ash are given in Table 1. The polycarboxylate based superplasticizer (SP) was used in the mixture.

| <b>Chemical Composition</b>           | CEM I 42,5 R | FA    |
|---------------------------------------|--------------|-------|
| CaO                                   | 63,48        | 1,73  |
| SiO <sub>2</sub>                      | 20,35        | 55,73 |
| Al <sub>2</sub> 0 <sub>3</sub>        | 4,47         | 29,76 |
| Fe <sub>2</sub> O <sub>3</sub>        | 3,8          | 5,41  |
| Na <sub>2</sub> O                     | 1,43         | 1,96  |
| K <sub>2</sub> 0                      | 0,19         | 3,11  |
| MgO                                   | 1,02         | 3,3   |
| SO <sub>3</sub>                       | 2,26         | 0,3   |
| Free Lime                             | 1,3          | -     |
| Loss of Glow                          | 2,63         | -     |
| Insoluble Residue                     | 0,65         | -     |
| Spesific gravity (g/cm <sup>3</sup> ) | 3,1          | 2,2   |
| Blaine (cm <sup>2</sup> /gr)          | 3200         | 3700  |

Table 1. Chemical and physical properties of cement and fly ash

### Methods

In the experimental study, three batches were produced, one for control and three for each batch (Table 2). Fly ash and marble powder were substituted by 5, 10 and 20% of cement. Aggregate and substitute materials were mixed dry in the concrete, then 1/3 of the mixing water was added and stirring was continued. After mixing, cement and fly ash were added to the concrete until the homogenous mixture was maintained, and 1/3 of the mixing water was added to the superplasticizer was added to the concrete for 3 min. then the mixing was terminated. 10 x 10 x 10 cm cube specimens were placed in the sample containers for 3, 7 and 28 days compressive strength. The specimens were covered with an impermeable cover for 24 hours at  $23 \pm 2$  ° C and the relative humidity of 55-60%, then the samples taken from the sample cup were kept in lime saturated water at  $20 \pm 2$  ° C until the test day. Compressive strength according to TS 12390-3 was applied to the samples.

| <b>Table 2.</b> Mix design (kg/m | Table | 2. | Mix | design | $(kg/m^3)$ |
|----------------------------------|-------|----|-----|--------|------------|
|----------------------------------|-------|----|-----|--------|------------|

| Sr No | Type of Mix      | W/P ratio | Cement | FA    | Sand | Marble | Coarse Aggregate |        | Watar   | сD          |
|-------|------------------|-----------|--------|-------|------|--------|------------------|--------|---------|-------------|
| 5r.10 |                  |           |        |       |      |        | (0-5)            | (5-15) | vv ater | <b>5.</b> P |
| 1     | (0% MP + 0% FA)  |           | 4088,0 | 0     |      | 0      | 2141             |        |         |             |
| 2     | (0% MP + 5% FA)  |           | 3883.6 | 204.4 |      | 0      | 2141             |        |         |             |
| 3     | (0% MP +10%FA)   |           | 3679.2 | 408.8 |      | 0      | 2141             |        |         |             |
| 4     | (0% MP + 20% FA) | 0,34      | 3270.4 | 817.6 | 4435 | 0      | 2141             | 4033   | 1374    | 132         |
| 5     | (5% MP +0% FA)   |           | 3883.6 | 0     |      | 204.4  | 2141             |        |         |             |
| 6     | (10% MP + 0% FA) |           | 3679.2 | 0     |      | 408.8  | 2141             |        |         |             |
| 7     | (20% MP + 0% FA) |           | 3270.4 | 0     |      | 817.6  | 2141             |        |         |             |

# **3. RESULTS**

## **Fresh Property of SCC**

The following specifications are determined in accordance with EFNARC rules, which are accepted worldwide:

### **Filling Capability:**

The ability of the SCC to flow and completely fill all gaps in the mold under its own weight. The ability to fill is generally measured by the sedimentation flow (Figure 1) and the cubes in which the concert is performed (Figure 2). According to the EFNARC manual, the Flow should be between 70 and 80 cm.



Figure 1 Slump flow



Figure 2 Mix groups

Table 3. Fresh properties of concrete with slump test

| Sr. No | Type Of Mix      | Slump (cm)75±5 cm |
|--------|------------------|-------------------|
| 1      | (0% MP +0% FA)   | 80cm              |
| 2      | (0% MP +5% FA)   | 79cm              |
| 3      | (0% MP +10%FA)   | 75cm              |
| 4      | (0% MP +20%FA)   | 72cm              |
| 5      | (5% MP +0% FA)   | 75cm              |
| 6      | (10% MP + 0% FA) | 80cm              |
| 7      | (20% MP + 0% FA) | 80cm              |

### Hardened Properties of SCC

Compressive strength is the average of at least three standard cured strength specimens made from the same concrete sample and tested at the same age. The vast majorities of cases strength requirements for concrete are at an age of 28 days of curing. After 28 days, the concrete cubes were examined for the purpose of their compressive strengths.

During the test comparison, the same size of cube 10x10x10 cm was made by Self Compacting concrete (SCC). In addition, test specimens of Self Compacting concrete (SCC) cube was cured into water for 3, 7 and 28 days. The results of the mixtures are given in Table 4.

Table 4. Compressive strenght (MPa)

| Sr No | Type of Mix      | 3 Days | 7 Days | 28 Days |
|-------|------------------|--------|--------|---------|
| 1     | (0% MP +0% FA)   | 55.16  | 62.08  | 68.20   |
| 2     | (0% MP +5% FA)   | 65.56  | 70.67  | 81.46   |
| 3     | (0% MP +10%FA)   | 53.10  | 59.52  | 70.80   |
| 4     | (0% MP +20%FA)   | 41.86  | 54.56  | 59.57   |
| 5     | (5% MP +0% FA)   | 60.38  | 63.78  | 59.57   |
| 6     | (10% MP + 0% FA) | 58.55  | 59.87  | 67.56   |
| 7     | (20% MP + 0% FA) | 53.77  | 60.42  | 67.08   |

#### 4. CONCLUSION

The marble powder and fly ash usage by substitution to cement has no direct negative effect of workability of SCC. For instance, in fresh property such as filling and passing ability increases by using of 20% marble powder and 5% fly ash by substitution by cement in binder material. In addition, the increase of marble powder in SCC and slump flow shows a direct relationship between them. Moreover, the increase of marble powder in SCC increases the passing ability. In compressive strength would be taken into account and marble powder can be used up to 20% and fly ash to 5%.

As a result, it was observed that the contribution of fly ash in SCC was more effective than the contribution of waste marble dust and could be used as powder material.

### REFERENCES

- Belaidi, A. S. E., Azzouz, L., Kadri, E., & Kenai, S. (2012). Effect of natural pozzolana and marble powder on the properties of selfcompacting concrete. Construction and Building Materials, 31, 251-257.
- Dhiyaneshwaran, S., Ramanathan, P., Baskar, I., & Venkatasubramani, R. (2013). Study on durability characteristics of selfcompacting concrete with fly ash. Jordan journal of civil engineering, 159(3164), 1-12. Domone PLJ. 2006a. Mortar tests for material selection and mix design of SCC, Concrete International.
- Domone, P. L. (2006). Self-compacting concrete: An analysis of 11 years of case studies. Cement and concrete composites, 28(2), 197-208.
- Domone, P. L. (2007). A review of the hardened mechanical properties of self-compacting concrete. Cement and concrete composites, 29(1), 1-12.
- EFNARC. 2002. Specification and guidelines for self-compacting concrete. European Federation of Producers and Applicators of Specialist Products for Structures.
- Gaywala, N. R., & Raijiwala, D. B. (2011). Self compacting concrete: A concrete of next decade. Journal of Engineering Research & Studies, 2(4).
- Murthy, N. K., Rao, N., Reddy, I. R., & Reddy, M. V. S. (2012). Mix Design procedure for self-compacting concrete. IOSR Journal of Engineering, 2(9), 33-41.

Okamura H, Ouchi M. 2003. Self-compacting concrete. Journal of Advanced Concrete Technology 1(1):5-15.

- Okamura H, Ozawa K. 1995. Mix design for self-compacting concrete. Concrete Library of Japanese Society of Civil Engineers 25(6):107-120.
- Shetty. M. S., 2012." Concrete Technology" (Theory and Practice), S.Chand & Company Limited, New Delhi, Seventh Edition.
- Tayeb, B., Abdelbaki, B., Madani, B., & Mohamed, L. (2011). Effect of marble powder on the properties of self-compacting sand concrete. The Open Construction and Building Technology Journal, 5(1).



# Design and Simulation of LED Driver Circuit with PFC

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**Abstract:** This work aimed at analyzing power factor corrected AC/DC Converter for LED driving purpose. The study involves analysis, circuit design, simulation and evaluation.

In recent years, the use of LEDs in lighting applications has become common due to their highest efficiency. Based on this fact, drivers are required to improve their efficient capability. As a result of research, efficient drivers must have long lifetime, small amount of components, high power factor and low harmonic distortion.

Behind of efficient capability idea, the chosen topology should make easy to control and design low cost line for low powered lighting application. Proposed Led driver designed in boost converter topology with single stage power factor correction and hysteresis current control mode. The design easily maintains output current protection, voltage step-up regulation and constant output voltage with only one semiconductor switch.

In this context, Boost Type Converter LED driver design has been established and simulated in PSIM application. As can be observed, proposed LED Driver's input current waveform matches nearly pure sinusoidal waveform which one of the basic of design. The results of simulation; low total harmonic distortion and high-power factor and efficiency values support our assertion.

Keywords: LED Driver, Boost Converter, Power Factor Correction, Single Stage PFC, Hysteresis Current Control

### **1. INTRODUCTION**

The development of semiconductor technology led to the fact that LED lighting could be used in various fields and it could replace conventional lighting. However, because of the LED's "lighting emitting diodes" structure, LED systems require a driver circuit; and despite many advantages stemming from the use of the LED systems, when the driver circuits are not designed properly, there could be many cases of shifting in light spectrum, power and lifetime decrease. For driving LED units, passive circuit components or linear regulators can be used. But the use of SMPS topologies is becoming necessary especially in the case in which the power efficiency or international standards are discussed. SMPS is used in various fields because of its different topologies with different solutions and it could obtain a considerable place in power electronics due to the fact that it is considerably efficient and open to new developments.

The increase of the use of power electronic devices caused several negative impacts such as harmonics in electric networks, reactive power losses, distortions at the frequency value. Although there are many ways that are used to prevent these distorting effects, the most popular methods have been applied by power electronic solutions. The standards and restrictions which are internationally recognized, require to resolve the negative impacts within the power electronic devices resulted by these devices and not to reflect these impacts to electric networks.

Within the scope of this study, it is aimed to design of a power factor corrected boost type AC/DC converter that obtains voltage regulation, current protection, constant voltage. After the design of this converter that is considered for high powered LED lighting applications, converter design has been simulated in PSim software by using the idealized values. The principal aim of this simulation is to observe the quality of the AC sinusoidal obtained from the electric grid, the obtained power factor, the total harmonic distortion and the fact that the circuit could supply with the standard requirements "EN 61000-3-2 C" that is necessary for the lighting application of a circuit.

### 2. MATERIAL AND METHODS

#### **Boost Converter**

It is also known as Step-Up Converter. This converter is used to obtain a higher output voltage than the one in input. When the power switch is turned on, the inductance is connected to the rectified source and the energy in the source is stored in this inductance. When the switch is turned off, the inductance current is forced to flow on diode and load. The voltage in the inductance is negative [5]

The output voltage is given as Vout = Vin/(1-D) (1)

In this case, D symbolizes the duty cycle of the switch and D=ton/T (2)

T symbolizes the time of the period. 1/T is the switching frequency of semiconductor switch. [4]

Due to the fact that D varies within the marge of 0 < D < 1, the load voltage will vary within the marge of Vout, Vin < Vout  $< \infty$ 

The inductor and output capacitor can calculate by bellowed equations. Output ripple voltage can see as  $\Delta Vo$  in capacitance equation. Ripple current shown as  $\Delta Io$  in inductance equation.

$$C = \frac{D}{R(\Delta Vo/Vo)f}$$
(3)

$$L = \frac{V_S \times D}{f_S \times \Delta Io} \tag{4}$$

#### Active PFC Circuit Based on Boost Converter

The boost topology that is used the power factor correction methods could function in intermittent and uninterrupted flow modes. The study, known as transition mode or critical flow mode is the study in which the switching frequency is adjusted within the boundaries of intermittent and uninterrupted flow modes. In Fig.1 and Fig.2, the boost converter with the uninterrupted flow mode and the wave shapes could be seen. This topology gives the input voltage to the output, by increasing this voltage during this process. The converter could function in the period of grid, that is the reason why the input voltage does not have transition ripple. This situation prevents the grid current distortion on a very close point zero transition. Moreover, the input switching current of the converter is continuous, because it has a serial connection with the boost inductance. The high frequency S switch has no impact on the input current. Hence, the input current has less high frequency content; there is less EMI and smaller filters are sufficient. The Co output capacitance limits the voltage at the moment of turn-off of the S switch to the output voltage on the diode and it protects the switch.



Figure 1. Active PFC Circuit Based on Boost Converter [3]


Figure 2. Voltage and current signal forms of Continuous Conduction Mode PFC Boost Converter [3]

On the converter above, the control method determines whether the inductance current would be continuous or discontinuous. The discontinuous-conduction mode works in a particular frequency and there are some interruptions on switching current. They are used in very limited occasions due to the high current peaks and EMI. These high peaks stem from after the idle times needed by sudden input voltages, necessary to protect the discontinuous-conduction mode in all input variations. On the other hand, the critical-conduction mode converter uses a variation of hysteresis current control in a way in which the lowest limit of the converter is zero current. This technical variable is a technic of frequency control; while the input current control is naturally stable, it eliminates the reverse recovery losses of rectifier. As the turn-on time within the given current and voltage parameters remains same, the turn-off time variable. As a result, the switching frequency of power converter obtains its highest value while instant input voltage is in the lowest value.

For the power factor correction, as it can be seen in the Fig.2, the input current of the boost front regulator is forced to be in harmony with the waveform of the input voltage, when the switch is turned on and turned off on a very high frequency. This power factor correction control circuits pursue a control about the current on the boost inductance by using PWM pulses. The inductance becomes a controlled current source when the operating frequency is chosen in a level in which the inductance is in continuous conduction. The reference of the rectified source voltage and current waveform and leads to the fact that the inductance current is in the same phase and sinusoidal with the source voltage to provide a high-power factor.

#### **Control of Power Factor Corrected Converter**

Power factor corrected AC/DC converters are power electronic devices identify with PFC circuits and PFC technique terms at these days. These converter's one of the most important advantages are simple structure, easy to control and highly viable. [1]

Inherent of single-phase power corrected AC/DC converters acquire by PFC unit connected to output of single phase fullwave rectifier and one or more high frequency DC/DC converter. While Continuous-conduction mode (CCM) is preferred to high power applications, discontinuous-conduction mode (DCM) is preferred to low power for reduce to power losses. Main current can observe as highly frequent sinusoidal tracking and continuous current signal along the period rather than passive methods. In this way, power factor can be close to one and THD rate can reduce. [2]

Single stage converters are improved by combine of PFC unit's and DC regulator convertor's switches and control loops. Semiconductor component's control signal trigger to regulation and PFC occurs by itself. However, PFC and regulation aren't quality as much as two stage converters but even in this condition single stage converters are highly demanded because of low cost and easy to control characteristic.



Figure 3.1. Single-Stage PFC Converter

Figure 3.2. Two-Stage PFC Converter

In our work, hysteresis current control method is used for semiconductor switch component control in power factor corrected LED driver design.



Figure 4.1. Principle Block Diagram of Hysteresis Control



Figure 4.2. Hysteresis Control's Input Current Waveform

Hysteresis control's block diagram and input current's waveform can be observed at Fig.4.1. and Fig.4.2. In this control method aims to limit the inductance real current between two different sinusoidal signals under variable frequency.

when increasing inductance current reached to upper reference current, Switch signal goes off and once again inductance current fall to lower reference current, switch signal goes on. Hereby Switch frequency becomes variable.

Even though input current signal is smooth, this control method has disadvantages like hard switching problem at main diode and main switch.

# **3. RESULTS AND DISCUSSION**

#### Simulation

Single-stage PFC Power LED driver circuit has been designed at PSim simulation software. Design structure based on Boost topology and hysteresis current control. Power LED is modelled with ideal diode and 40ohm resistor in simulation.

After usage of suitable circuit component (Fig.5.1.), established circuit is simulated with 50ns time steps for a 100ms. Allowed upper and lower limit of hysteresis current control set to  $\pm$ %10 increasing/decreasing (total allowed bandwidth %20) from ideal sinusoidal current waveform.



Figure 5.1. PSim Block diagram of Single-stage Hysteresis controlled PFC LED Driver

Simulation is run several times with 50ns between 80-100ms and 0-100ms. Transient analysis practiced on simulation diagrams. Determined regime observed approximately 50-60ms after simulation start.

#### **R-C Selection for Voltage Error Amplifier (PI Controller)**

Voltage reference set 5 Volt DC for voltage error amplifier. Various PI controller values which taken from similar PI controller designs, tested in simulation. As observed after numerous tests, desired values also support formula at below and this value set for voltage error amplifier.

$$fc = 1/2\pi RC \, Hz \tag{5}$$

#### **Actual Current Sample**

Actual current sample measured from return of circuit line by current sensor and its small-signal form with different sensor gain will compare to 5sinwt signal values taken multiplier output. But peak current measured 27.092A by Iret amperemeter. Comparator current was 5A sinusoidal signal, because of that current sensor's gain rate must reduce for proper comparison. It will cause shrinkage and resemble to 5sinwt at output of current sensor value. Selected gain rate is 0.185 for current sensor in simulation.

#### **Simulation Diagrams**



Figure 5.2. Input and Output Voltage Signals



Figure 5.3. AC Input Voltage and Current Signals



Figure 5.4. IGBT Gate Trigger Signal with Different Time Periods



Figure 5.5. Comparison of IGBT Gate Signal and Inductance Current Signal



Figure 5.6. AC Input Current Sample of Half Period

| Real Power |        |                | × |       | Ар     | Apparent Power |  |  |
|------------|--------|----------------|---|-------|--------|----------------|--|--|
| Time       | From   | 8.0000050e-002 |   | Time  | From   | 8.0000050e-002 |  |  |
| Time       | То     | 1.0000000e-001 |   | Time  | То     | 1.000000e-001  |  |  |
| Vac v      | s. lac | 3.7851045e+003 |   | Vac v | s. lac | 3.7931676e+003 |  |  |

Figure 5.7. Measured Real and Apparent Power Values

| Pe          | ower Factor        | × | THD                   |                   |  |  |  |
|-------------|--------------------|---|-----------------------|-------------------|--|--|--|
| Time From   | rom 8.0000050e-002 |   | Fundamental Frequency | 5.0000000e+001 HZ |  |  |  |
| Time To     | 1.000000e-001      |   | lac                   | 6 53064576-002    |  |  |  |
| Vac vs. lac | 9.9787433e-001     |   | NGC 1                 | 0.00004076-002    |  |  |  |

Figure 5.8. Measured Power Factor and Total Harmonic Distortion

### 4. CONCLUSIONS

Nowadays, LED lighting and power LEDs are highly demanded technologies, it's expected to demand is increasing and will increase more with new improvements. Driver circuits also have big importance and role in that improvements. In this study, proposed design is a power factor corrected boost type Power LED driver aim to offer simple structured and low cost specifically.

Basically, the proposed circuit steps up input voltage from AC mains to 400Vdc output continuous voltage. The study investigates possibility to drive a LED module with this PFC boost type convertor.

Simulated LED driver circuit has been established with one IGBT switch and one diode and it performed to its characteristic only two semiconductor components. As a results of simulation power factor value measured 0,997, THD observed %6,5 (Fig.5.8) and AC input current signal limited to wave set values. Quality of AC input current signal waveform can observe at Fig.5.6.

Opinions are noted following lines which acquired from this simulation work will consider to in later studies for improve the techniques.

EMI filter should use before bridge rectifier to reduce interference.

IGBT used for because of high output power level at power LED load. At the low power and high frequency level MOSFETs should be used

Switching frequency is variable because of hysteresis current control increase harmonic effects

Hard switching on diode and semiconductor switch

Occurred current and voltage stress can observe on components

Harmonic losses and efficiency values possible to improve to use of additional snubber circuits and soft switching.

The output current protection in case of higher and constant output voltage regulation condition must be known and LED load model should be chosen in accordance with this case.

Measured power factor supplies to required standards in good condition like "EN 61000-3-2 C" for lighting application. Besides measured THD is open to improvement but it could reasonable at low-cost design. When proposed design is built up again with non-ideal, real values, can be observed decreased power factor and increased THD.

### REFERENCES

- [1] Bodur, H., (2011). Güç Elektroniğinde Güç Faktörü Düzeltme Yöntemleri Ders Notu, Y.T.Ü. Elektrik Elektronik Fakültesi Elektrik Mühendisliği, İstanbul.
- [2] Şahin E, Büyükkatırcı O, Akın B, AC-DC Dönüştürücü Tabanlı Güç Faktörü Düzeltme Devresi Tasarımı ve Uygulaması "Eleco 2014 Sempozyumu", Yıldız Teknik Üniversitesi Elektrik Mühendisliği, İstanbul
- [3] Basu, S., (2006), "Single Phase Active Power Factor Correction Converters", Chalmers University of Technology, Sweden.
- [4] Matsuo, M., Matsui, K., Yamamoto, I., Ueda, F., (2000), "A Comparison of Various Dc-Dc Converters and Their Application to Power Factor Correction", IEEE 2000, Japan.
- [5] Rossetto, L., Spiazzi, G., Tenti P., (1999), "Boost PFC with 100-Hz Switching Frequency Providing Output Voltage Stabilization and Compliance with EMC Standards", IEEE Transactions on Industry Applications, Vol.36 No.1, Italy.



# Crystallization in Honey and Creamed Honey

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**Abstract:** Honey, having an animal and vegetable origin and a high nutritional value, has a very important place in nutrition. The crystallization of honey is a natural event. Honey is a highly concentrated sugar solution. Honey contains more than 60% sugars. The two principal sugars in honey are fructose and glucose. The glucose in Honey reaches saturation and crystallizes physically. Honey contains less than 20% water. Flower honey tends to crystallize. The rate of crystallization depends on the glucose, fructose ratio and water content in honey. Crystallization is late in immature honey. The crystallization times of the honeys also vary. The crystallization. Change occurs only in the physical view. The optimal temperature for crystallization is around 14 °C. Many factors affect crystallization in honey. The producers apply a high temperature to the crystallized honey so that the frozen honey does not freeze or the frozen honey is restored, which increases the amount of HMF harmful to human health. Honey is crystallized under controlled conditions, creamed honey is obtained. Creamed honey is composition, taste, nutritive value and quality are not different from run honey. In children's balanced diet, creamy honey in non-flowing and drier consistency is very important. Creamed honey has a soft texture and can be drivable feature to bread. The consumption of creamed honey in the world is widespread. But it is not common in Turkey.

Keywords: Honey, Crystallization, Creamed Honey, Fructose, Glucose,



# Lycopene: Chemistry, Sources, Bioavailability, and Benefits for Human Health

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Abstract: For decades, recommendations for the consumption of fruits and vegetables are increasing because of being rich sources of various phytochemical compounds. There are mostly bioactive substances that show beneficial and nutritional actions. Lycopene is a member of the carotenoid's family. It exhibits close characteristics and functions of some carotenoids that promote it to be an excellent fighter against a series of health problems and diseases. Lycopene is a red pigment which can be found in fresh fruits like tomato, rosehips, watermelon, autumn olive and in their processed products. Type of variety and several environmental factors have major influences on the lycopene contents of fruits. Moreover, the conditions during and after processing could lead to different variations in its content in the final products. Beyond being a color agent, this substance has other functions in its original environments (fruits) to protect them against oxidative stresses, harmful lights, and others. Due to its hydrophobicity and the intake of oils and fats, the bioavailability of lycopene is found to be high after the consumption with lipophilic rich products. After that, this substance would be concentrated in some organs and tissues where it will exert its numerous health effects. Lycopene is considered as an effective antioxidant and agent against several cancer types such as prostate and skin cancer. Also, it has a crucial role to reduce cardiovascular diseases, neurological impairments, diabetes, and related diseases. For this reason, this paper has reviewed the main characteristics of lycopene and its important sources in the human diet. Many recent studies in the literature have reported the efficient contribution and the relation of this substance to fight different diseases. Furthermore, the supplementation of foods with this potential active compound is recommended to be a good alternative to many existing treatments without showing any side effects.

Keywords: Lycopene, antioxidant, health

#### **1. INTRODUCTION**

Fruits and vegetables contain two essential metabolites. The primary metabolites are the organic compounds which are directly associated with the growth and the development of the plant. The secondary metabolites play important roles in the reproduction (attraction of pollinators), pigmentation of plants (flowers, leaves, fruits...). Also, human use the secondary metabolites for specific applications as fragrances, dyes or for even biological and medicinal purposes (Verpoorte & Memelink, 2002; Kumari et al., 2017).

In the last decade, studies have focused on the different classes and main functions that these metabolites provide. It has been discovered that the secondary metabolites which present biological activities are found in three major groups of compounds, phenolic compounds, terpenes and steroids, and alkaloids (Bourgaud et al., 2001; Siddiqui et al., 2017). Belonging to the family of terpenoids, carotenoids are a part of these secondary metabolites. The biosynthesis of these tetraterpenoids starts by the formation of isopentenyl diphosphate and followed by several reactions (desaturation, isomerization, cyclization, an introduction of oxygen...) leads to the formation of various types of carotenoids (Fraser & Bramley, 2004; Rodriguez-Amaya, 2019).

Depending on the chemical structure (cyclic, acyclic) and on the presence or absence of oxygen and other functional groups (hydroxyl, epoxy groups), carotenoids can be classified into many types (Stephen et al., 2017). Until today, more than 600 carotenoids have been characterized in plants and organisms. However, only 5 principal types including lycopene, lutein,  $\beta$ -carotene,  $\alpha$ -carotene, and cryptoxanthin which are fat soluble, mostly from plant origin, are found in human plasma (Chidambara Murthy et al., 2019). The chemical structure of the compounds affects their color characteristics, antioxidant activities, and biological activities against diseases (Rao & Rao, 2007).

In order to take benefits from all the advantages of carotenoids, they find an increasing use in the food industry, cosmetics, and pharmaceutical industry. In the food industry, and in order to increase satisfaction and to attract the consumers, the carotenoids are added to foodstuffs to improve the color and to increase the visual perception (Ramesh & Muthuraman, 2018). Lycopene is one of the important carotenoids in foods. this review has discussed the chemical structure, the bioavailability, the antioxidant activity, the main sources of the lycopene, and also its health promoting functions.

#### Lycopene Chemistry

Nature has a plenty of natural compounds that give plants, animal and even microorganisms their specific color. Carotenoids are among those compounds that contribute coloring of the fruits and vegetables (Bruno et al., 2007). Lycopene similarly to other carotenoids is a natural pigment principally synthesized by plants (Gerster, 1997). Also, it can be found in microorganisms and in some animals such as lobster and Flamingo (Clinton, 1998). It may give a color that ranges from pink to red (Rocha et al., 2015). Besides its contribution to color of fruits and vegetables, lycopene is known to have a role to absorb light during photosynthesis and consequently protects plants from photosensitization (Rao & Agarwal, 1999).



**Figure 1.** The structures of lycopene and pro-lycopene (1) All-trans-lycopene, (2) prolycopene, (3) 13-cis-lycopene, (4) 9-cis-lycopene, (5) 5-cis-lycopene (Lowe et al., 2018).

Lycopene is a carotenoid with simple hydrocarbon structure consisting of 8 isoprenoids that give the known tetraterpenoids ( $C_{40}H_{56}$ ). It is characterized with long double bonds that has 11 conjugated double bonds which give its interesting biological and physiological activities (**Figure 1**) (Rocha et al., 2015; Lowe et al., 2018). The predominant natural lycopene occurs in all-trans form. Its many other geometric isomer forms, mono- or poly-cis isomers can occur under conditions like high temperatures, oxygen, and catalysts (Stahl & Sies, 2003; Rocha et al., 2015).

Human plasma contains a mixture of lycopene isomers, up to 50% of total lycopene, are cis forms (Rao & Agarwal, 1999). Furthermore, in the last years, apolycopenoids (cleavage products of lycopene) have been detected in human plasma (Lowe et al., 2018). Like other carotenoids, lycopene is a hydrophobic compound soluble in oil and non-polar solvents (Sandei, 2018). However, and in contrary to  $\beta$ -carotene which is a cyclic molecule, the lycopene is among the acyclic carotenes. In addition to that, in polar media and at high concentration, it tends to aggregate and precipitate as a sort of crystals (Lowe et al., 2018).

#### Lycopene Sources

The main food sources of lycopene in our diet are fruits and vegetables, predominately fresh tomatoes and tomato processed products such as ketchup (Gerster, 1997). However, red fruits such as watermelon, grapefruit or even seafood are sources but at a low contribution compared with that of tomatoes and its products (Kohlmeier et al., 1997). In addition, other new sources such as red carrot, bitter melon, rosehip and autumn olives are among the main sources of lycopene (Kohlmeier et al., 1997). The fresh fruits of Autumn olive have a lycopene contents which ranges from 5 to 20 time compared to that of tomato and its products (Fordham et al., 2002).

Lycopene content is influenced by several factors such as fruits variety, degree of ripeness, the environmental conditions (light, irrigation, temperature, climate, planting location), storage and processing conditions (Rocha et al., 2015; Sgherri et al., 2015). During fruit ripening, the total chlorophyll levels decrease to disappear, whereas lycopene levels increase (Sandei, 2018). Moreover, supplemental lights (red, blue) on tomato had promoted the lycopene synthesis in fruits (Xie

et al., 2019). Also, lights and oxygen are among the important influencing factors during processing and storage of lycopene containing products (Xianquan et al., 2005). Murakami et al. (2018) proved the instability of lycopene under light irradiation by promoting its isomerization.

Unlike the other studied bioactive compounds (quercetin, ascorbic acid, kaempferol) in tomato paste (hot break production), lycopene was the most stable bioactive compounds even under the subsequent evaporation and sterilization, with only 20% of loss (Koh et al., 2012). It was suggested that the existing of antioxidant (e.g. ascorbic acid, phenolic compounds) could be responsible for stability during processing. Moreover, the presence of lycopene in the fruit (tomato) or in some kinds of matrices could provide more stability (autoxidation, isomerization) than the pure forms lycopene (Takeoka et al., 2001; Xianquan et al., 2005).

The storage stability of tomato juice had been studied. It was found that the post-processing stability of lycopene was strongly linked to cultivar, storage, time, and processing method (Grupta et al., 2010). After processing of lycopene sources, lycopene is liberated. Due to its hydrophobic nature, processed lycopene has shown more availability than that from raw tomatoes (Bruno et al., 2007, Sgherri et al., 2015). The lycopene contents of some food sources have shown in Table 1.

| Emite como finite producto                         | Lycopene(µg) Per | Emita como fruita producta                        | Lycopene(µg) |
|--|------------------|---|--------------|
| Fruits, some fruits products                       | 100 g            | Fruits, some fruits products                      | Per 100 g    |
| Tomatoes, sun-dried                                | 45902            | Papayas, raw                                      | 1828         |
| Tomato products, canned, puree, without salt added | 21754            | Grapefruit, raw, pink and red, all areas          | 1419         |
| Catsup   | 12062            | Beverages, Acai berry drink, fortified            | 899          |
| Vegetable juice cocktail, canned                   | 7119             | Peppers, sweet, red, sauteed                      | 484          |
| Guavas, common, raw                                | 5204             | Grapefruit juice, pink or red, with added calcium | 297          |
| Tomatoes, crushed, canned                          | 5106             | Sapote, mamey, raw                                | 199          |
| Watermelon, raw                                    | 4532             | Persimmons, Japanese, raw                         | 159          |
| Tomatoes, red, ripe, canned, stewed                | 4088             | Guava nectar, canned, with added ascorbic acid    | 35           |
| Guava sauce, cooked                                | 3909             | Asparagus, cooked, boiled, drained                | 30           |
| Tomatoes, red, ripe, cooked                        | 3041             | Cabbage, red, raw                                 | 20           |

Table 1. Lycopene content in some food sources (USDA, 2019).

# **Bioavailibility of Lycopene**

The lycopene is not synthesized in human body (Kirpichenkova et al., 2018). After the ingestion of foods and its release in the duodenum, lycopene, similarly to other carotenoids are combined in complex with fatty acids and bile acids. As a result, lycopene containing micelles has been formed. These micelles consist of shells of hydrophilic nature, where the hydrophobic core contains lycopene (Lowe et al., 2018). After that, lycopene is absorbed and packaged in small intestine into chylomicrons, via a passive controlled diffusion. Subsequently, it is secreted into the lymphatic system and bloodstream and finally delivered to liver and body organs (Figure 2) (Bruno et al., 2007; Sandei, 2018). It is known that lycopene is among the carotenoids which concentrate at high levels found in adrenal glands, testes and prostate (Rao & Agarwal, 1999).

Tomato products, like ketchup, is an excellent source with high concentration of lycopene. Furthermore, tomato processing leads to liberate lycopene from its binding matrices, and the addition of lipophilic phase (oils) make the lycopene more available for micelles formation and the subsequent intestinal absorption (Giovannucci et al., 2002). Therefore, any factors that cause lipid malabsorption (dietary compounds, drugs, diseases) may lead to disrupting the micelle formation process and by consequence the reduce the bioavailability of carotenoids as well as lycopene (Bruno et al., 2007).





#### Antioxidant Activities of Lycopene

Foods, mainly of plant origin, are important natural sources for a mixture of antioxidant agents such as vitamin C (ascorbic acid),  $\alpha$ -tocopherol and carotenoids (Arnao et al., 2001). The light absorbing capacity that characterized by the presence of double bonds in the polyene backbone in carotenoids. They offer also a substantial antioxidant capacity to reduce the oxidative stress in plants. And also, these effects are seen in animals and human who consumed these compounds (Stahl, & Sies, 2003).

Carotenoids such as lycopene, are also a known as efficient antioxidant which have a role in scavenging free radicals (Komurogluet al., 2018). They display an ability to disrupt the autooxidation of lipids by quenching lipid peroxyl radicals at initial steps by electron transfer, hydrogen abstraction or by addition (Lowe et al., 2018). This carotenoid remains undamaged after the transfer of energy from singlet oxygen, which enable it to undergo many cycles of quenching (Bruno et al., 2007).

Several health benefits are hypothesized to be linked with carotenoids consumption, and substantially by lycopene which has the highest activity because of their ability to prevent the oxidative damages. In general, due to the lipophilic nature of carotenoids, lycopene is mostly located in cell membranes and in lipoprotein components. (Rao & Agarwal, 1999).

Recently, the relationship between the antioxidant activity of lycopene and its contribution in reducing diseases have markedly investigated. Xu et al. (2019), have shown that lycopene supplementation could help to decrease the oxidative stress in hemodialysis patients. Arnao et al. (2001) reported that a mixture of hydrophobic and hydrophilic antioxidant activities in samples, could contribute to the total antioxidant capacity.

Also, the combination of lycopene and anthocyanins at some levels, have affect the uptake of lycopene and may decrease the cellular antioxidant activity (Phan et al., 2019). The antioxidant combination composed of lycopene and green tea extract have shown an improvement in clinical parameters in gingivitis patients, and it is concluded that such supplementation could be promising for periodontal health (Tripathi et al., 2019).

#### Health Benefits of Lycopene

Besides in food industry, the lycopene sources have been widely used in medicine, in cosmetics and other areas (Song et al., 2019). The wide uses of lycopene are arised from its various beneficial health effects. Many studies had focused on the protective effects of lycopene against diseases such as cardiovascular and neurological diseases, prostate and skin cancers and diabetes.

#### Lycopene and Cardiovascular Diseases

Cardiovascular diseases (CVD) are among diseases responsible for the morbidity and mortality causes around the world. Various factors are included in the etiology of CVD, mainly the oxidative stress and the inflammation. For instance, the

oxidation of low-density lipoproteins (LDL) in vessels could develop atherosclerotic lesions (Di Pietro et al., 2016). Despite the complex nature of CVD, several phytochemicals have been suggested as preventing against this group of diseases (Costa-Rodrigues et al., 2018). Dietary antioxidants such as carotenoids may play crucial activities against oxidation and inflammation, therefore they play a key role in reducing cardiovascular health risks (Maria et al., 2015; Di Pietro et al., 2016). In spite of the controversial cardiovascular health effects, in vitro studies have suggested lycopene to have a potential action to prevent CVD in humans (Arab & Steck, 2000).

Hypertension and ischemic heart diseases are implicated in inflammations that lead to heart failure complications. Biddle et al. (2015) have investigated the effect of lycopene on the inflammation in female patients with heart failure. They found the levels of CRP (C-reactive protein) decreased. It was suggested that this natural antioxidant could delay or prevent from those complications. Yilmaz et al. (2018) have carried out a study on the effect of lycopene in kidney and heart of rats exposed to Aflatoxin B1 (in terms of time and different doses). They found that the lycopene had protective effects against the nephrotoxicity and cardiotoxicity of Aflatoxin. Thanks to cheap, easily accessible, long shelf life and wide spectrum of impact (Maria et al., 2015; Biddle et al., 2015) lycopene may be the reason of preference in the protection against CVD.

### Lycopene and Prostate Cancer

Since decades, the consumption of lycopene-rich products such as tomato and tomato-based products, have been associated to decrease the risk of cancers. Lycopene intake has shown clear evidence to reduce prostate cancer and a frequent consumption is recommended (Giovannucci, 1999; Giovannucci, 2002). Jiang et al. (2019) have studied the therapeutic effects and the capacity to inhibit prostate cancer by lycopene at different doses. It was clearly found that lycopene treatment has reduced the progression of prostate cancer and suppressed the inflammatory responses. Its potential therapeutic contribution was dependent on doses and time of treatment. Overall, lycopene is considered as promising therapeutic dietary agent that has anti-cancer and anti-inflammatory properties which could substitute the chemical therapies and furthermore without any side effects.

### Lycopene and Skin Cancer

A correlation between the uses of lycopene and the reduction of skin cancer's risk has been established. The carotenoids in general act as light filters in biological systems to reduce light exposure that helps to prevent from oxidative stresses (Stahl, & Sies, 2003). The studies shown that UV lights could lead to photosensitivity, photoaging, erythema and increase the risk of cancer on the skin after its exposure to UV radiation, the generation of singlet oxygen, hydroxyl and other free radicals is induced. In general carotenoids, or specifically, lycopene is one of the substances that play a role in quenching those UV-induced oxidative compounds. As a result, it was suggested that lycopene could have a positive action against free radicals to hinder the occurring of possible carcinogenic effect in the skin (Gerster, 1997; Clinton, 1998).

#### Lycopene and Neurological Diseases

Some studies have demonstrated that lycopene and lycopene containing product have beneficial action to protect the neurological system. Hua et al. (2019) revealed that lycopene has relieved the spinal cord ischemia/reperfusion injury that induce neurological deficits, neuronal cell death, and neuroinflammation. Malekiyan et al. (2019) have found that lycopene alone or in combination with insulin, have exhibited an effective protection for hippocampal neuroglia against streptozotocin, and thereby have reduced the impairment of learning and memory. Due to its neuroprotective function and therapeutic action, it is suggested that lycopene could be an ideal candidate for the protection of neurological system from menacing disorders and inflammations.

#### Lycopene and Diabetes

Beside other diseases, many investigations have been done in relationship between lycopene and diabetes. Komuroglu et al. (2018) have studied the influence of lycopene administration on the activity of some enzymes as inflammation markers in rats during 28 days. They revealed that group of rats which had given lycopene, exhibited less inflammation markers. So, it's considered that lycopene is useful for the prevention complications of diabetes and related inflammation. Yin et al. (2019) have studied the effects of lycopene on diabetic rats. They found that the lycopene reduced the blood glucose level, improved lipid disorders, insulin sensitivity and insulin resistance. Consequently, the application lycopene and lycopene-rich products can be helpful for the prevention and treatment of diabetes and to regulate the metabolism of glucose and lipids.

#### Lycopene and Bone Health

For a long time, it is believed that lycopene could have beneficial implications in bones health. By several ways, lycopene interferes for the inhibition of hormones and reactive oxygen species which can be responsible for mineral resorption and osteoporosis (Rao et al., 2003). A study on postmenopausal women have been carried out by treating them with lycopene supplemented juices and capsules. The results brought an interesting finding in which the different supplementation forms of lycopene may have ability to reduce the bone resorption (Mackinnon et al., 2011). A study on female rats which received a daily lycopene treatment during 12 weeks, it demonstrated that the beneficial action of the compound to suppress the bone turnover and to improve their strength and their microarchitecture (Ardawi et al., 2016). As a result, lycopene in complement or supplementation forms could be a good alternative to improve and protect the bones health.

#### 2. CONCLUSION

Lycopene, one of the carotenoid compounds, has been reported to prevent the formation of many diseases such as cardiovascular, neurological, diabetes, prostate cancer due to its strong antioxidant properties. Some fruits and vegetables, such as tomatoes and watermelons, are important sources of lycopene. Lycopene, which has a lipophilic property, can be taken directly from the food or consumed as a food supplement. Consumption of lycopene resources is important for the protection of human health.

#### REFERENCES

- Ardawi, M. S. M., Badawoud, M. H., Hassan, S. M., Rouzi, A. A., Ardawi, J. M., AlNosani, N. M., ... & Mousa, S. A. (2016). Lycopene treatment against loss of bone mass, microarchitecture and strength in relation to regulatory mechanisms in a postmenopausal osteoporosis model. *Bone*, 83, 127-140.
- Arab, L., & Steck, S. (2000). Lycopene and cardiovascular disease. The American journal of clinical nutrition, 71(6), 1691S-1695S.
- Arnao, M. B., Cano, A., & Acosta, M. (2001). The hydrophilic and lipophilic contribution to total antioxidant activity. Food chemistry, 73(2), 239-244.
- Biddle, M. J., Lennie, T. A., Bricker, G. V., Kopec, R. E., Schwartz, S. J., & Moser, D. K. (2015). Lycopene dietary intervention: a pilot study in patients with heart failure. *The Journal of cardiovascular nursing*, *30*(3), 205.
- Bourgaud, F., Gravot, A., Milesi, S., & Gontier, E. (2001). Production of plant secondary metabolites: a historical perspective. *Plant science*, *161*(5), 839-851.
- Bruno, R. S., Wildman, R. E., & Schwartz, S. J. (2007). Lycopene: food sources, properties, and health. In *Handbook of nutraceuticals* and functional foods (pp. 55-72). CRC press.
- Chidambara Murthy, K. N., Shivapriya, M., Monika, P., & Tejashree, B. (2019). Challenges in Optimal Utilization of Bioactive Molecules Clinically. *Bioactive Molecules in Food*, 109-136.
- Clinton, S. K. (1998). Lycopene: chemistry, biology, and implications for human health and disease. Nutrition reviews, 56(2), 35-51.
- Costa-Rodrigues, J., Pinho, O., & Monteiro, P. R. R. (2018). Can lycopene be considered an effective protection against cardiovascular disease?. *Food chemistry*, 245, 1148-1153.
- Di Pietro, N., Di Tomo, P., & Pandolfi, A. (2016). Carotenoids in cardiovascular disease prevention. JSM Atheroscler, 1, 1-13.
- Dufossé, L. (2018). Microbial Pigments From Bacteria, Yeasts, Fungi, and Microalgae for the Food and Feed Industries. In *Natural and Artificial Flavoring Agents and Food Dyes* (pp. 113-132). Academic Press.
- Fraser, P. D., &Bramley, P. M. (2004). The biosynthesis and nutritional uses of carotenoids. *Progress in lipid research*, 43(3), 228-265.
- Fordham, I. M., Zimmerman, R. H., Black, B. L., Clevidence, B. M., & Wiley, E. R. (2002, August). Autumn olive: A potential alternative crop. In XXVI International Horticultural Congress: Berry Crop Breeding, Production and Utilization for a New Century 626 (pp. 429-431).
- Gerster, H. (1997). The potential role of lycopene for human health. Journal of the American College of Nutrition, 16(2), 109-126.
- Giovannucci, E. (1999). Tomatoes, tomato-based products, lycopene, and cancer: review of theepidemiologic literature. *Journal of the national cancer institute*, 91(4), 317-331.
- Giovannucci, E., Rimm, E. B., Liu, Y., Stampfer, M. J., & Willett, W. C. (2002). A prospective study of tomato products, lycopene, and prostate cancer risk. *Journal of the National Cancer Institute*, 94(5), 391-398.
- Gupta, R., Balasubramaniam, V. M., Schwartz, S. J., & Francis, D. M. (2010). Storage Stability of Lycopene in Tomato Juice Subjected to Combined Pressure– Heat Treatments. *Journal of agricultural and food chemistry*, 58(14), 8305-8313.
- Hua, Y., Xu, N., Ma, T., Liu, Y., Xu, H., & Lu, Y. (2019). Anti-Inflammatory Effect of Lycopene on Experimental Spinal Cord Ischemia Injury via Cyclooxygenase-2 Suppression. *Neuroimmunomodulation*, 1-9.
- Jiang, L. N., Liu, Y. B., & Li, B. H. (2019). Lycopene exerts anti-inflammatory effect to inhibit prostate cancer progression. *Asian journal of andrology*, 21(1), 80.

- Kirpichenkova, E. V., Korolev, A. A., Onishchenko, G. G., Nikitenko, E. I., Denisova, E. L., Fetisov, R. N., ... &Fanda, E. A. (2018). Study of consumption frequency of the main sources of lycopene and its quantification in students' diet. *IP Pavlov Russian Medical Biological Herald*, 26(4), 474-483.
- Koh, E., Charoenprasert, S., & Mitchell, A. E. (2012). Effects of industrial tomato paste processing on ascorbic acid, flavonoids and carotenoids and their stability over one-year storage. *Journal of the Science of Food and Agriculture*, 92(1), 23-28.
- Kohlmeier, L., Kark, J. D., Gomez-Gracia, E., Martin, B. C., Steck, S. E., Kardinaal, A. F., ... & Martin-Moreno, J. M. (1997). Lycopene and myocardial infarction risk in the EURAMIC Study. *American Journal of Epidemiology*, 146(8), 618-626.
- Komuroglu, A. U., Yur, F., Ekin I. H. (2018). Influence of lycopene administration on neopterin, myeloperoxidase and gamma glutamyl transferase in diabetic rats. *Fresenius Environmental Bulletin*. 27(6), 4285-4292.
- Kumari, M., Kumar, S., Yadav, A., Singh, R. S., & Roy, C. (2017). Secondary Metabolites: Evolutionary Perspective, In Vitro Production, and Technological Advances. In *Plant Secondary Metabolites, Volume Two* (pp. 27-52). Apple Academic Press.
- Lowe, G. M., Graham, D. L., & Young, A. J. (2018). Lycopene: Chemistry, Metabolism, and Bioavailability. In Lycopene and Tomatoes in Human Nutrition and Health (pp. 1-20). CRC Press.
- Mackinnon, E. S., Rao, A. V., Josse, R. G., & Rao, L. G. (2011). Supplementation with the antioxidant lycopene significantly decreases oxidative stress parameters and the bone resorption marker N-telopeptide of type I collagen in postmenopausal women. Osteoporosis International, 22(4), 1091-1101.
- Malekiyan, R., Abdanipour, A., Sohrabi, D., & Jafari Anarkooli, I. (2019). Antioxidant and neuroprotective effects of lycopene and insulin in the hippocampus of streptozotocin-induced diabetic rats. *Biomedical reports*, 10(1), 47-54.
- Maria, A. G., Graziano, R., & Nicolantonio, D. O. (2015). Carotenoids: potential allies of cardiovascular health?. Food & nutrition research, 59(1), 26762.
- Murakami, K., Honda, M., Takemura, R., Fukaya, T., Kanda, H., &Goto, M. (2018). Effect of thermal treatment and light irradiation on the stability of lycopene with high Z-isomers content. *Food chemistry*, 250, 253-258.
- Phan, M. A. T., Bucknall, M. P., & Arcot, J. (2019). Interferences of anthocyanins with the uptake of lycopene in Caco-2 cells, and their interactive effects on anti-oxidation and anti-inflammation in vitro and ex vivo. *Food chemistry*, 276, 402-409.
- Ramesh, M., &Muthuraman, A. (2018). Flavoring and coloring agents: Health risks and potential problems. In Natural and artificial flavoring agents and food dyes (pp. 1-28). Academic Press.
- Rao, A. V., & Agarwal, S. (1999). Role of lycopene as antioxidant carotenoid in the prevention of chronic diseases: a review. Nutrition research, 19(2), 305-323.
- Rao, L. G., Krishnadev, N., Banasikowska, K., & Rao, A. V. (2003). Lycopene I—effect on osteoclasts: lycopene inhibits basal and parathyroid hormone-stimulated osteoclast formation and mineral resorption mediated by reactive oxygen species in rat bone marrow cultures. *Journal of medicinal food*, 6(2), 69-78.
- Rao, A. V., & Rao, L. G. (2007). Carotenoids and human health. Pharmacological research, 55(3), 207-216.
- Rocha, C. E., Roehrs, R., & Roehrs, M. (2015). Chemistry, Sources and Benefits of Lycopene. In Food Sources, Potential Role in Human Health and Antioxidant Effects (pp. 105-113). Nova.
- Rodriguez-Amaya, D. B. (2019). Natural food pigments and colorants. Bioactive Molecules in Food, 867-901.
- Sandei, L. (2018). 9 Lycopene and Tomatoes. In Lycopene and Tomatoes in Human Nutrition and Health (pp. 149-178). CRC Press.
- Sgherri, C., Pérez-López, U., &Pinzino, C. (2015). Antioxidant properties of food products containing lycopene are increased by the presence of chlorophyll. Lycopene: Food Sources, Potential Role in Human Health and Antioxidant Effects Edited by bailey JR. New York: Nova Science Publishers, inc, 39-90.
- Sharmeen, Z., Bashir, S., Kan, A. A. (2018). Physiochemical Assay of Lycopene Supplemented Yoghurt. AJAHS. 3(2). 44-49.
- Siddiqui, M. W., Ayala-Zavala, J. F., Gonzalez-Rios, H., Gonzalez-Aguilar, G. A., Ansorena, R., & Lopez-Romero, J. C. (2017). Applications of Plant Secondary Metabolites in Food Systems. In *Plant Secondary Metabolites, Volume Two* (pp. 221-258). Apple Academic Press.
- Song, P., Li, D., Wang, X., & Zhong, X. (2019). Lycopene protects from perfluorooctanoic acid induced liver damage and uterine apoptosis in pregnant mice. Int J Clin Exp Med, 12(1), 212-219.
- Stahl, W., & Sies, H. (2003). Antioxidant activity of carotenoids. Molecular aspects of medicine, 24(6), 345-351.
- Stephen, N. M., Gayathri, R., Niranjana, R., Prasad, Y., Das, A. K., Baskaran, V., & Ganesan, P. (2017). Carotenoids: types, sources, and biosynthesis. In *Plant Secondary Metabolites, Volume Two* (pp. 103-132). Apple Academic Press.
- Takeoka, G. R., Dao, L., Flessa, S., Gillespie, D. M., Jewell, W. T., Huebner, B., ... & Ebeler, S. E. (2001). Processing effects on lycopene content and antioxidant activity of tomatoes. *Journal of Agricultural and Food Chemistry*, 49(8), 3713-3717.
- Tang, G., Ferreira, A. L. A., Grusak, M. A., Qin, J., Dolnikowski, G. G., Russell, R. M., &Krinsky, N. I. (2005). Bioavailability of synthetic and biosynthetic deuterated lycopene in humans. *The Journal of nutritional biochemistry*, 16(4), 229-235.
- Tripathi, P., Blaggana, V., Upadhyay, P., Jindal, M., Gupta, S., & Nishat, S. (2019). Antioxidant therapy (lycopene and green tea extract) in periodontal disease: A promising paradigm. *Journal of Indian Society of Periodontology*, 23(1), 25.

- US Department of Agriculture (USDA), Agricultural Research Service, Nutrient Data Laboratory. USDA National Nutrient Database for Standard Reference. Available at: <u>http://www.ars.usda.gov/nutrientdata</u>. Accessed:19 Feb 2019.
- Verpoorte, R., & Memelink, J. (2002). Engineering secondary metabolite production in plants. *Current opinion in biotechnology*, 13(2), 181-187.
- Xianquan, S., Shi, J., Kakuda, Y., &Yueming, J. (2005). Stability of lycopene during food processing and storage. *Journal of medicinal food*, 8(4), 413-422.
- XIE, B. X., WEI, J. J., ZHANG, Y. T., SONG, S. W., SU, W., SUN, G. W., ... & LIU, H. C. (2019). Supplemental blue and red light promote lycopene synthesis in tomato fruits. *Journal of Integrative Agriculture*, *18*(3), 590-598.
- Yilmaz, S., Kaya, E., Karaca, A., &Karatas, O. (2018). Aflatoxin B1 induced renal and cardiac damage in rats: Protective effect of lycopene. *Research in veterinary science*, *119*, 268-275.
- Yin, Y., Zheng, Z., & Jiang, Z. (2019). Effects of lycopene on metabolism of glycolipid in type 2 diabetic rats. *Biomedicine & Pharmacotherapy*, 109, 2070-2077.
- Yonekura, L., & Nagao, A. (2007). Intestinal absorption of dietary carotenoids. Molecular nutrition & food research, 51(1), 107-115.



# Synthesis and Separation of the Spiro[oxindole-cyclopropane] Diasteriomers with Anti-HIV-1 or Anti-HIV-2 Activity

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**Abstract:** The main objective of the current study was design and synthesis of a new group of compounds with anti-HIV-1 and anti-HIV-2 activities. The introduction of new asymmetric centers into the molecule made possible diastereselective separation of enantiomers. Such a technique can significantly improve the biological properties of the obtained substances and thus improve the outcomes of the antiretroviral therapy.

Keywords: organic chemistry, organic synthesis, spiro[cyclopropane-oxindole], diasteroselective synthesis

# **1. INTRODUCTION**

The unique structural features of spirooxindoles together with diverse biological activities have made them privileged structures in new drug discovery. According to a large number of publications, such spiro compounds have different biological properties (Pavlovska, 2015). Among them are substances with anti-HIV activity (Jiang, 2006, Jiang, 2006a). It is known that some of spirooxindole derivatives have a high ability to inhibit HIV reverse transcriptase, which is responsible for the reproduction of the virus. This ability is manifested at concentrations close to the concentrations of active drugs (EC50 = 50 nM).

Currently there is no effective vaccine, and the existing medical treatment of HIV infections does not ensure a definitive cure from the virus. Moreover, medicines currently used to treat HIV infection can induce the development of additional side effects over time. Therefore, the development of new antiviral drugs that will be more active and less toxic than existing ones is of major importance. It is important to develop antiviral drugs that will be more active and less toxic than existing ones.

We have previously shown that spirooxindole derivatives have significant activity against another retroviral enzyme - integrase (Surmava, 2010). However, it should be mentioned that the biological properties of these derivatives were tested with application of racemic mixtures and not individual enantiomers. Taking into account the fact that often only one of the enantiomers is active, it can be assumed that the use of enantiomerically pure derivatives will increase the activity by half, which will reduce the overall dosage and, consequently, diminish the toxicity.

# 2. MATERIALS AND METHODS

Research methodology includes fine organic synthesis methods and classical analysis methods (HPLC, NMR, IR, elemental analysis, etc.). Antiviral and cytotoxic testing was performed on the MT-4 cell culture infected with HIV-1 (IIIB) and HIV-2 (ROD) viruses.

All used solvents were of reagent quality, and all commercial reagents were used without additional purification. Removal of all solvents was carried out under reduced pressure. Analytical TLC plates were Silufol® UV-254 (Silpearl on aluminium foil). IR spectra were recorded on a Spectrum 100 FT-IR spectrophotometer (Perkin–Elmer) using the universal ATR sampling accessory. 1H and 13C NMR spectra have been recorded for CDCl<sub>3</sub> 2-% solution on a "Bruker -Avance III" (400.13 and 100.61 MHz). Melting points were determined on a Boëtius melting point apparatus (PHMK, VEB Wägetechnik Rapido, Radebeul, Germany).

Isatines **1a,b** were prepared using the methods [Sucman, 2012, Jiang, 2006]. Hydrolysis of the esters has been carried out with formation of individual *cis*- and *trans*-acids **2** (Figure 1) according to the method (Sucman, 2018).



Figure 1. Starting materials

#### 3. RESULTS AND DISCUSSION

Earlier, our group attempted an enantioselective synthesis of spiro[cyclopropane-oxindoles] [Noole,2012]. However, it was not possible to achieve significant results on the required derivatives. High values of enantioselectivity (more than 95%ee) were obtained with a certain change in the structure of the final substance, but this led to a complete loss of the biological properties of the molecule. This fact can be explained by the fact that the surface and/or the size of the new molecule do not correspond to the size of the active center of enzymes.

For the purpose of the obtaining of individual stereoisomers and increasing the effectiveness of the hit compounds, they were derivatized by introducing certain asymmetric groups in their structure and the formed diastereomers were separated.

There are several reaction centers in the structure of spiro[cyclopropane-oxindoles], which we tried to use in the obtaining of diastereomeric derivatives. First of all, various amides **3**, **4** and **5**, **6** were obtained from acids **2a**, **b** (Fig.2).



Figure 2. Synthesis of diastereomeric amides 3-6

It should be noted that the reactions were carried out in mild conditions of peptide synthesis [Nhut, 2015], because of epimirization of one of the chiral centers in cyclopropane moiety in case of usual synthetic way, throw acid chlorides. In the first case, the diastereomers were separated by chromatography. In the second entry chromatographic separation was impossible, but it was possible to recrystallize them, with separation of individual compounds. Use of leucine and valine as chiral reagents lead to the diastereomers, which could not be separated with application of the previously described methods.

The next step was the introduction of the chiral fragment in the molecule due to substitution of the hydrogen of oxindole amide. The syntheses were carried out by alkylation according to the schemes presented in Figure 3.



Figure 3. Synthesis of diastereomers 7-10 by alkylation

From the mixture of **6** and **7**, one of the diastereomers was isolated in individual form by recrystallization. The mixture of **9** and **10** appeared as chromatographically indivisible oil.

Compounds 13 and 14 have been synthesized to increase the number of target compounds. The amino derivative 11 was obtained from nitro compound 1c by reduction with hydrogen on Pd/C. Further, after condensation with dehydroabietic chloride 12 (Fig.4), a mixture of a new type of amides 13,14 was obtained that was then separated by column chromatography.



Figure 4. Synthesis of compounds 12,13

The structures of all separated compounds have been confirmed with application of different physico-chemical methods of analysis, including HPLC, NMR, IR, elemental analysis, etc.

# 4. CONCLUSION

A number of compounds with potentially high antiviral activity were obtained as a result of different synthetic pathways with application of chiral reagents followed by separation of diastereomers.

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#### REFERENCES

- Jiang, T., Kuhen, K.L., Wolff, K., Yin, H., Bieza, K., Caldwell, J., Bursulaya, B., Wub, T. Y.-H., and He, Y.,2006. Design, synthesis and biological evaluations of novel oxindoles as HIV-1 non-nucleoside reverse transcriptase inhibitors. Part IBioorganic & Medicinal Chemistry Letters 16: 2105–2108
- Jiang, T., Kuhen, K.L., Wolff, K., Yin, H., Bieza, K., Caldwell, J., Bursulaya, B., Tuntland, T., Zhang, K., Karanewsky, D., and He, Y. 2006. Design, synthesis and biological evaluations of novel oxindoles as HIV-1 non-nucleoside reverse transcriptase inhibitors. Part II. Bioorganic & Medicinal Chemistry Letters 16: 2109–2112;
- 3. Surmava, S., Elefthetiou, P., Geronikaki, A., Petrou, C., Macaev, F., and Sucman, N. HIV-1 integrase inhibition by novel spiroisatin-ciclopropane derivatives. In: XVIII International AIDS Conference. Viena: Austria, 2010, p. 56
- 4. MD 2012/4202
- Noole, A., Sucman, N. S., Kabeshov, M. A., Kanger, T., Macaev, F. Z., Malkov, A.V. Highly enantio- and diastereoselective generation of two quaternary centers in spirocyclopropanation of oxindole derivatives. 2012. Chemistry: A European Journal 18 (47): 14929-14933.
- 6. US 2015/0329593 A1,
- Pavlovska, T. L., Redkin, R. G., Lipson, V. V., and Atamanuk, D. V. 2015. Molecular diversity of spirooxindoles. Synthesis and biological activity. Molecular Diversity 20(1): 299–344.
- Sucman, N., Boldscu, V., Kravtsov, V.CH., Baca, S.G., Macaev, F.Z. The structure of 5'-bromo-2'-oxo-1',2'-dihydrospiro-[cyclopropane-1,3'-indole]-2-carboxylic acid. In: 9 th International conference on materials science and condensed matter physics. Moldova. 25-28 september, 2018. p. 166.



# Cutting Production of *Hedera helix* L., *Tamarix gallica* L. and *Chaenomeles japonica* (Thunb.) Lindl.) Used in Landscape Applications

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Abstract: Due to the rapidly growing world population and developing technology, the big city life has affected the physical and psychological health of humanity. In this sense, plants have a positive effect on human health in all respects due to their relaxing effects and the benefits they provide in air pollution with the abundant oxygen they bring to the environment. Therefore, the selection of plant species to be used in landscaping works especially in big cities is very important. These selected plants should not be harmed by negative external factors, and they should make a significant contribution to avoiding heavy metal-borne environmental pollution by living fast and growing for a long time. On the other hand, the selected plants in park and garden landscaping arrangements should not have toxic and toxic effects. In this study, in this study, the production of the forests, vine and Japanese lunar species, which have a live leaf mass for a long period of time and which have a rapid psychological development by closing the concrete surfaces, and which have been used to produce a relaxing psychological effect, have been examined. For this purpose, the rooting medium of the steels is composed of 50% sand + 50% perlite. The first one of these rooting media was applied to the 1% IBA (Indole -3-Butyric Acid) solution, and the second one was applied to the 2% IBA solution. One-way ANOVA was applied to determine whether there was any difference between species and rooting environments and Duncan Test was applied for grouping in case of difference. All statistical analyzes were performed in SPSS package statistics program. As a result of the examination and root counts conducted one year after the establishment of the experiment, a statistically significant difference was found between the hormone applications and P <0.01 confidence level, especially after the counts made in the roots longer than 5 cm. In this context, according to the Duncan test applied at the confidence level P <0.05, 1% and 2% IBA solution were included in the first group and control parcel was in the second group. When evaluated in terms of species, there was also statistical difference at P <0.01 confidence level according to ANOVA. As a result of the Duncan test performed at the P < 0.05 confidence level in this context, the forest group was in the first group with 90.7% and 90.3% rooting percentages, while the Japanese group was in the second group with 75.6% rooting percentage.

Keywords: Cutting, Rooting, IBA, Landscape, Ornamental Plant.

### **1. INTRODUCTION**

Urban life, which grows every day and shows a complex structure, adversely affects the psychological and biological health of the society with increasing population and industrialization activities. For this reason, society needs functional and functional green areas, relaxing and sustainable architecture. In this context, the living biomass, which enables the quality living conditions required by the societies, should be established both in the city centers and around the cities. The fact that green areas are afforestation and urban forests, especially the areas that improve the quality of life by cleaning the air, are very important in terms of eliminating many negativities and getting people to have a happier life. As a matter of fact, the hot and dry islets caused by reflections due to the angle and duration of the sun's rays in high build cities have negative effects on the life cycle of all living things and also negatively impacts the effects of global climate change (KONIJNENDIJK AND RANDRUP, 2004). For this reason, the use of species that are rapidly growing and spreading very rapidly to concrete and iron surfaces in the context of green architectural works provides significant benefits. (RAGHEB ET AL., 2016). For this purpose, the production of high quality and healthy seedlings of these species is also very important for their use (ÜRGENÇ, 1998). At the beginning of these species are the European ivy (*Hedera helix* L.), common tamarisk (*Tamarix gallica* L.) and the Japanese quince (*Chaenomeles japonica* (Thunb.) Lindl.).

In this study, the production of the European ivy, common tamarisk and Japanese quince species, which have a lively leaf mass for a long time and which have a rapid psychological effect and which has a relaxing psychological effect by closing the concrete surfaces, has been studied.

#### 2. MATERIAL AND METHODS

European ivy, which is one of the species working on the material as a material, is a species with high climber and spreading properties which can be seen in the form of many pure and mixed forests in Euro-Siberian plant region ( $\ddot{U}RGENÇ$ , 1998; GENÇ, 2012). Therefore, it is frequently preferred in landscaping. In this research, Devrek origin of the European ivy is used. Another type is the infamous; It is a common species in salty, semi-arid and poor coniferous soils. It has a decorative appearance due to its highly flowering branches and is widely used in landscaping (GENÇ, 2012). In this study, branch steels obtained from common tamarisk individuals of Konya origin were used. The last species used in the rooting studies in the research is the Japanese lunar. This species is often branched, and there are spikes on the shoots. Oval shaped, glossy dark green leaves ( $\ddot{U}RGENÇ$ , 1998). Rooting studies were carried out in Gökçebey Forest Nursery production greenhouses, daytime ambient temperature in greenhouses was 21.3 ° C, night temperature was 13.6 ° C, rooting medium; % 50kum +% 50perlit, rooting environment temperature 18.2 ° C, rooting environment humidity 82.6% and rooting environment 68.4% (Figure 1).



Figure 1. Rooting Conditions of Cuttings

Cuttings used in the rooting studies were taken in the last week of April 2018. Cuttings were selected and healthy branches were selected and the steel was 12 cm long and 0.7 cm thick. (Figure 2).



Figure 2. Cutting Samples By Used in Research

Cuttings taken from healthy branches of the midlet were brought into the laboratory and kept in rooting medium under greenhouse conditions after hiding in a cool ( $4 \circ C$ ) environment for 2 weeks (Figure 3).



Figure 3. Transfer of Cuttings to Rooting Conditions.

In order to increase the rooting success of the cuttings used in the three species used in the study before cuttings were taken into rooting medium, some pre-treatments shown in Table 1 were applied. These pretreatments were performed in a replica of 10 cuttings and 4 replicates (Figure 4).

| Table 1. Pretreatments Applications to Cutt | ings |
|---|------|
|---|------|

| Pretreatment       | Hedera helix L     | Tamarix gallica L  | Chaenomeles japonica |
|--------------------|--------------------|--------------------|----------------------|
|                    |                    |                    | (Thunb.) Lindl.      |
| 1% IBA (Indole -3- | 40 pieces cuttings | 40 pieces cuttings | 40 pieces cuttings   |
| Butyric Acid)      |                    |                    |                      |
| 2% IBA (Indole -3- | 40 pieces cuttings | 40 pieces cuttings | 40 pieces cuttings   |
| Butyric Acid)      |                    |                    |                      |
| Control            | 40 pieces cuttings | 40 pieces cuttings | 40 pieces cuttings   |

In order to determine the differences between the pretreatment and control samples applied to the 3 different branches, Duncan test was used for variance analysis (ANOVA) and groupings. SPSS package statistical program was used to perform these analyzes.



Figure 4. Roots of Three Species Cuttings

In the study, it was found that the first primary and secondary roots were removed in determining whether the cuttings were rooted (Figure 5).



Figure 5. Primary and Secondary Roots in Cuttings Accepted as Rooted

# 3. RESULTS AND DISCUSSION

#### Results

As a result of the examination and root counts conducted one year after the establishment of the experiment, a statistically significant difference was found between the hormone applications and P <0.01 confidence level, especially after the counts made in the roots longer than 5 cm. In this context, according to the Duncan test applied at P <0.05 confidence level, 1% and 2% IBA solution were included in the first group and control parcel was in the second group. When evaluated in terms of species, there was also statistical difference at P <0.01 confidence level according to ANOVA. As a result of the Duncan test performed at the P <0.05 confidence level in this context, the forest group was in the first group with 90% and 90.3% rooting percentages, while the Japanese group was in the second group with a percentage of 75.6% rooting (Table 2).

**Table 2.** Variance Analysis of Rooted Steels by Pretreatment and Species and Duncan Test Results.

|                    | Percentage of Rooting (%) |                            |                   |  |  |  |  |  |
|--------------------|---------------------------|----------------------------|-------------------|--|--|--|--|--|
|                    | F=146,84**                |                            |                   |  |  |  |  |  |
| Pretreatment       |                           |                            | Chaenomeles       |  |  |  |  |  |
|                    | Hedera helix L            | Tamarix gallica L<br>90,3a | japonica (Thunb.) |  |  |  |  |  |
|                    |                           |                            | Lindl.            |  |  |  |  |  |
| 1% IBA (Indole -3- | 00.60                     | 00.3a                      | 75.2%             |  |  |  |  |  |
| Butyric Acid)      | 90,0a                     | 90,3a                      | 75,2a             |  |  |  |  |  |
| 2% IBA (Indole -3- | 01.85                     | <u>90 5a</u>               | 77 59             |  |  |  |  |  |
| Butyric Acid)      | 91,0a                     | 90,5a                      | 11,5a             |  |  |  |  |  |
| Control            | 52,3b                     | 58,6b                      | 38,3b             |  |  |  |  |  |

a, b and c: Different letters indicate different groups. \*\*: P<0.01

#### Discussion

Research has been studied in urban landscaping and green architecture studies and the effects of some pretreatments on the percentage of rooting of the three species of green species, which cover the bare surfaces in a short period of time with rapid growth characteristics were investigated. *Hedera helix L.*, which is the first of these species, has an important value both in its natural field and its landscaping works with its spreading and wrapping properties. As a matter of fact, 1% and 2% IBA applications applied to the cuttings of the species significantly affected the rooting percentage compared to the control samples and increased them significantly (Table 1). In another study, it was found that 2% IBA solution had a significant effect on rooting of forest ivy steels with 89.6% rooting success (GENEVE, 1990). In the study, Tamarix gallica L. is another species in which the effects of pre-treatment on the success of rooting cuttings are examined. Accordingly, the 1% and 2% IBA solution applied to Tamarix gallica L. steels increased the rooting success of the steels

to the control samples at a very high level (Table 1). In another study conducted in this type, it was found that the steels of the type of pre-treatment had positive effects on rooting success (DRABU ET AL., 2012). In the scope of the research, some of the pre-treatments on the success of the cuttings of the cuttings are examined by *Chaenomeles japonica* (Thunb.) Lindl. As a result of the pretreatment, it was determined that the success of rooting in the steel of the species was lower than the other two species, but higher than the control samples (Table 1). In another study conducted for the vegetative production of this species, it is emphasized that especially the rooting and production environments created with the plant growth regulators increase the vegetative production capability of the species and that the success of rooting can be reduced to 70-80% in this way (PANAVAS, 1994). According to these comparative results, it is possible to say that the percentage of rooting was significantly increased in all three types of pretreatments in order to increase the rooting success of the cuttings.

#### 4. CONCLUSION

It is possible to make the following suggestions in the light of the findings of the research;

• European ivy, common tamarisk and Japanese quince, which is widely used in landscape applications, can be easily rooted by the effects of IBA hormone applied in 1% and 2% doses applied to the branch cuttings. According to the results of the study, it is sufficient to use 1% IBA because of Duncan Range Test.

• Due to their rapid growth and convincing properties, these species may also have the potential to be used for preliminary expansions and permanent stabilization studies in dune and semi-arid zones afforestation studies, and therefore this situation should be investigated.

• It is important that these species are taken from branches that are not exposed to high levels of sunlight when receiving branch cuttings.

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#### REFERENCES

- Drabu, S., Chaturverdi, S., Sharma, M. (2012). Tamarix gallica-An Overview, Asian Journal of Pharmaceutical and Clinical Research 5:124-131.
- Genç, M. (2012). Süs Bitkisi Yetiştiriciliği [Production of Ornamental Plants], Süleyman Demirel Üniversitesi, Orman Fakültesi, Yayın No:55, Isparta.
- Geneve, R.L. (1990). Root Formation in Cuttings of English Ivy Treated with Paclobutrazol or Uniconazole, HortScience 25(6): 709-714.
- Konijnendijk, C.C., Randrup, T.B. (2004). Urban Forestry, Danish Forest and Landscape Research Institute, Hoersholm, Denmark.
- Panavas T. (1994). Optimization of the growth medium for the micropropagation of Japanese quince (*Chaenomeles japonica* Thunb.). Biologija 3: 44–49.
- Ragheb, A., El-Shimy, H., Ragheb, G. (2016). Green Architecture: A Concept of Sustainability, Procedia-Social and Behavioral Sciences 216(2016): 778-787.
- Ürgenç, S. (1998). Ağaç ve Süs Bitkileri Fidanlık ve Yetiştirme Tekniği [Nursey and Cultivation Techniques of Trees and Ornamental Plants]. İ.Ü Orman Fakültesi Yayın No: 3676/418, İstanbul.



# Accuracy Of UAV-Derived Digital Terrain Model (DTM) And Its Effect On Canopy Height Model (CHM) In Berkelah Tropical Rainforest, Malaysia

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Abstract: Tropical forest plays a crucial role in the storage of a large amount of carbon, typically the aboveground biomass of trees which is affected by deforestation and degradation. UNFCCC intended to reduce carbon dioxide(CO2) emission from land use change and deforestation through (REDD+) program and its MRV Mechanism. There is a need in tropical countries for low cost, accurate and timely information to determine tree parameters like height and diameter. Direct measurement of tree height with measuring tape is the most accurate, but it is not a viable option for forest monitoring and evaluation. Light Detection And Ranging (LiDAR) is one of the remote sensing technology used to estimate tree height with the best result in terms of accuracy. However, the cost of LiDAR can pose financial constraints, especially when the study area needs temporal data to monitor vegetation change. 3D photogrammetry using Unmanned Aerial Vehicle (UAV) images is a potentially cost-effective alternative method. The quality of a UAV-DTM directly influences the estimation of tree height and AGB and carbon stock. The quality of the UAV-DTM in its turn is influenced by the number and configuration of the Ground Control Points (GCPs). Therefore, this thesis present accuracy assessment of UAV DTM with different number and layout of GCPs and its effect on CHM. The accuracy of UAV DTM with 4,6,8 and ten ground control point, when compared to checkpoints which measured by DGPS, achieved RMSE of  $\pm 0.9$  m and  $R^2$  of 0.98 for all DTM'S While  $R^2$  of 0.9 and RMSE of ±1.5 m ALS. On the other hand, The accuracy of UAV DTM with 4,6,8 and ten ground control point, when compared to ALS DTM, achieved RMSE of  $\pm 3.6$  m,  $\pm 3.53$  m,  $\pm 3.51$ m, and ±3.50 m and R2 of 0.66,0.68,0.67 and 0.67 respectively. The accuracy assessment of UAV tree height in comparison to ALS tree height revealed that RMSE of  $\pm 2.18$  m and R<sup>2</sup> of 0.6. Additionally, the comparison was made between UAV tree height and ALS tree height in relatively closer altitude of DTM height. The accuracy assessment revealed that  $R^2$  of 0.88 and RMSE of ±2 m. Furthermore, the AGB was computed using an allometric equation which utilized Diameter at Breast height (DBH), tree height and wood density. AGB and carbon stock computed using the adjusted tree height. The result revealed that mean biomass of 0.04Mg, 0.05Mg, 0.06Mg and 0.06 for RMSE -2,-1,0,1 and 2 respectively.

Keywords: UAV, Digital terrain model, Tropical rainforest, Canopy Height Model, Remote Sensing



# The Impact of Aero Technogenic Emissions of Magnesite Production on Experimental Forest Cultures and Soils, on The Example of the Combine Magnesite South Ural, Russia

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Abstract: This study was aimed at studying the chemistry of the soil and condition of the experimental forest cultures of pine (Pínus sylvéstris L.), Sukachev larch (Larix sukaczewii D y l.) and silver birch (Betula pendula Roth.) on the background of a significant decrease in aero-emissions of Combine Magnesite. Forest cultures were planted in rows in 1980-1983 by the Ural Forest Experiment Station (All-Russian Research Institute of Forestry and Forestry Mechanization) for the purpose to study the suitability of soil for reforestation in various zones of magnesite pollution. When laying the test sites, peat was applied with a layer of 12 cm, a peat layer of 2 cm, nitrogen-phosphorus-potassium fertilizers, a weak solution of sulfuric acid (to reduce the pH of the soil). The entry of maximum volumes of magnesite dust into the atmosphere in 1963 reached 182.5-328.5 thousand t/day. In 1978, new electrostatic precipitators were installed at the plant, and dust emissions decreased to 70-90 t/day. The level of snow pollution for the period from 1983 to 2019 showed a consistently high alkaline pH of snow water of 9.1-10.4 units in the zone of strong dust (1 km from the Combine). Analysis of the content of exchangeable cations in the soil (Mg<sup>2+</sup> and Ca<sup>2+</sup>) showed a significant increase in exchangeable magnesium, which can cause salt formation in the soil and adversely affect the plants. An increase in the content of exchangeable Mg cations with respect to Ca cations up the soil profile, as well as a comparison of this indicator with background indicators, indicates the technogenic character of this process. In the background natural soils of exchangeable Ca<sup>2+</sup> is several times more than Mg<sup>2+</sup>. The results of chemical analysis of soil samples showed significant changes in soil chemistry at the source of pollution, which is an increase in pH by 2-3 units, accumulation of exchangeable magnesium and some heavy metals in the soil (iron, zinc, cobalt, copper dominate). All these changes negatively affect the growth and living conditions of plants, and in the impact zone they lead to the death of forest-forming species. The level of soil pollution characterizes the accumulated effect of aero technogenic pollution, since pollutants have been sorbed into the soil absorbing complex for over 50 years. Peat layer 12 cm as an improver increased the growth of birch compared with the option without peat in diameter 2.8 times (t = 9.23 with p < 0.001), and in height 2.1 times (t = 10.2with p < 0,001). Analyzing birch and pine under similar conditions (peat layer 2 cm), pine height is 20% more (t = 2.46 with p = 0.02), and diameter is 70% more (t = 6.34 with p<0.001) than birch. The surviving specimens of larch in the variant with acid showed the best growth compared with birch in the same version: diameter is more than 88% (t = 4.75at p <0.001), and height is 23% (t = 1.89 at p = 0.06). In comparison with ordinary pine (a variant with peat of 2 cm), the diameter of larch is 26% less (t = 2.40 with p = 0.02). It can be assumed that in the zone of heavy pollution, coniferous species in the remaining variants show better growth than birch. As a result of our research, it was found that the growth and condition of larch under conditions of contaminated magnesite production is better than that of birch and pine. The positive effect of lowland peat introduced during planting as an ameliorant, which reduced the negative impact of aero technogenic emissions of the Combine, was revealed. Accordingly, in a zone of heavy pollution, it is possible to create forest crops using peat as an ameliorant in sufficient quantities (for example, lowland peat not less than 12 cm thick). Creating cultures in areas of medium and low pollution is possible from any species studied, without adding peat.

Keywords: aero-technogenic emissions, experimental forest cultures, reforestation, snow and soil pollution, lowland peat.



# Modelling of Above Ground Biomass (AGB) of Mangroves in Kedah, Malaysia using Landsat 8 (OLI) Imagery

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Abstract: Mangroves are recognized as ecosystem that grow and dominate the coastal areas of tropical and sub-tropical regions across the world. The high adaptability properties of these halophytic trees enable them to thrive in a harsh condition such as the intertidal zones. They not only provide ecological and socio-economic support, but also play pivotal role in offsetting an excess of carbon from the atmosphere. Despite the crucial roles provided by mangroves, the ecosystem has degraded at an alarming rate mainly due to anthropogenic activities. Remote sensing technology provides a new dimensional perspective in monitoring and estimating of tree biomass. Therefore, this study aimed at (1) estimating the AGB of mangroves in Kedah, Malaysia, (2) investigating the relationships between mangrove stand parameters with spectral reflectance recorded from Landsat 8 Operational Land Imager (OLI) data, and (3) developing predictive models for estimating the AGB of mangroves by combining the ground and Landsat 8 (OLI) data. For the purpose of this study, a total of 81 mangroves stand data set measuring at 100 m × 100 m were collected throughout Kedah, Malaysia. Within the stand, seven randomly selected plots were established and all individual mangroves parameter (diameter at breast height (DBH) and height) were measured, and tree species were identified at species level. From 81 stands, the data was split into two independent data sets for developing and validating the models (56 and 25 stands, respectively). Multiple regression technique with least square approach was used in the model development process. From several good candidate models, a model consists of four predictive variables (bands 3 and 6, NDVI and ratio 2) seems to be the best model due to its simplicity and predictive ability ( $p \le 0.001$ ,  $R^2 = 0.56$ , adj.  $R^2 = 0.53$ ). Validation of the model has resulted in Mallow's prediction criterion ( $C_p$ ) value of 4.28 and Root Mean Squared Error (RMSE) of 4.11 m<sup>3</sup>/ha. The information from this study may provide useful input for future research and can be crucial tools for the government and stakeholders in future decision making for the sustainability of mangrove resources.

Keywords: Modelling, Above Ground Biomass, Mangroves, Remote Sensing, Landsat 8 (OLI)



# **Evaluation of Forest Fire Watch-Towers Location (Çankırı Sample)**

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**Abstract:** Due to the increase in the demand for wood raw materials, it is of great importance that the forest resources, which are limited due to increase, are effectively managed, used and maintained according to the sustainable forest management approach. Forest fires pose a threat to both nature and people. The most important stage of the fight against forest fires is to be informed at the first moment of the fire and to direct this information and fire extinguishing teams urgently. At this stage, forest fire watch-towers serve as an early warning system. The intervention of forest fires is the most important factor in increasing the success of forest fires. For this reason, it is expected that the vision of the forest fire watch-towers (FFW) will be spread throughout the area of responsibility. Geographical Information Systems (GIS) can be used for the determination of FFW locations or evaluation of existing FFW locations. The aim of this study is to evaluate the FFW locations in Çankırı Forest Management Directorate and to put forward the FFW requirement levels in a practical way. As a result of the study, it was found that 124,261.8 ha (27%) of the total area of responsibility area of 458,888.8 ha is forest area and the visible forest area from the existing FFW locations is 33,094.5 ha and the invisible forest area is 91,167.3 ha. According to the obtained results, it was determined that FFW locations did not see all of the forest areas and the visible area was not acceptable (26.6%). It is considered that it would be beneficial to use GIS location detection for FFW locations to the maximum extent since it is important to make maximum use of GIS and that such analyzes provide advantages in decision making.

Keywords: Viewshed analysis, weighted overlay, forest watch tower, mapping

#### **1. INTRODUCTION**

Forests are limited natural resources that occur spontaneously or in labour over a long period of time. Since the demand for wood raw materials is increasing day by day, it is of great importance to protecting these resources within the framework of effective use, management and sustainability forest management understanding (DPT, 2018). There are many negative factors that threaten the existence of forests (Doğan and Tüzer, 2011). Among these factors, forest fires are one of the most important (Küçükosmanoğlu, 1985). Forest fires pose much threat to both nature and people (Baş, 2014). The most important stage of the fight against forest fires is to be informed at the first moment of the fire and to direct the fire-fighting teams to the scene (Bilici, 2009). At this stage, forest fire watch towers (FFW) serve as an early warning system (Akay et al., 2011). Forest fires are easier to intervene than they are when they start growing, and it is the most important factor that increases success in the struggle. For this reason, it is expected that the forest fire watch towers will dominate the entire field of vision. It is possible to meet this expectation in a practical and fast way with today's technologies and software. Geographical Information Systems (GIS) can be used to determine the positions of the FFWs effectively.

In this study, it has been tried to reveal the FFW activity based on the existing FFW locations within the scope of Çankırı Forest Management Directorate (FMD), which is chosen as the study area. Afterwards, a solution has been sought to help decision determine where a new FFW location can be for better surveillance.

#### 2. MATERIALS AND METHODS

The study area is the forests located within the responsibility of Çankırı Forest Management Directorate under the Ankara Regional Directorate of Forestry, and three FFWs observing these forests (Figure 1). The average elevation of the study area is 1046 m, minimum elevation is 540 m and maximum elevation is 1980 m. The average slope is 17% and the main aspect is South-West.



Figure 1. Location of the study area

This study was carried out with a two-stage methodology. In the first stage, the ratio and the amount of the forest areas within these areas were determined and not seen from the existing FFW locations. This was done using the Viewshed Analysis tool included in the ArcGIS 10.3 TM software (Figure 2).



Figure 2. Viewshed analysis (ESRI, 2019a)

In the second stage, the weighted registration tool used in ArcGIS 10.3 TM software was used to create a decision support system to further improve this current situation (Figure 3). This approach has the ability to evaluate and map various criteria together. In this study, the weighting of the criteria was taken as equal and produced as both raster and vector data as a single map output.



Figure 3. Weighted overlay analysis (ESRI, 2019b)

In order to determine the criteria, commonly used criteria have been determined by using both national and international publications and General Directorate of Forestry No: 285. These criteria are information about the distance between forests and non-forest areas, altitude and distance of each of them. This information was transferred to the GIS environment for analysis (Figure 4).



Figure 4. Factors: a. Forested areas, b. Elevation, c. FFW distances

# 3. RESULTS AND DISCUSSION

#### Results

Çankırı OİM has a total area of 458,888.8 hectares with 124,261.8 ha (27.1%) forested area and 334,627.0 ha (72.9%) non-forest. After the visibility analysis, the observed area was 106,691.9 ha (23.3%) and the invisible area was 352,196.9 ha (76.7%) (Figure 5).



Figure 5. Viewshed analysis results

Areas with visible forest areas are 33,094.5 ha (26.6%). In the area of responsibility, forests and invisible areas were found to be 91167.3 ha (73.4%). What can be done next is of great importance for decision makers and practitioners. In this study, the need for new locations or locations can be determined at three different levels of need if the need for future years is seen. Here, the map is obtained according to the low, medium and high need levels (Figure 6).



Figure 6. FFW need levels

According to the criteria discussed in this study; The red areas in Figure 6 indicate the priority and high level of need, the yellow areas indicate the medium level of neediness and the areas with green represent the low need level.

#### Discussion

According to the results, it was determined that FFW locations could not see all of the forest areas and the visible area was not acceptable (26.6%). It is very important that primarily be utilized to the maximum extent from the GIS to analyze and identify the current location of the required FFW area in Turkey. While the analysis of the methodology in this study will vary according to the criteria to be covered in this context, it is thought that it will provide an advantageous decision-making platform in the selection of FFW locations. FFW is currently being carried out within the framework of the notification no. 285 (Application Principles for the Prevention and Extinction of Forest Fires). Several meetings were held in recent years regarding the updating of this notification, but no improvement was made. In today's world, where GIS technology and usage is rapidly spreading, the most suitable areas for the location of YGK location can be accurately identified with a few hours of work. The use of this technology is critically important.

#### 4. CONCLUSION

According to the results of the FFW analysis, they were determined as not acceptable and suitable for the fight against forest fire. First of all, these deficiencies and weaknesses need to be eliminated. Afterwards, to evaluate the effectiveness of the FFW units in the next studies and to evaluate with new methods and approaches, FFW will increase diversity in its efficiency and it is thought that this issue will be enriched by considering the different aspects.

#### REFERENCES

Akay, A. E., Sivrikaya, F., Yenilmez, N., and Taylan, H. (2011). Yangın gözetleme kulelerinin lokasyonlarının CBS ortamında görünürlük analizi ile değerlendirilmesi. Ulusal Akdeniz Orman ve Çevre Sempozyumu, 24-26.

Anonymous, 1995. 285 Sayılı Tebliğ: Orman Yangınlarının Önlenmesi ve Söndürülmesinde Uygulama Esasları, Ankara.

- Baş, R. 2014. Türkiye'de orman yangınları nedenleri, zararları ve yangınlara karşı alınacak önlemler. İstanbul Üniversitesi Orman Fakültesi Dergisi, 27(2), 52-73.
- Bilici, E. 2009. Orman Yangın Emniyet Yolları ve Şeritleri İle Orman Yol Şebekelerinin Entegrasyonu, Planlamaları ve Uygulamaları Üzerine Bir Araştırma (Gelibolu Milli Parkı Örneği). İstanbul Üniversitesi Orman Fakültesi Dergisi, 59(2), 85-101.

Doğan, S., and Tüzer, M. 2011. Küresel iklim değişikliği ve potansiyel etkileri. CÜ İktisadi ve İdari Bilimler Dergisi, 12(1), 21-34.

DPT, 2018. Onuncu Beş Yıllık Kalkınma Planı (2014-2018), Retrieved in March, 4, 2019 from http://www.dpt.gov.tr

- ESRI, 2019a. Viewshed Analysis, Retrieved in March, 4, 2019 from https://learn.arcgis.com/en/projects/i-can-see-for-miles-and-miles/lessons/perform-a-viewshed-analysis.htm
- ESRI, 2019b. Weighted Overlay Analysis, Retrieved in March, 4, 2019 from http://desktop.arcgis.com/en/arcmap/10.3/tools/spatial-analyst-toolbox/how-weighted-overlay-works.htm

Küçükosmanoğlu, A. 1985. Orman yangınları. İstanbul Üniversitesi Orman Fakültesi Dergisi, 116-124.



# Determination of Antimicrobial Potential and Chemical Contents Of *Echinacea purpurea* (L.) Moench And *Erica arborea* L. Extracts

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**Abstract:** *Echinacea purpurea* (L.) Moench (Purple coneflower/Ekinezya) and *Erica arborea* L. (Tree heath/Funda) plants were extracted by using 65% ethanol to isolated their active constituents. The antimicrobial activities of extracts were investigated against 19 microorganisms (*Enterobacter aerogenes* ATCC 13048, *Salmonella infantis, Listeria monocytogenes* ATCC 7644, *Klebsiella pneumoniae, Pseudomonas aeruginosa* DSMZ 50071, *Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis* ATCC 29212, *Listeria innocua, Salmonella enteritidis* ATCC 13075, *Enterococcus durans, Salmonella typhimurium* SL 1344, *Candida ablicans* ATCC 10231, *Enterococcus faecium* ATCC 25923, *Staphylococcus aureus, Staphylococcus epidermidis* DSMZ 20044, *Bacillus subtilis* DSMZ 1971, *Escherichia coli* ATCC 25922 and *Serratia marcescens*) by using the disk diffusion method and volatile components of the Purple coneflower and Tree heath were determined by GC-MS analysis. The extracts of Purple coneflower and Tree heath showed antibacterial activity against tested microorganisms at different levels.

**Keywords:** *Echinacea purpurea, Erica arborea*, Purple coneflower, Ekinezya, Tree heath, Funda, disc diffusion, GC-MS

# **1. INTRODUCTION**

The use of aromatic plants for different purposes is based on ancient times. Medicinal aromatic plants and their medicinal derivatives are of interest in many countries around the world nowadays different purposes such as herbal medicines, herbs, phytotherapeutics, phytopharmaceuticals and traditional drugs. Herbal Medicinal Products name has been found suitable for use in these kinds of herbal products in order to use a common term by European Medicines Evaluation Agency (EMEA) in European Union countries (Kartal, 2004).

It has been known for many years that natural products have a potential as therapeutic agents especially for infectious diseases (Clardy and Walsh, 2004; Altuner et al., 2010). The many studies support that natural products have been used for hundreds of years to treat several diseases caused by bacteria, fungi, viruses and parasites (Jones, 1996). The new researches have showed that natural products have been a potential of providing opportunities as new drug precursor. *Echinacea*, one of the most important of these plants, is the second top-selling plant in the US market because of its immune modulator properties. Moraes et al. established an in vitro repository of *Echinacea* sp. to produce healthy plants in an effort to identify the active constituents responsible for the immune enhancing activities (Moraes et al.,2011). Pugh et al. reported differences in immune enhancing activities of *Echinacea* shoot cultures, and then, they demonstrated that monocyte and macrophage immune activation is due to lipoproteins and lipopolysaccharides (LPS) of bacterial endophytes (Pugh et al., 2005). Besides, herbal teas are prepared from aerials parts of *Erica arborea* and *Erica manipuliflora* have been popularly used as diuretic, astringent and treatment of urinary infections in Turkey (Baytop, 1999). Many studies have revealed that several species from *Ericaceae* plants such as *Calluna vulgaris* L., *Ledum groenlandicum* Retzius and *Vaccinium mirtillus* L. possess in vivo anti-inflammatory activity (Orhan et al., 2007).

This paper is concerned with ethanol (65%) extracts of *E. purpurea* – (leaf), *E. purpurea* – (flower) and *E. arborea*, their antimicrobial activities against 19 microorganisms by using the disk diffusion method. Furthermore, chemical components of these extracts were determined by GC-MS analysis.

### 2. MATERIALS AND METHODS

## **Plant Samples**

In this study, *E. purpurea* was cultivated in Gölköy (Kastamonu/Turkey) in 750 m at 2018. *E. arborea* was collected from İnebolu (Kastamonu/Turkey) at sea level, at 2018. *E. arborea* (leaf) parts of *E. purpurea* (individually leaf and flower) were used. Plant materials were dried at room temperature and on draft for three weeks. All the plants were identified by Assist. Prof. Dr. Kerim GÜNEY, Department of Forest Engineering, Faculty of Forestry, Kastamonu University.

# **Extraction method**

The plants were washed thoroughly 2-3 times with water, and then, they were air-dried under shade. Afterwards, the dried plant materials were ground in a mixer, the powder was kept in the amber glass bottle. About 10-30 g ground plant samples were extracted with 250 mL of ethanol (%65) in a Soxhlet apparatus by continuous heat extraction for 24 hours. All extract solutions were filtered through Whatman No.1 paper. Then, filtrates were evaporated with a rotary evaporator. The filtrates were freeze dried and stored in refrigerator at about 4°C after sealed with paraffin for further studies.

### **Determination of Antimicrobial Activities**

### **Preparation of Extract Stock**

Extract stocks to test the antimicrobial activity were prepared by dissolving 1 mg of extract in each 3 mL of ethanol for disk diffusion test.

#### Strains

In order to analyse the antimicrobial activity of plants extracts, 19 microorganisms namely, Enterobacter aerogenes ATCC 13048, Salmonella infantis, Listeria monocytogenes ATCC 7644, Klebsiella pneumoniae, Pseudomonas aeruginosa DSMZ 50071, Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis ATCC 29212, Listeria innocua, Salmonella enteritidis ATCC 13075, Enterococcus durans, Salmonella typhimurium SL 1344, Candida ablicans ATCC 10231, Enterococcus faecium ATCC 25923, Staphylococcus aureus, Staphylococcus epidermidis DSMZ 20044, Bacillus subtilis DSMZ 1971, Escherichla coli ATCC 25922 and Serratia marcescens were used.

#### **Preparation of Innocula**

All strains were incubated according to their requirements as it was previously mentioned by Altuner and Canlı (2012) and Canlı et al (2015). For the inocula, morphologically similar colonies of each organism were transferred into physiological saline (Canlı et al, 2016a, Onbaşlı, 2013) and, to adjust equal the number of the colonies in the solution, 0.5 McFarland standard was used (Hammer et al., 1999; Altuner et al., 2012a and b). Thus, standard inocula is adjusted to contain approximately 10<sup>8</sup> cfu/mL<sup>-1</sup> for bacteria and 10<sup>7</sup> cfu/mL<sup>-1</sup> for *C. albicans* (Canlı et al, 2016c and d).

#### **Disk Diffusion Test**

The disk diffusion test was applied accordingly the previous studies (Andrews, 2003; Canlı et al, 2014). Petri dishes containing Mueller Hinton Agar were used for disk diffusion test (İlhan et al., 2006; Canlı et al, 2016e). 20, 40 and 80  $\mu$ L of extracts were loaded on empty sterile antibiotic disks (SAD). Disks were kept at 40°C for 24 h in aseptic conditions (Altuner et al., 2010). Microorganism suspensions were inoculated on the surfaces of the Mueller Hinton plates and left in aseptic conditions for 2-3 minutes before applying disks as described in the previous studies (Altuner et al., 2010). Inhibition zones were defined in mm by the method mentioned by Altuner et al. (2014).

#### Controls

Empty SAD was used as negative controls for disk diffusion test, where broth medium inoculated with each microorganism was used to control microorganisms.

# GC-MS (Gas chromatography-mass spectroscopy) Analysis

GC-MS analysis was performed using Shimadzu GCMS QP 2010 ULTRA series device. Samples were passed through RTX-5MS capillary column (30 m x 0.25 mm x 0.25  $\mu$ m). Helium gas was used as carrier gas. The injection temperature was maintained at 250° C. GC-MS analysis was performed according to the procedure in the literature. The oven is heated to 40° C and is allowed to stand at this temperature for 3 minutes. Then the temperature is increased by 4 degrees per minute to 240° C and waited for 10 minutes. Finally, the temperature is increased by 4 degrees per minute to 260° C and kept for 65 minutes and is completed to a total of 78 minutes. The injection temperature was maintained

at 250° C and the injection volume was determined as 1  $\mu$ l. The intermittent temperature is 250° C and the ion boiling temperature is 200°C. It was treated with hexane.

### 3. RESULT AND DISCUSSION

The GC-MS results of the extracts are presented in Table 1, and the antimicrobial activity test results are presented in Table 2.

|          | E          | chinacea  | purpurea – (leaf)  | Echi       | nacea pr  | ırpurea – (flower)  | Erica arborea |           | arborea                                  |
|----------|------------|-----------|--|------------|-----------|---|---------------|-----------|--|
| Pea<br>k | R.<br>Time | Area<br>% | Compenent Name   | R.<br>Time | Area<br>% | Compenent Name  | R.<br>Time    | Area<br>% | Compenent<br>Name                        |
| 1        | 3.561      | 1.23      | Cyclopentane, 1,3-<br>dimethyl-, cis-  | 29.460     | 3.57      | Tetracosane   | 41.699        | 1.93      | Hexadecanoic<br>acid, methyl ester       |
| 2        | 22.539     | 2.05      | Anethole <(Z)->  | 48.933     | 1.52      | Indolo[2,3-<br>a]quinolizin-4(12H)-<br>one, 1,2,3,6,7,12b-<br>hexahydro-3,12b-<br>dimethyl- | 45.989        | 2.38      | Methyl 9,12,15-<br>octadecatrienoat<br>e |
| 3        | 29.501     | 1.04      | Eicosane   | 75.821     | 5.38      | γ-Sitosterol  | 46.273        | 2.25      | Phytol                                   |
| 4        | 34.491     | 1.76      | Widdrol  |            |           |   | 50.407        | 1.17      | 1-Octanol, 2-<br>butyl-                  |
| 5        | 35.566     | 1.73      | Cycloheptane, 4-<br>methylene-1-methyl-2-<br>(2-methyl-1-propen-1-<br>yl)-1-vinyl-         |            |           |   | 54.238        | 1.58      | DI-(9-<br>OCTADECENOY<br>L)-GLYCEROL     |
| 6        | 37.678     | 4.44      | 4,8,13-Duvatriene-1,3-<br>diol   |            |           |   | 60.666        | 1.90      | Pentacosane                              |
| 7        | 37.929     | 1.08      | (+) - Isonootkatone  |            |           |   | 65.379        | 8.16      | γ-Sitosterol                             |
| 8        | 38.127     | 2.15      | 3-Buten-2-one, 4-(3-<br>hydroxy-6,6-dimethyl-<br>2-<br>methylonogyalabayyl)                |            |           |   | 65.855        | 23.9<br>2 | Squalene                                 |
| 9        | 39.609     | 10.63     | 2-Butenal, 2-methyl-4-<br>(2,6,6-trimethyl-1-<br>cyclohexen-1-yl)-                         |            |           |   | 67.039        | 2.36      | β-Amyrin                                 |
| 10       | 40.795     | 1.08      | 1-Heptatriacotanol   |            |           |   | 67.725        | 1.83      | I-Heptacosanol                           |
| 11       | 41.205     | 1.18      | Isoaromadendrene<br>epoxide  |            |           |   | 68.113        | 7.39      | Hexatriacontane                          |
| 12       | 41.384     | 1.60      | 6-Isopropenyl-4,8a-<br>dimethyl-<br>1,2,3,5,6,7,8,8a-<br>octahydronaphthalene-<br>2,3-diol |            |           |   | 70.333        | 1.72      | α-Amyrin                                 |
| 13       | 41.620     | 1.68      | Octadecane   |            |           |   | 74.600        | 4.11      | 1-Heptacosanol                           |
| 14       | 41.723     | 10.94     | Hexadecanoic acid,<br>methyl ester   |            |           |   | 75.594        | 9.46      | Hexatriacontane                          |
| 15       | 42.092     | 1.44      | 4,6,10,10-Tetramethyl-<br>5-<br>oxatricyclo[4.4.0.0(1,4)]<br>dec-2-en-7-ol                 |            |           |   | 75.915        | 1.99      | β-Amyrin                                 |
| 16       | 45.860     | 10.06     | 9,12-Octadecadienoic<br>acid (Z,Z)-, methyl ester  |            |           |   | 77.196        | 8.45      | dl-a-Tocopherol                          |
| 17       | 46.017     | 6.51      | Methyl 9,12,15-<br>octadecatrienoate   |            |           |   |               |           |  |
| 18       | 46.330     | 4.47      | Phytol   |            |           |   |               |           |  |
| 19       | 46.607     | 2.61      | Methyl stearate  |            |           |   |               |           |  |
| 20       | 48.081     | 1.79      | N-Isobutyl-<br>(2E,4Z,8Z,10E)-<br>dodecatetraenamide                                       |            |           |   |               |           |  |
| 21       | 50.429     | 4.64      | Pentacosane  |            |           |   |               |           |  |
| 22       | 50.534     | 1.01      | cis-11-Eicosenoic acid,<br>methyl ester  |            |           |   |               |           |  |
| 23       | 52.544     | 1.65      | Tetracosane  |            |           |   |               |           |  |

**Table 1.** Echinacea purpurea – (leaf), Echinacea purpurea – (flower) and Erica arborea, GC-MS aroma analysis results

GC-MS aroma analysis of for *Echinacea purpurea* – (leaf) clearly showed the presence of twenty three (23) compounds. Hexadecanoic acid, methyl ester was identified as a major chemical constituent (10,94%) followed by 2-Butenal, 2-methyl-4-(2,6,6-trimethyl-1-cyclohexen-1-yl) (10,63%) and 9,12-Octadecadienoic acid (Z,Z)-, methyl ester (10,06%). The highest values for Echinacea purpurea - (leaf) are as follows; Methyl 9,12,15-octadecatrienoate (6,51%),

Pentacosane (4,64%), Phytol (4,47%), 4,8,13-Duvatriene-1,3-diol (4,44%), Methyl stearate (2,61%), 3-Buten-2-one, 4-(3-hydroxy-6,6-dimethyl-2-methylenecyclohexyl) (2,15%), Anethole  $\langle Z \rangle > (2,05\%)$ , N-Isobutyl-(2E,4Z,8Z,10E)dodecatetraenamide (1,79%), Widdrol (1,76%), Cycloheptane, 4-methylene-1-methyl-2-(2-methyl-1-propen-1-yl)-1vinyl (1,73%), Octadecane (1,68%), Tetracosane (1,65%), 6-Isopropenyl-4,8a-dimethyl-1,2,3,5,6,7,8,8aoctahydronaphthalene-2,3-diol (1,60%), 4,6,10,10-Tetramethyl-5-oxatricyclo[4.4.0.0(1,4)]dec-2-en-7-ol (1,44%), Cyclopentane, 1,3-dimethyl-, cis- (1,23%), Isoaromadendrene epoxide (1,18%), (+) – Isonootkatone (1,08%), 1-Heptatriacotanol (1,08%), Eicosane (1,04%) and cis-11-Eicosenoic acid, methyl ester (1,01%).

GC-MS aroma analysis of for *Echinacea purpurea* – (flower) clearly showed the presence of three (3) compounds.  $\gamma$ -Sitosterol was identified as a major chemical constituent (5,38%) followed by Tetracosane (3,57%) and Indolo[2,3-a]quinolizin-4(12H)-one, 1,2,3,6,7,12b-hexahydro-3,12b-dimethyl- (1,52%).

GC-MS aroma analysis of for *Erica arborea* clearly showed the presence of sixteen (16) compounds. Squalene was identified as a major chemical constituent (23,92%) followed by Hexatriacontane (9,46%), dl- $\alpha$ -Tocopherol (8,45%),  $\gamma$ -Sitosterol (8,16%), Hexatriacontane (7,39%), 1-Heptacosanol (4,11%), Methyl 9,12,15-octadecatrienoate (2,38%),  $\beta$ -Amyrin (2,36%), Phytol (2,25%),  $\beta$ -Amyrin (1,99%), Hexadecanoic acid, methyl ester (1,93%), Pentacosane (1,90%), 1-Heptacosanol (1,83%),  $\alpha$ -Amyrin (1,72%), DI-(9-OCTADECENOYL)-GLYCEROL (1,58%) and 1-Octanol, 2-butyl-(1,17%).

According to the results of GC-MS aroma analysis, the highest chemical component ratio of the *Erica arborea* species is squalene. Squalene is a triterpene and an intermediate in the biosynthesis of sterols in the plant and animal world (Psomiadou, E., Tsimidou, M., 1999). The richest known source of squalene is shark liver oil. In vegetable oils, squalene is found over broad ranges (Tuberoso, C.I.G. et al., 2007). Squalene is the main component of skin surface polyunsaturated lipids and shows some advantages for the skin as an emollient and antitumor compound (Huang, Z.R., et al., 2009).

|                  | E<br>purp | Echinacea<br>purpurea – (leaf) |    |    | Echinacea<br>purpurea –<br>(flower) |    |    | Erica arborea |    |  |
|------------------|-----------|--------------------------------|----|----|-------------------------------------|----|----|---------------|----|--|
|                  | 20        | 40                             | 80 | 20 | 40                                  | 80 | 20 | 40            | 80 |  |
|                  | μL        | μL                             | μL | μL | μL                                  | μL | μL | μL            | μL |  |
| E. aerogenes     | -         | -                              | -  | -  | -                                   | -  | -  | -             | -  |  |
| S. infantis      | -         | -                              | -  | -  | -                                   | 8  | -  | -             | -  |  |
| L. monocytogenes | -         | -                              | 8  | -  | -                                   | -  | -  | 8             | 8  |  |
| K. pneumoniae    | 8         | 8                              | 8  | 8  | 8                                   | 8  | 8  | 8             | 8  |  |
| P. aeruginosa    | -         | -                              | -  | -  | -                                   | -  | -  | -             | -  |  |
| P. fluorescens   | -         | -                              | -  | -  | -                                   | -  | -  | -             | -  |  |
| S. kentucky      | 7         | 7                              | 7  | -  | -                                   | 7  | -  | -             | 7  |  |
| E. faecalis      | -         | -                              | -  | -  | -                                   | -  | -  | -             | -  |  |
| L. innocua       | -         | -                              | 7  | -  | -                                   | -  | -  | -             | -  |  |
| S. enteritidis   | 7         | 8                              | 8  | -  | 7                                   | 7  | -  | -             | -  |  |
| E. durans        | -         | -                              | -  | -  | 9                                   | 10 | -  | -             | -  |  |
| S. typhimurium   | -         | 7                              | 7  | -  | -                                   | -  | -  | -             | -  |  |
| C. ablicans      | -         | -                              | -  | -  | -                                   | -  | -  | -             | -  |  |
| E. faecium       | 7         | 7                              | 7  | -  | 7                                   | 7  | 7  | 7             | 8  |  |
| S. ayreus        | -         | -                              | -  | _  | 9                                   | 11 | -  | 7             | 8  |  |
| S. epidermidis   | -         | -                              | 7  | -  | -                                   | -  | -  | -             | 7  |  |
| B. subtilis      | -         | -                              | 7  | -  | -                                   | -  | -  | -             | 7  |  |
| E. coli          | -         | -                              | -  | -  | -                                   | -  | -  | -             | -  |  |
| S. marcescens    | -         | -                              | -  | 7  | 7                                   | 8  | -  | -             | -  |  |

**Table 2.** Disk Diffusion Test Results of extracts at the *Echinacea purpurea* – (leaf), *Echinacea purpurea* – (flower) and *Erica arborea* concentration

Disk diffusion test results showed that E. purpurea – (leaf) is active against L. monocytogenes, K. pneumoniae, S. kentucky, L. innocua, S. enteritidis, S. typhimurium, E. faecium, S. epidermidis and B. subtilis with inhibition zones between 7-8 mm, where E. purpurea – (flower) is active against S. infantis, K. pneumoniae, S. kentucky, S. enteritidis,
E. durans, E. faecium, S. ayreus and S. marcescens with inhibition zones between 7-11 mm, where E. arborea is active against L. monocytogenes, K. pneumoniae, S. kentucky, E. faecium, S. ayreus, S. epidermidis and B. subtilis with inhibition zones between 7-8 mm.

#### 4. CONCLUSION

In terms of antimicrobial activity, it was observed that the extracts were effective only for a few of the tested species. Consequently, these extracts require an antimicrobial agent for use in the medicine and cosmetics sector.

The importance of the study is due to the biological activities of some of these compounds. The present study, which reveals the presence of components in *Echinacea purpurea* and *Erica arborea* suggest that the contribution of these compounds on the pharmacological activity should be evaluated.

#### REFERENCES

- Altuner E.M., Çeter T., İşlek C. 2010. Investigation of antifungal activity of *Ononis spinose* L. ash used for the therapy of skin infections as folk remedies. Mikrobiyoloji Bülteni, 44, 633-639.
- Altuner, E.M. and I. Akata. 2010. Antimicrobial activity of some macrofungi extracts. Sakarya Üniversitesi Fen Bilimleri Enstitüsü Dergisi. 14(1):45-49.
- Altuner, E.M. 2011. Investigation of antimicrobial activity of *Punica granatum* L. fruit peel ash used for protective against skin infections as folk remedies especially after male circumcision. African Journal of Microbiology Research. 5(20):3339-3342.
- Altuner, E.M. and K. Canlı. 2012. In vitro antimicrobial screening of Hypnum andoi A.J.E. Sm. Kastamonu U. J. For. Fac. 12:97-101.
- Altuner, E.M., I. Akata and K. Canli. 2012a. *In vitro* antimicrobial screening of *Bovista nigrescens* (Pers.). Kastamonu U. J. For. Fac. 12:90-96.
- Altuner, E.M., I. Akata and K. Canlı. 2012b. In vitro antimicrobial screening of Cerena unicolor (Bull.) Murrill (Polyporaceae Fr. Ex Corda). Fresen. Environ. Bullet. 21:3704-3710.
- Altuner, E.M., K. Canlı and I. Akata. 2014. Antimicrobial screening of *Calliergonella cuspidata*, *Dicranum polysetum* and *Hypnum cupressiforme*. Journal of Pure and Applied Microbiology. 8(1):539-545.
- Andrews, J.M. 2003. BSAC standardized disc susceptibility testing method (version 6). Journal of Antimicrobial Chemotherapy. 60:20-41.
- Baytop T. 1999. "Türkiye'de Bitkilerle Tedavi, Geçmişte ve Bugün" Nobel Tıp Kitabevleri, İstanbul, p. 208.
- Canlı, K., B. Çetin, E.M. Altuner, Y. Türkmen, U. Üzek and H. Dursun. 2014. In vitro antimicrobial screening of Hedwigia ciliata var. leucophaea and determination of the ethanol extract composition by gas chromatography/mass spectrometry (GC/MS). Journal of Pure and Applied Microbiology. 8(4):2987-2998.
- Canlı, K., E.M. Altuner and I. Akata. 2015. Antimicrobial screening of *Mnium stellare*. Bangladesh Journal of Pharmacology. 10:321-325.
- Canlı, K., E.M. Altuner, I. Akata, Y. Türkmen and U. Üzek. 2016a. In vitro antimicrobial screening of Lycoperdon lividium and determination of the ethanol extract composition by gas chromatography/mass spectrometry. Bangladesh Journal of Pharmacology. 11(2):389-394.
- Canlı, K., I. Akata and E.M. Altuner. 2016c. *In vitro* antimicrobial activity screening of *Xylaria hypoxylon*. African Journal of Traditional, Complementary and Alternative Medicines. 13(4):42-46.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016d. *In vitro* antimicrobial screening of *Aquilaria agallocha* roots. African Journal of Traditional, Complementary and Alternative Medicines. 13(5):178-181.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016e. *In vitro* antimicrobial activity screening of *Rheum rhabarbarum* roots. International Journal of Pharmaceutical Sciences Invention, 5(2):1-4.
- Clardy J., Wash C. 2004. Lessons from natural molecules. Nature, 432, 829-837.
- Hammer, K.A., C.F. Carson and T.V. Riley. 1999. Antimicrobial activity of essential oils and other plant extracts. Journal of Applied Microbiology. 86:985-990.
- Huang, Z.R., Lin, Y.K., Fang, J.Y. 2009. Biological and pharmacological activities of squalene and related compounds: Potential uses in cosmetic dermatology. Molecules 2009, 14, 540–554.
- İlhan, S., F. Savaroğlu, F. Çolak, C.F. Iscen and F.Z. Erdemgil. 2006. Antimicrobial activity of *Palustriella commutata* (Hedw.) Ochyra extracts (Bryophyta). Turk. J. Biol, 30:149-152.
- Jones F.A. 1996. Herbs useful plants. Their role in history and today. European Journal of Gastroenterology and Hepatology, 8, 1227-1231.

- Kartal, M. 2004. Avrupa birliği ülkelerinde tıbbi bitkisel ürünlerin ruhsatlandırılması, Ankara Üniversitesi Eczacılık Fakültesi Farmakognozi Anabilim Dalı.
- Moraes R.M., Lata H., Sumyanto J., Pereira A.M.S., Bertoni W., Joshi V.C., Pugh N.D., Khan I.A. and Pasco D.S. 2011. "Characterization and Pharmacological Properties of in Vitro Propagated Clones of *Echinacea tennesseensis* (Beadle) Small," Plant Cell, Tissue and Organ Culture", Vol. 106, No. 2, pp. 309-315.
- Onbaşlı, D., G. Yuvalı Çelik, E.M. Altuner, B. Altunsoy and B. Aslim. 2013. *In vitro* antimicrobial, antioxidant, and antibiofilm activities of *Bryum capillare*, a bryophyte sample. Current Opinion in Biotechnology, 24 (Supplement 1): 113.
- Orhan, İ., Küpeli, E., Terzioğlu, S. and Yeşilada, E. 2007. "Bioassay-guided isolation of kaempferol-3-3O-β-D-galactoside with antiinflammatory and antinociceptive activity from the aerial part of *Calluna vulgaris* L." Journal of Ethnopharmacology, 114 (2007), pp. 32-37.
- Psomiadou, E., Tsimidou, M. 1999. On the role of squalene in olive oil stability. J Agric Food Chem. 1999, 47, 4025–4032.
- Pugh N.D., Balachandran P., Lata H., Dayan F.E., Joshi V., Bedir E., Makino T., Moraes R., Khan I. and Pasco D.S. 2005. "Melanin: Dietary Mucosal Immune Modulator from *Echinacea* and Other Botanical Supplements," International Immunopharmacology, Vol. 5, No. 4, pp. 637-647.
- Tuberoso, C.I.G., Kowalczyk, A., Sarritzu, E. and Cabras, P. 2007. Determination of antioxidant compounds and antioxidant activity in comercial oilseeds for food use. Food Chem. 2007, 103, 1494–1501.



# Some Medicinal Plant Extracts in Turkey: Antimicrobial Activity and Gc-Ms Analysis

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**Abstract:** In this study, the antimicrobial activities of the ethanolic extracts of *Inula viscosa* (False yellowhead/Sümenit) and *Chrysophthalmum montanum* (Tutça), used in the folk-medicine, were tested against gramnegative and gram-positive bacteria as well as yeast-like fungi by the agar diffusion method. 19 microorganisms namely, *Enterobacter aerogenes* ATCC 13048, *Salmonella infantis, Listeria monocytogenes* ATCC 7644, *Klebsiella pneumoniae, Pseudomonas aeruginosa* DSMZ 50071, *Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis* ATCC 29212, *Listeria innocua, Salmonella enteritidis* ATCC 13075, *Enterococcus durans, Salmonella typhimurium* SL 1344, *Candida ablicans* ATCC 10231, *Enterococcus faecium* ATCC 25923, *Staphylococcus aureus, Staphylococcus epidermidis* DSMZ 20044, *Bacillus subtilis* DSMZ 1971, *Escherichia coli* ATCC 25922 and *Serratia marcescens* were used. Gas-Chromatography Mass Spectrometry (GC-MS) analysis of their ethanol extracts were carried out in Shimadzu GCMS-QP-2010 system. The extracts of False yellowhead and Tutça showed antibacterial activity against tested microorganisms at different levels.

Keywords: Inula viscosa, False yellowhead, Sümenit, Chrysophthalmum montanum, Tutça, disc diffusion, GC-MS

## **1. INTRODUCTION**

Microbial evolution and antibiotic resistance as a result of the increase in use of anti-infective drugs worldwide has been defined as the major threat for the public health in the 21<sup>st</sup> century by World Health Organization (Syed et al., 2010). Medicinal plants have been used for a wide variety of purposes for many thousand of years in Turkey and all over the world. In particular, extracts and oils of these plants have formed the basis of many applications, including raw and processed food preservation, pharmaceutical, alternative medicine, and natural therapies (Baytop, 1984). Although a tremendous progress has been made in human medicine in the last decades; bacterial, fungal and viral diseases are still threatening the public health in the developing countries (Cos et al., 2006). In these countries, the major problem is not only the relative unavailability of medicines but also, the extensive drug resistance has also a large impact on human health (Okeke et al., 2005).

*Inula viscosa* (L.) Aiton (Compositae) (common local name: Taioon) is a perennial plant distributed in different regions of the Mediterranean Basin (Çelik, T. and Aslantürk, O., 2010). In traditional medicine, *Inula viscosa* has many uses, including anti-inflammatory, anthelmintic, lung disorders, antipyretic, antiseptic, and antiphlogistic activities [9,10] in addition to treating gastroduodenal disorders. Crude extracts prepared from different parts of Inula viscosa exhibit antifungal, antioxidant, antiulcerogenic and anthelmintic properties and prevent zygote implantation (Al-Dissi, N. Et al., 2001). In previous studies, it was reported the potent antiproliferative and antimicrobial activities of an *Inula viscosa* methanol extract. Chemical analysis showed that *Inula viscosa* contains many biologically active compounds, including flavonoids and terpenoids (Talib, W. and Mahasneh, A., 2010).

This paper is concerned with antimicrobial activity of ethanol (65%) extracts of *I. viscosa* and *C. montanum* against 19 microorganisms by using the disk diffusion method, and chemical components of the *I. viscosa* and *C. montanum* were determined by GC-MS analysis.

#### 2. MATERIALS AND METHODS

## **Plant Samples**

In this study, *I. viscosa* (above ground parts) was collected from Aydıncık (Mersin/Turkey), at sea level at 2018. *C. montanum* was collected from Eskiköy (Adıyaman/Sincik) in 1100 m, at 2018. *I. viscosa* (above ground parts) of *C. montanum* (above ground parts) were used. Plant materials were dried at room temperature and on draft for three weeks. All the plants were identified by Assist. Prof. Dr. Kerim GÜNEY, Department of Forest Engineering, Faculty of Forestry, Kastamonu University.

## **Extraction method**

The plants were washed thoroughly 2-3 times with water, and then, they were air dried under shade. Afterwards, the dried plant materials were ground in a mixer, the powder was kept in the amber glass bottle. About 10-30 g ground plant samples were extracted with 250 mL of ethanol (%65) in a Soxhlet apparatus by continuous heat extraction for 24 hours. All extract solutions were filtered through Whatman No.1 paper. Then, filtrates were evaporated with a rotary evaporator. The filtrates were freeze dried and stored in refrigerator at about 4°C after sealed with paraffin for further studies.

## **Determination of Antimicrobial Activities**

## **Preparation of Extract Stock**

Extract stocks to test the antimicrobial activity were prepared by dissolving 1 mg of extract in each 3 mL of ethanol for disk diffusion test.

#### Strains

In order to analyse the antimicrobial activity of plants extracts, 19 microorganisms namely, Enterobacter aerogenes ATCC 13048, Salmonella infantis, Listeria monocytogenes ATCC 7644, Klebsiella pneumoniae, Pseudomonas aeruginosa DSMZ 50071, Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis ATCC 29212, Listeria innocua, Salmonella enteritidis ATCC 13075, Enterococcus durans, Salmonella typhimurium SL 1344, Candida ablicans ATCC 10231, Enterococcus faecium ATCC 25923, Staphylococcus aureus, Staphylococcus epidermidis DSMZ 20044, Bacillus subtilis DSMZ 1971, Escherichla coli ATCC 25922 and Serratia marcescens were used.

#### **Preparation of Innocula**

All strains were incubated according to their requirements as it was previously mentioned by Altuner and Canlı (2012) and Canlı et al (2015). For the inocula, morphologically similar colonies of each organism were transferred into physiological saline (Canlı et al, 2016a, Onbaşlı, 2013) and to adjust equal the number of the colonies in the solution, 0.5 McFarland standard was used (Hammer et al., 1999; Altuner et al., 2012a and b). Thus standard inocula is adjusted to contain approximately  $10^8$  cfu/mL<sup>-1</sup> for bacteria and  $10^7$  cfu/mL<sup>-1</sup> for *C. albicans* (Canlı et al, 2016c and d).

#### **Disk Diffusion Test**

The disk diffusion test was applied as it was mentioned in the previous studies (Andrews, 2003; Canlı et al, 2014). Petri dishes containing Mueller Hinton Agar were used for disk diffusion test (İlhan et al., 2006; Canlı et al, 2016e). 20, 40 and 80  $\mu$ L of extracts were loaded on empty sterile antibiotic disks (SAD). Disks were kept at 40°C for 24 h in aseptic conditions (Altuner et al., 2010). Microorganism suspensions were inoculated on the surfaces of the Mueller Hinton plates and left in aseptic conditions for 2-3 minutes before applying disks as described in the previous studies (Altuner et al. (2014).

#### Controls

Empty SAD was used as negative controls for disk diffusion test, where broth medium inoculated with each microorganism was used to control microorganisms.

#### GC-MS (Gas chromatography-mass spectroscopy) Analysis

GC-MS analysis was performed using Shimadzu GCMS QP 2010 ULTRA series device. Samples were passed through RTX-5MS capillary column (30 m x 0.25 mm x 0.25  $\mu$ m). Helium gas was used as carrier gas. The injection temperature was maintained at 250° C. GC-MS analysis was performed according to the procedure in the literature. The oven is heated to 40° C and is allowed to stand at this temperature for 3 minutes. Then the temperature is increased by 4 degrees per minute to 240° C and waited for 10 minutes. Finally, the temperature is increased by 4 degrees per minute to 260° C and kept for 65 minutes and is completed to a total of 78 minutes. The injection temperature was maintained

at 250° C and the injection volume was determined as 1  $\mu$ l. The intermittent temperature is 250° C and the ion boiling temperature is 200°C. It was treated with hexane.

## 3. RESULT AND DISCUSSION

The GC-MS results of the extracts are presented in Table 1, and the antimicrobial activity test results are presented in Table 2.

|      |            | ula viscosa | Chrysophthalmum montanum   |            |           |  |
|------|------------|-------------|--|------------|-----------|--|
| Peak | R.<br>Time | Area<br>%   | Compenent Name   | R.<br>Time | Area<br>% | Compenent Name   |
| 1    | 46.663     | 6.80        | 2-one, 3,3a,4,7,8,8a-<br>hexahydro-7-methyl-3-<br>methylene-6-(3-<br>oxobutyl)-, [3aR-<br>(3a.alpha.,7.beta.,8a.alph<br>a) | 3.703      | 1.19      | 2,2-<br>dimethoxybutane  |
| 2    | 47.636     | 8.04        | 2-one, 3,3a,4,7,8,8a-<br>hexahydro-7-methyl-3-<br>methylene-6-(3-<br>oxobutyl)-, [3aR-<br>(3a.alpha.,7.beta.,8a.alph<br>a) | 41.707     | 2.20      | Hexadecanoic acid,<br>methyl ester   |
| 3    | 47.750     | 4.78        | 2-one, 3,3a,4,7,8,8a-<br>hexahydro-7-methyl-3-<br>methylene-6-(3-<br>oxobutyl)-, [3aR-<br>(3a.alpha.,7.beta.,8a.alph<br>a) | 45.841     | 1.17      | Metil linoleat   |
| 4    | 54.703     | 1.05        | Pentacosane  | 45.983     | 1.05      | Methyl 6-<br>octadecenoate   |
| 5    | 60.696     | 3.00        | Pentacosane  | 46.585     | 1.03      | Methyl stearate  |
| 6    | 65.313     | 1.01        | Heptacosyl acetate   | 48.065     | 1.39      | Methyl-6-(2,6,6-<br>trimethylcyclohex-<br>1-enyl)hexa-1,3,5-<br>trienyl]cyclohex-1-<br>en-1-<br>carboxaldehyde |
| 7    | 65.561     | 4.66        | γ-Sitosterol   | 50.404     | 1.11      | Octacosane   |
| 8    | 65.886     | 42.84       | Squalene   | 54.689     | 9.68      | Tetracosane  |
| 9    | 68.132     | 3.50        | Hexatriacontane  | 57.341     | 1.28      | Hexacosane   |
| 10   | 68.365     | 1.00        | cis-1-Chloro-9-<br>octadecene  | 60.677     | 12.95     | Tetratetracontane  |
| 11   | 69.264     | 1.30        | Hexacosanoic acid,<br>methyl ester   | 65.852     | 23.76     | Squalene   |
| 12   | 75.623     | 3.18        | Hexatriacontane  | 68.117     | 6.78      | Tetracontane   |
| 13   |            |             |  | 75.595     | 4.25      | Hexatriacontane  |

 Table 1. Inula viscosa, Chrysophthalmum montanum, GC-MS aroma analysis results

GC-MS aroma analysis of for *Inula viscosa* clearly showed the presence of twelve (12) compounds. Squalene was identified as a major chemical constituent (42,84%) followed by 2-one, 3,3a,4,7,8,8a-hexahydro-7-methyl-3-methylene-6-(3-oxobutyl)-, [3aR-(3a.alpha.,7.beta.,8a.alpha) (8,04%), 2-one, 3,3a,4,7,8,8a-hexahydro-7-methyl-3-methylene-6-(3-oxobutyl)-, [3aR-(3a.alpha.,7.beta.,8a.alpha) (6,80%), 2-one, 3,3a,4,7,8,8a-hexahydro-7-methyl-3-methylene-6-(3-oxobutyl)-, [3aR-(3a.alpha.,7.beta.,8a.alpha) (6,80%), 2-one, 3,3a,4,7,8,8a-hexahydro-7-methyl-3-methylene-6-(3-oxobutyl)-, [3aR-(3a.alpha.,7.beta.,8a.alpha) (6,80%), 2-one, 3,3a,4,7,8,8a-hexahydro-7-methyl-3-methylene-6-(3-oxobutyl)-, [3aR-(3a.alpha.,7.beta.,8a.alpha) (4,78%),  $\gamma$ -Sitosterol (4,66%), Hexatriacontane (3,50%), Hexatriacontane (3,18%), Pentacosane (3,00%), Hexacosanoic acid, methyl ester (1,30%), Pentacosane (1,05%), Heptacosyl acetate (1,01%) and cis-1-Chloro-9-octadecene (1,00%).

GC-MS aroma analysis of for *Chrysophthalmum montanum* clearly showed the presence of thirteen (13) compounds. Squalene was identified as a major chemical constituent (23,76%) followed by Tetratetracontane (12,95%), Tetracosane

(9,68%), Tetracontane (6,78%), Hexatriacontane (4,25%), Hexadecanoic acid, methyl ester (2,20%), Methyl-6-(2,6,6-trimethylcyclohex-1-enyl)hexa-1,3,5-trienyl]cyclohex-1-en-1-carboxaldehyde (1,39%), Hexacosane (1,28%), 2,2-dimethoxybutane (1,19%), Metil linoleat (1,17%), Octacosane (1,11%), Methyl 6-octadecenoate (1,05%) and Methyl stearate (1,03%).

According to the results of GC-MS aroma analysis, the highest chemical component ratio of the *Inula viscosa* and *Chrysophthalmum montanum* species is squalene. Squalene is a triterpene and an intermediate in the biosynthesis of sterols in the plant and animal World (Psomiadou, E., Tsimidou, M., 1999). The richest known source of squalene is shark liver oil. In vegetable oils, squalene is found over broad ranges (Tuberoso, C.I.G. et al., 2007). Squalene is the main component of skin surface polyunsaturated lipids and shows some advantages for the skin as an emollient and antitumor compound (Huang, Z.R., et al., 2009).

|                  | In       | ula visco | osa      | Chrysophthalmun<br>montanum |          |          |
|------------------|----------|-----------|----------|-----------------------------|----------|----------|
|                  | 20<br>uL | 40<br>uL  | 80<br>цГ | 20<br>uL                    | 40<br>uL | 80<br>иL |
| E. aerogenes     | -        | -         | -        | -                           | -        | -        |
| S. infantis      | -        | -         | -        | -                           | -        | -        |
| L. monocytogenes | -        | 8         | 8        | -                           | -        | -        |
| K. pneumoniae    | 8        | 8         | 8        | -                           | -        | 8        |
| P. aeruginosa    | -        | -         | -        | -                           | -        | -        |
| P. fluorescens   | -        | -         | -        | -                           | -        | -        |
| S. kentucky      | -        | 7         | 7        | -                           | -        | -        |
| E. faecalis      | -        | -         | -        | -                           | 7        | 8        |
| L. innocua       | -        | 9         | 10       | -                           | -        | -        |
| S. enteritidis   | -        | -         | -        | -                           | 7        | 7        |
| E. durans        | -        | 11        | 14       | -                           | -        | -        |
| S. typhimurium   | -        | 7         | 9        | -                           | 7        | 8        |
| C. ablicans      | -        | 8         | 10       | -                           | -        | -        |
| E. faecium       | 7        | 7         | 7        | -                           | -        | -        |
| S. aureus        | 8        | 10        | 11       | -                           | 7        | 9        |
| S. epidermidis   | -        | -         | -        | -                           | -        | -        |
| B. subtilis      | -        | -         | -        | -                           | -        | -        |
| E. coli          | -        | -         | -        | -                           | -        | -        |
| S. marcescens    | -        | 7         | 8        | -                           | -        | -        |

Table 2. Disk Diffusion Test Results of extracts at the Inula viscosa and Chrysophthalmum montanum concentration

Disk diffusion test results showed that I. viscosa is active against L. monocytogenes, K. pneumoniae, S. kentucky, L. innocua, E. durans, S. typhimurium, C. ablicans, E. faecium, S. aureus and S. marcescens with inhibition zones between 7-14 mm, where C. montanum is active against K. pneumoniae, E. faecalis, S. enteritidis, S. typhimurium, and S. aureus with inhibition zones between 7-9 mm.

#### 4. CONCLUSION

In conclusion, this study provides new scientific information about *Inula viscosa* and *Chrysophthalmum montanum*, interested with their secondary metabolites, antibacterial potential and chemical. The antibacterial activities of *I. viscosa* and *C. montanum* may be attributed to the various phytochemical constituents present in the extract. Further work on the types of chemical constituents and purification of individual groups of bioactive components could reveal the full potential of the *I. viscosa* and *C. montanum* extract to inhibit several pathogenic microbes.

#### REFERENCES

- Altuner E.M., Çeter T., İşlek C. 2010. Investigation of antifungal activity of *Ononis spinose* L. ash used for the therapy of skin infections as folk remedies. Mikrobiyoloji Bülteni, 44, 633-639.
- Altuner, E.M. and I. Akata. 2010. Antimicrobial activity of some macrofungi extracts. Sakarya Üniversitesi Fen Bilimleri Enstitüsü Dergisi. 14(1):45-49.
- Altuner, E.M. and K. Canlı. 2012. In vitro antimicrobial screening of Hypnum andoi A.J.E. Sm. Kastamonu U. J. For. Fac. 12:97-101.
- Altuner, E.M., I. Akata and K. Canli. 2012a. *In vitro* antimicrobial screening of *Bovista nigrescens* (Pers.). Kastamonu U. J. For. Fac. 12:90-96.
- Altuner, E.M., I. Akata and K. Canlı. 2012b. In vitro antimicrobial screening of Cerena unicolor (Bull.) Murrill (Polyporaceae Fr. Ex Corda). Fresen. Environ. Bullet. 21:3704-3710.
- Altuner, E.M., K. Canlı and I. Akata. 2014. Antimicrobial screening of *Calliergonella cuspidata*, *Dicranum polysetum* and *Hypnum cupressiforme*. Journal of Pure and Applied Microbiology. 8(1):539-545.
- Al-Dissi, N.; Salhab, A.; Al-Hajj, H. 2001. Effects of *Inula viscosa* leaf extracts on abortion and implantation in rats. J. Ethnopharmacol. 77, 117–121.
- Andrews, J.M. 2003. BSAC standardized disc susceptibility testing method (version 6). Journal of Antimicrobial Chemotherapy. 60:20-41.
- Baytop, T. 1984. Health treatment in Turkey using plant extracts. Publication of the İstanbul University No. 3255.
- Canlı, K., B. Çetin, E.M. Altuner, Y. Türkmen, U. Üzek and H. Dursun. 2014. In vitro antimicrobial screening of Hedwigia ciliata var. leucophaea and determination of the ethanol extract composition by gas chromatography/mass spectrometry (GC/MS). Journal of Pure and Applied Microbiology. 8(4):2987-2998.
- Canlı, K., E.M. Altuner and I. Akata. 2015. Antimicrobial screening of *Mnium stellare*. Bangladesh Journal of Pharmacology. 10:321-325.
- Canlı, K., E.M. Altuner, I. Akata, Y. Türkmen and U. Üzek. 2016a. *In vitro* antimicrobial screening of *Lycoperdon lividium* and determination of the ethanol extract composition by gas chromatography/mass spectrometry. Bangladesh Journal of Pharmacology. 11(2):389-394.
- Canlı, K., I. Akata and E.M. Altuner. 2016c. *In vitro* antimicrobial activity screening of *Xylaria hypoxylon*. African Journal of Traditional, Complementary and Alternative Medicines. 13(4):42-46.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016d. *In vitro* antimicrobial screening of *Aquilaria agallocha* roots. African Journal of Traditional, Complementary and Alternative Medicines. 13(5):178-181.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016e. *In vitro* antimicrobial activity screening of *Rheum rhabarbarum* roots. International Journal of Pharmaceutical Sciences Invention, 5(2):1-4.
- Cos, P., Vlietinck, A.J., Vanden Berghe D. and Maes L. 2006. Anti-infective potential of natural products: How to develop a stronger in vitro 'proof-of-concept'. Journal of Ethnopharmacology, 106, 290-302.
- Çelik, T.; Aslantürk, O. 2010. Evaluation of Cytotoxicity and Genotoxicity of *Inula viscosa* Leaf Extracts with Allium Test. J. Biomed. Biotechnol.
- Hammer, K.A., C.F. Carson and T.V. Riley. 1999. Antimicrobial activity of essential oils and other plant extracts. Journal of Applied Microbiology. 86:985-990.
- Huang, Z.R., Lin, Y.K., Fang, J.Y. 2009. Biological and pharmacological activities of squalene and related compounds: Potential uses in cosmetic dermatology. Molecules 2009, 14, 540–554.
- İlhan, S., F. Savaroğlu, F. Çolak, C.F. Iscen and F.Z. Erdemgil. 2006. Antimicrobial activity of *Palustriella commutata* (Hedw.) Ochyra extracts (Bryophyta). Turk. J. Biol, 30:149-152.
- Okeke I.N., Laxmaninarayan R., Bhutta Z.A., Duse A.G., Jenkins P., O'Brien T.F., PablosMendez A. and Klugman, K.P. 2005. Antimicrobial resistance in developing countries. Part 1: recent trends and current status. Lancet Infectious Diseases, 5, 481-493.
- Onbaşlı, D., G. Yuvalı Çelik, E.M. Altuner, B. Altınsoy and B. Aslim. 2013. *In vitro* antimicrobial, antioxidant, and antibiofilm activities of *Bryum capillare*, a bryophyte sample. Current Opinion in Biotechnology, 24 (Supplement 1): 113.
- Psomiadou, E., Tsimidou, M. 1999. On the role of squalene in olive oil stability. J Agric Food Chem. 1999, 47, 4025–4032.
- Syed, G.W., Syed, A.S., Oh, L.A. 2010. Risk Evaluation Under Various Speculations of Antibiotic Usage; A Cohort Survery Among Outpatients of Pinang, Malaysia. Eur. J Gen Med, 7, 303-309.
- Talib, W.; Mahasneh, A. 2010. Antiproliferative Activity of Plant Extracts Used Against Cancer in Traditional Medicine. Sci. Pharm. 78, 33-45.
- Tuberoso, C.I.G., Kowalczyk, A., Sarritzu, E. and Cabras, P. 2007. Determination of antioxidant compounds and antioxidant activity in comercial oilseeds for food use. Food Chem. 2007, 103, 1494–1501.



# Mapping the Scientific Communications of Silvicultural System in Turkey During 1990-2018

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Abstract: Bibliometric methods are widely used for the analysis of scientific communications and their characteristics by information scientists. On the other hand, social network analysis (SNA) techniques measure the relationships among people. The objective of this research is to analyze and map the intellectual property of the silvicultural as a research field in Turkey between two periods: 1988-2008 and 2009-2018 using bibliometric and SNA methods. We used the "thesaurus" of Commonwealth Agricultural (CAB) abstract to define the search terms. A total of 22 terms were extracted from CAB Abstracts based on the word "silvicultural system", including 15 narrower terms and related terms. Then, with the help of an expert in the field of the silvicultural, we finalized a test query consist of 32 terms. To conduct the research, (Retrieved 03.25.2019) a total of articles were downloaded from Web of Science (WoS) Clarivate Analytics (online academic database) using a compound text query having at least a Turkish address. We extracted 280, 1800 records all document types between 1988-2008, and 2009-2018 from WoS respectively. Then, we used Bibliometrix an R package and Gephi to perform a bibliometric and social network analysis of the articles. The growth rate of scholarly communications between each period is 54%. Co-word analysis based on the authors' key-word in the articles show that the network structure of articles between 1998 and 2008 consist of three clusters depicting climate change, biomass and sustainable in the left side of the graph- land use, remote sensing in the center, and forest management in the right side of the graph. Whereas network structure of articles between 2009 and 2018 is much denser than the previous network consists of six clusters having climate change in the center of the graph followed by forest management on the top, greenhouse in the left side, sustainability in the left corner, water in the bottom, and mann-kedall, client in the left corner of the graph. Results indicate that climate change as a research topic plays a pivotal role in the network structure connecting all clusters with each other.

Keywords: Bibliometric, Mapping, Social network analysist, silvicultural system, R, Gephi, VOSviewer



# ORAL PRESENTATION

# Assessment on The Damage of Harvesting in the Residual Stand

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**Abstract**: In the sustainable forest management approach, although forestry requires harvesting, this process cannot be completed without damage. However, it is necessary to focus on the methods that determine the types and degrees of damage, and the measures that should be taken. As a result of the harvesting in the residual stand, various damage groups are seen in the sapling, forest soil and the wood raw material. Particularly, damage to the residual stand makes the forest vulnerable to various insect epidemics, especially the secondary bark beetle infestation. Within the scope of this study, it will be tried to determine the degree of damage created by the harvesting activities on the individuals in the residual stand and the possible measures. In this study, all of a section with 6,9 hectares of area in the sub-district directorate within borders of Daday Forest Management Directorate was taken into consideration and the wound status was observed in the individuals residual in the stand after harvesting. In the section, 804 pcs. of 553,156 m3 planted bark trees were stamped and 33 trees were indicated to be wounded after the harvesting residual in the stand.

Keywords: Harvesting, skidding, residual stand, damage

## 1. INTRODUCTION

Harvesting is defined as "All efforts made to increase the quantity and usefulness level of scarce goods" in the most general approach (Türker, 2000; Acar, 2004). Harvesting in forestry, in the shortest definition, is the utilization of primary and secondary products that are material wealth and supply by making ready for consumption (Acar, 2004; Enez, 2008).

The task of forestry is to operate forests according to the principles of forestry science and technology, to meet the need for various forest products of the country in a continuous, adequate and economical way and to secure the other functions of forests that are indispensable for the society and the country (Gürtan, 1975). What first comes to mind when considered harvesting in forestry is the production of primary forest products (log, pole, stake, rod, traverse, industrial wood, fire wood, fiber and cellulose).

The harvesting of wood raw materials in our country begins with stamping made in line with the silvicultural demands of the forest. The trees stamped in this stage are quantified by considering Planted Barked Trunk Volume (DKGH-PBTV). Thus, the eta required to be taken according to the plan is determined. However, this amount does not specify the amount of product to be obtained. For this, the trees are required to be cut, limbing, bucking, measured and divided according to standards, product prepared in the cutting area to be carried to forest road, that is skidding and loaded, transported to forest storages, unloaded and stacked. At the end of this process, the volume of round wood obtained specifies the amount of products (Acar and Şentürk, 1996; Karaman, 1997; Topalak, 1998, Eker, 2004; Enez, 2008; Ünver, 2008; Buğday, 2011; Erdaş et.al., 2014). This creates the harvesting cycle of the wood raw material in the forestry (Figure 1).



Figure 1. Harvesting cycle

As can be seen from the cycle, the damage on the residual stand is created during the cutting, felling and skidding stages. Harvesting work is made together with the public in our country. This makes the residual stand damage that can occur each time depending on the skill and method used by the workers.

We can classify the damages that can occur during harvesting as damage on the wood raw material produced, damage on the forest soil and damages on the residual stand (trees and saplings). Damage on the residual stand particularly creates wounds on the trees, infection foci for bark insects and causes severe, economic and ecological losses (Ünver, 2008). It is known that wounded trees cause an effect increasing beetle harm (Lempérière, 1994), therefore the wounds occurred on the tree trunks can weaken the tree, enables entrance of insects and other disease causing organisms into the tree (Neely 1988). Wounds of various sizes on the trunks of trees causes the trees to become stressed and sensitive to the attacks of bark beetle (DeGomez and Celaya, 2013). Damages occurred in the stand as a result of harvesting negatively impacts the product quality as well as the efficiency of the forest.

Low resin pressure caused by the wounds increase the exposure risk of trees to beetle attacks. In many studies, it has been stated that trees wounded due to harvesting activities or different reasons affect the beetle damage in the forest. For these reasons, it is important to determine the numbers and densities of wounds which are the focal points of infection for bark insects in the residual stand as a result of harvesting.

In this study, it is aimed to assessment the damaging in the residual stand as a result of harvesting and the possible measures that can be taken.

# 2. MATERIAL AND METHODS

This study was carried out within the administrative boundaries of the Kastamonu Regional Directorate of Forestry, Daday Forest Sub-District Directorate (Figure 2). Two adjacent sections where harvesting activities are carried out were used. Harvesting sections are Karaçam stand and the distribution of stand types are as Çkcd3, ÇkMbc2, Çkbc4, Çkc3, Çkcd3-1, Çkcd3-2, Çkd2. Average elevation of sections is 1210 m, average slope is 38%. The aspect of the sections is northwest. The total area of the harvesting sections is 14.9 ha and 1341 trees are stamped by the relevant operating unit and an eta in the volume of 1058,743 m3 was taken in Planted Barked Trunk volume. Harvesting was made by log method, which we can express as motor-manual. Removing from the section, in other words, skidding activities are made by agricultural tractors and with both ends on the ground.





#### Figure 2. Study area

To determine the damage on the residual stand, full area of sections where harvesting is made was evaluated. All damages caused on the saplings in the residual stand, overturn of trees, trunk breakage, top breakage and root wounded are not taken into consideration due to not exceeding more than a couple cases in the sections where the study was carried out. Observations were made after the harvesting activities are ended, measurements were made on the individual of the residual stand that are larger than 8 cm diameter.

Wounds were evaluated according to their severity and height on the tree. Wounds caused on the individual of the residual stand were divided into five different Wound Severity Class by taking into consideration the areas of wounds (Table 1) (Meng, 1978, Ünver, 2008).

| ound Severity Classification | Wounded size (cm <sup>2</sup> ) |  |
|------------------------------|---------------------------------|--|
| I. Class                     | < 10                            |  |
| II. Class                    | 11-50                           |  |
| III. Class                   | 51-200                          |  |
| IV. Class                    | 201-500                         |  |
| V. Class                     | >500                            |  |

Table 1. Wound Severity Classification (Meng, 1978, Ünver, 2008)

The height of wounds on the trees are evaluated in three groups as bottom, middle and top, according to the height from the ground they are found on the trunk (Table 2) (Youngblood, 1990; Ünver, 2008).

Table 2. Grouping of Wound Locations According to Their Height

| Wound Location | Height of Wound Location (cm) |
|----------------|-------------------------------|
| Bottom         | 0-35                          |
| Middle         | 36-137                        |
| Тор            | >137                          |

The diameter d1,30 of the trees that were wounded during the harvesting were measured with diameter gauge, the size (width - length) of the wounds caused on the trees were measured with steel tape meter. The aspect of the wound on the tree was identified by a compass. The area of the wound was calculated by multiplication of the width and length of the wound

# 3. RESULTS AND DISCUSSION

#### Results

As a result of harvesting 2412 pieces of 410,745 m3 and 646 ster wood were obtained. Wounds were identified in a total of 33 trees in the residual stand in the harvesting area (Figure 3)



Figure 3. Wounded tree

Table 3 shows the distribution of 33 wounded trees according to their diameter stages. When the table is examined, it is seen that the wounds occurred in the trees with a diameter of 36-43.9 cm, with the highest ratio of 30.4%.

| Diameter class (cm) | Ν  | %      |
|---------------------|----|--------|
| 12-15,9             | 1  | 3,03   |
| 16-19,9             | 6  | 18,2   |
| 20-23,9             | 2  | 6,1    |
| 24-27,9             | 3  | 9,1    |
| 28-31,9             | 5  | 15,2   |
| 32-35,9             | 3  | 9,1    |
| 36-39-9             | 5  | 15,2   |
| 40-43,4             | 5  | 15,2   |
| 44-47,9             | 1  | 3,0    |
| 52-55,9             | 1  | 3,0    |
| 56-59,9             | 1  | 3,0    |
| Total               | 33 | 100,00 |

Table 3. Distribution of Wounded Trees According to Diameter Stages

The aspect of the wounds occurred on the tree trunks are 49% south, 21% north, 24% east and 6% west. The difference in the distribution of wounds by aspect and focusing on some aspect shows that the removal from the section was made by means of specific skidding paths.

Table 4. Distribution of Wounded Trees in the Residual Stand According to Wound Severity Classes

| Wound Severi | ty Classes | Ν  | %    |
|--------------|------------|----|------|
| I.           | Sınıf      | -  | -    |
| II.          | Sınıf      | -  | -    |
| III.         | Sınıf      | 6  | 18,2 |
| IV.          | Sınıf      | 8  | 24,2 |
| <b>V.</b>    | Sınıf      | 19 | 57,6 |
| Tota         | 1          | 33 | 100  |

When the distribution of wounds is looked at, it is seen that they are distributed between 56-3200 cm2 and correspond to Class V wound severity with an average of 777,70 cm2 (Table 4).

Table 5. Distribution of Wounded Trees in the Residual Stand According to Wound Height Classes

| Height Classes | Ν  | %    |
|----------------|----|------|
| Bottom         | 25 | 75,8 |
| Middle         | 7  | 21,2 |
| Тор            | 1  | 3,0  |
| Total          | 33 | 100  |

When the height of wounds from the ground is looked at, it is seen that they occur up to 150 cm height from the bottom, and its average corresponds to 30.45 cm height at the bottom part (Table 5).

#### Discussion

The average of wound height is seen to be the same with the wound height average of the removal activity previously performed. Ünver (2008) stated in his study that the surroundings of the trees cut in the harvesting area and the trees found on the skidding route were observed. However, in our study, all harvesting area is being evaluated and similar results were obtained in terms of wound heights. The reason for this can be stated as the study areas being in similar slope groups and use of similar removal technique. Therefore, it is seen that harvesting activities, especially the increase of mechanization level during the removal stage, are not sufficient and also the workers are required to pay attention. Also, instead of directly employing the public of the region, trained and specialist people should be employed.

When the wound severity is evaluated, while Ünver (2008) stated that Class III wound severity was the most, our study shows Class V wound severity as the most. The reason for this may be the difference in the distribution of trees in the stand according to their diameter classes. In this study, the most are 36-44 cm diameter groups with 30.4%.

The degree of slope in the study area being high shows that the control of especially the large assets during skidding is difficult and progressed by hitting the residual individuals. For this, one of the workers should accompany the product during skidding and intervene in a controlled way when required with simple hand tools such as sapin.

The damages occurred on the residual stand during harvesting sends an invitation to primarily bark beetle and many other environmental damages. In the studies conducted, it was identified that *Dendroctonus micans*, a species of bark beetle, has damaged 78% (Eroğlu, 1995), 88% (Özcan et al., 2006) 84.4% (Alkan Akıncı, 2006, Alkan Akıncı et al., 2009), 70.73% (Özcan, 2009; Özcan et.al., 2011) of the wounded trees. Some bark beetle are also known to be damaging in the forests of the region (Akkuzu and Güzel., 2015; Ozcan, 2017; Tuncay et. al., 2018). This study, forming the results of first observation, constitutes the part of a project to present the relations of wound-beetle as a result of annual observation after harvesting.

#### REFERENCES

- Acar H H (2004). Ormancılıkta Transport Ders Notları, Karadeniz Teknik Üniversitesi Orman Fakültesi Orman Mühendisliği Bölümü, 268s, Trabzon,.
- Acar H H & Şentürk N (1996). Dağlık Orman Alanlarındaki Üretim Çalışmalarında Mekanizasyon. İstanbul Üniversitesi Orman Fakültesi Dergisi, İstanbul, B, 46,1-2-3-4: 76-94.
- Alkan Akıncı H (2006). Doğu Ladini Ormanlarında *Dendroctonus micans* (Kugelann)'ın Populasyon Dinamiğine Etki Eden Etmenler ve *Ips typographus* (Linnaeus) ile Diğer Kabuk Böceği Türleri (Coleoptera, Scolytidae)'nin Populasyon Düzeyleri ve Etkileşimleri [Factors Affecting Population Dynamics of *Dendroctonus micans* (Kugelann) and Population Levels and Interactions of *Ips typographus* (Linnaeus) and Other Bark Beetle Species (Coleoptera, Scolytidae) In Oriental Spruce Forests. PhD thesis ]. Karadeniz Teknik Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, Trabzon.
- Akinci H, Ozcan G & Eroglu M (2009). Impacts of Site Effects on Losses of Oriental Spruce During *Dendroctonus micans* (Kug.) Outbreaks in Turkey. African Journal of Biotechnology, 8(16): 3934-3939
- Akkuzu E & Guzel H (2015). Edge Effects of Black Pine Forests on *Ips sexdentatus* (Boern.) Abundance. Šumarski list, 9 (10): 447–453.
- Buğday E (2011). Ormancılık Üretim Çalışmalarının Çevresel Zararları [Environmental Damages of Forest Harvesting Opreations. MS thesis]. Çankırı Karatekin Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi, Çankırı
- DeGomez T & Celaya B (2013). The Piñon Ips Bark Beetle. College of Agriculture and Life Science. The University of Arizona Cooperative Extension. 5p.
- Eker, M., 2004. Ormancılıkta Odun Hammaddesi Üretiminde Yıllık Operasyonel Planlama Modelinin Geliştirilmesi [Development of annual operational planning model for timber harvesting. PhD thesis]. Karadeniz Teknik Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, Trabzon.

- Enez K (2008). Ormancılıkta Üretim İşçiliğinde Antropometrik Verilerin ve Çalışma Duruşlarının Kaza Risk Faktörleri Olarak Değerlendirilmesi [Assessment of Anthropometric Data and Working Posture as Accident Risk Factors in Forest Harvesting Workmanship PhD thesis]. Karadeniz Teknik Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, Trabzon.
- Erdaş O, Acar H H & Eker M (2014). Orman Ürünleri Transport Teknikleri, Karadeniz Teknik Üniversitesi 232/39, ISBN:978-975-6983-75-1, 504s., Trabzon.
- Eroğlu M, (1995). *Dendroctonus micans* (Kug.) (Coleoptra, Scolytidae)'ın Populasyon Dinamiğine Etki Eden Faktörler Üzerine Araştırmalar. I. Ulusal Karadeniz Ormancılık Kongresi, 23–25 Ekim 1995, Trabzon, Bildiriler Kitabı.
- Gürtan H (1975). Dağlık ve Sarp Arazili Ormanlarda Kesim ve Bölmeden Çıkarma İşlerinde Uğranılan Kayıpların Saptanması ve Bu İşlemlerin Rasyonalizasyonu Üzerine Araştırmalar. Tübitak Yayınları, 250, 95 s. TOAG Seri No:38, Ankara.
- Karaman A (1997). Doğu Karadeniz Yöresinde Farklı Çalışma Koşullarında Kesim ve Sürütme İşlerinde İşgüçlüğü Kriterlerinin Araştırılması ve Verim Üzerine Etkisinin Belirlenmesi [Determining the Effects of Working Difficulties on the Production on the Study on These Factors Relating the Logging Operations on the Working Condiction in the East Karadeniz Forest Region. PhD thesis]. Karadeniz Teknik Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, Trabzon.
- Lempérière G (1994). Ecology of The Great European Spruce Bark Beetle Dendroctonus micans (Kug.). Ecologie, 25 (1): 31-38.
- Meng W (1978). Baumverletzungen Durch Transportvorgange Bei Der Holzernte–AusbabUnd Verteilung, Folgeschaden Am Holz und Versuch Ihrer Bevertung, Schriftenreihe Der LFV Baden-Württemberg, Band 53, 159 s.
- Neely D (1988). Tree Wound Closure. Journal of Aboriculture, 14 (6): 148-152.
- Özcan G E, Eroğlu M & Alkan Akıncı H (2006). Doğu Ladini Ormanlarında *Dendroctonus micans* (Kugelann) (Coleoptera: Scolytidae)'ın Zarar Durumu ve *Rhizophagus grandis* (Gyllenhal) (Coleoptera: Rhizophagidae)'ın Zararlının Populasyonuna Etkisi. Türk. Entomol. Derg., 30 (1):11-22.
- Özcan G E (2009). Maçka Orman İşletmesi Doğu ladini Ormanlarında Başlıca Kabuk Böceklerinin Savaş Olanaklarının Araştırılması [Investigation of the Possibilities of Pest Management of Major Bark Beetle Species in the Oriental Spruce Forests of Maçka Forestry Enterprise PhD thesis ]. Karadeniz Teknik Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, Trabzon.
- Özcan G E, Eroğlu M & Alkan Akıncı H (2011). Use of Pheromone-Baited Trapsf or Monitoring *Ips sexdentatus* (Boerner) (Coleoptera: Curculionidae) in Oriental Spruce Stands. African Journal of Biotechnology, 10 (72): 16351-16360.
- Özcan G E, 2017. Assessment of *Ips sexdentatus* Population Considering the Capture in Pheromone Traps and Their Damages Under Non-Epidemic Conditions. Šumarski list, 141: 47-55
- Topalak, Ö., 1998. Torul Orman İşletme Müdürlüğü Alacadağ Orman İşletme Şefliği Üretim Sırasında Mekanizasyon İhtiyacının Belirlenmesi [Torul Directorate of Forest District, Alacadağ Forest District, Definition Level of Mechanization During The Production Operation. PhD thesis]. Karadeniz Teknik Üniversitesi Fen Bilimleri Enstitüsü, Yüksek Lisans Tezi, Trabzon.
- Tuncay C, Tuncay S & Sevik H (2018). *Ips acuminatus, Ips sexdendatus* and *Pityoctenis curvidens* Damage in Kastamonu Karadere Forest Enterprise. International Journal of Trend in Research and Development, 5 (4): 61-64
- Türker M F (2000). Orman İşletmeciliği Ders Notu. Karadeniz Teknik Üniversitesi, Orman Fakültesi Ders Notları No:59, 226 s.Trabzon.
- Ünver S (2008). Endüstriyel Odun Hammaddesinin İnsan Gücüyle Sürütülmesi Sırasında Ortaya Çıkan Ürün Kayıpları ile Çevresel Zararların Belirlenmesi Üzerine Araştırma [Determining Environmental and Quantitative Effects of Human Power Based Ground Skidding and The Damage Prediction Model. PhD thesis]. Karadeniz Teknik Üniversitesi Fen Bilimleri Enstitüsü, Doktora Tezi, Trabzon.
- Youngblood A P (1990). Effects of Shelterwood Removal Methods on Established Regeneration in an Alaska White Spruce Stand. Can. J. Res., 20: 1378-1381.



# Investigation of the Flora and Stand Characteristics in the Transportation Zone from the Terrestrial to Coastal Districts in Bartin Region

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**Abstract:** The region of Bartin is home to important plant species belonging to different phytogeographic regions in terms of plant sociology due to the diversity of ecological conditions. In addition to this, pure and mixed secondary forest organizations, which are generally covered by Castanetum and Fagetum transition zones, consist of broad-leaved species. However, there are also pure coniferous stand organizations in the form of industrial plantations, which are established by artificial means in the transition zones starting from the terrestrial region of the Bartin region. This study carried out in Ulugecit district, which is in the Bartin-Amasra route and which represents the transition zone in the region in the best way, examines the distribution of taxa to the phytogeographical regions. There are 114 taxa in European-Siberian phytogeographic region, 19 taxa in Mediterranean region, 3 taxa in Iran-Turan region and 111 taxa in wide geographical variations. As a result of vegetation studies conducted in the transition zone, 92 species belonging to 41 families, 24 subspecies and 7 varieties were determined. The rate of endemism in the area is 5,64%. Generally, the ratio of broadleaved mixed stands is 81% and the ratio of pure stands is 19%. The only type of industrial plantation used is the *Pinus pinaster* Aiton. The quarries are single layer and the same aged and the closeness varies between 0.6-0.8 and the frequency is between 0.5-0.7. 65.4% of the stands in the research area are productive and 34.6% degraded.

Keywords: Flora, Stand, Terrestial, Coastal, Vegetation, Analysis, Bartın.

# **1. INTRODUCTION**

Turkey's geographical characteristics of different flora and fauna that direction, depending on the impact of emerging with different ecological conditions because has a rich biological potential. According to the latest statistical information, approximately 13000 plant species are found in our country and approximately 3000 of these total species are endemic species and our country's endemiz rate is close to 38%. This rate is 14.9% in Greece, 2.9% in France and 18.6% in Spain (AVCI, 2005). On the other hand as a joint result of different ecological effects in Turkey Euro-Siberian, Mediterranean and Irano-Turanian the flora of Turkey emerged by grouping the different number and types of flora field has the cardinal region should carefully protected and operate due to the high level of biodiversity (KILINÇ, 2011). For this reason, the transition zones or the areas where bridging areas, especially the main zone areas, should be subjected to floristic analyzes and examined. Only in this way the biodiversity policies and strategies whose objectives and objectives are specific will be produced in a healthy way. In this context, our country, which is in the process of entering the European Union, has to show the utmost importance for the conservation and sustainable management of biological diversity according to the international agreements signed by our country (ÇÖRTOĞLU, 2009).

In this study, in this study carried out in the Ulugecit-Ambarci region, which is located at the transition point from the terrestrial to coastal region, it is tried to reveal the vegetation and forest structure in this transition zone.

# 2. MATERIAL AND METHODS

The Ulugecit-Ambarci region where the research was conducted is located in the northeast of the Bartin province and is located at the crossroads of the mountains from the terrestrial and coastal areas. The average elevation of the research area is 600m as a result of the elevation measurements, and the overall characteristic is composed of north and northeast directions. Plant picking areas and forest stand dynamics were carried out under medium slope conditions. However, the area is sandy-clay and sandy-clayey-slime texture and has a clastic structure. Soils generally have moderate deep soil conditions, pH is 7.8 and organic matter is high. However, the average temperature in the research area is 17.2°C, the

average rainfall is 987.6 mm, the prevailing wind direction is north and the vegetation period is 6 months (ANONİM, 2010).

The samples of the plants in the research area and having different life forms were taken in accordance with the general vegetative sampling criteria and after being pressed, the necessary comparisons were made by bringing them to Bartin University, Faculty of Forest Herbarium in order to carry out the diagnoses. On the other hand, in order to determine the stand dynamics of the forest form at the crossing point where the research was conducted, transient experimental areas were taken according to the random sampling method of 25x40m dimensions. In this experimental area, various determinations have been made in terms of species or species dominating the stand, closeness, frequency, stratification and growth stage.

# 3. RESULTS AND DISCUSSION

## Results

As a result of the investigations and evaluations carried out in the research area, the transition from the terrestrial to the coastal region is dominated by the oriental beech (*Fagus orientalis Lipsky*.) And the hornbeam (*Carpinus betulus L*.) in the Fagetum zone in general. However, *Prunus laurocerasus* L., which is involved in the individual mixed forests in the leafy mixed forests, *Ficus carica subsp. Carica L., Morus nigra L., Morus alba L., Fraxinus angustifolia L., Cornus mas L., Quercus robur L., Quercus hartwissiana var. macrocarpa L., Salix alba L., Populus nigra L., Junglas regia L., Tilia tomentosa Moench. and Acer campestre L. (Figure 1).* 



Figure 1. Forests of broadleaves forest tree species in research area

As can be seen in Figure 1, stand dynamics of forest organizations consisting of broad-leaved species in the transition zone in the research area are generally single-stratified with shoot origin, similarity, canopy degree is 0.6-0.8 and density is 0.5-0.7 It is in the form of mixed forest organizations where the individual mixture varies.

As a result of the diagnoses carried out on the plant samples collected from the research area, it was determined that there were a total of 41 family plant species in this transition zone. The distribution of the taxa determined in the research area according to phytogeographic regions is given in Table 1.

| Table 1. Distribu | tion of Taxa Ac | cording to Phyto | ogeographical | Regions |
|-------------------|-----------------|------------------|---------------|---------|
|-------------------|-----------------|------------------|---------------|---------|

| Phytogeographical Area | Number of Taxa | Rate (%) |  |
|------------------------|----------------|----------|--|
| Euro-Siberian          | 114            | 46,3     |  |
| Mediterranean          | 19             | 7,7      |  |
| Irano-Turanian         | 3              | 1,2      |  |
| Wide Distribution      | 111            | 44,8     |  |

When the data in Table 1 are analyzed, it is determined that the phytogeographical region with the largest number of taxonomies in the research area is Euro-Siberian Region and that this region is followed by species with wide distribution area.

#### Discussion

In the area of research, Bartın region is located within the boundaries of the Euro-Siberian phytogeographic region. As a result of the diagnosis of plant samples collected from the area, this was determined by 114 taxa and 46.3% participation rate (Table 1). As a matter of fact, in studies conducted in the field of plant sociology, in the study conducted in many regions close to the research area, the dominance of Euro-Siberian region members is mentioned in the Western Black Sea belt (KILINÇ, 1975; KILINÇ, 2011). In the research area, mixed forests, which are generally composed of broad-leaved species, were determined in the experimental areas where they contain many tree species (Figure 1). These forest areas are generally located in the transition to the *Castanetum-Fagetum* and *Fagetum* and in the main zones. The same age, root and billet shoots in these forests, which density of 0.5-0.7 and the canopy degree of 0.6-0.8 varies between. As a matter of fact, in many studies where general evaluations of Western Black Sea Region forests are made, the dominance of mixed forest zones (ATA, 1995). Within the scope of this research, similar forest structures have been found to be dominant in the field as well from the examinations and evaluations made in the experimental areas. In this context, it is possible to say that the data obtained from the researches made in previous years and the findings obtained within the scope of this research are very similar.

#### 4. CONCLUSION

According to the results obtained from the research, there is an important plant biodiversity in the transition zones. This diversity, which belongs to 41 families, also contains plant elements belonging to many different phytogeographical regions. In this context, the vegetation structure in the region should be examined closely and especially the number of herbaceous species should be preserved over the years. However, these transitional zones should be protected and not allowed to be destroyed in order to ensure the future of plant communities in both land and coastal areas. However, leafy mixed forests, which are determined to be composed of root and billet shoots, should be operated in accordance with their natural structures in terms of preservation of vegetative biodiversity and radical silvicultural processes aimed at changing the natural structure, especially the establishment of industrial plantations and bond works in the transition zones should be avoided.

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#### REFERENCES

Anonim (2010). Bartın Orman İşletme Şefliği Amenajman Planı [Management Plan of Bartın Forest Range District]. Bartın.

- Ata, C. (1995). Silvikültür Tekniği, Z.K.Ü Bartın Orman Fakültesi, Üniversite Yayın No: 4, Fakülte Yayın No: 3, Bartın, 453 s.
- Avcı, M. (2005). Çeşitlilik ve Endemizm Açısından Türkiye'nin Bitki Örtüsü [Diversity and Endemism in Turkey's Vegetation]. İstanbul Üniversitesi Edebiyat Fakültesi Coğrafya Bölümü Coğrafya Dergisi, 13:27-55.
- Çörtoğlu, F.S. (2009). Avrupalılaşma Bağlamında Türkiye'de Doğa Koruma ve Biyolojik Çeşitlilik [Europeanization in Turkey in the Context of Nature Conservation and Biodiversity], Ankara Üniversitesi Yayınları, No:235, Ankara Üniversitesi Avrupa Toplulukları Araştırma ve Uygulama Merkezi Araştırma Dizisi No:34, ISBN: 978-975-482-837-5, Ankara.
- Kılınç, M. (1975). İç Anadolu-Bati Karadeniz Geçiş Bölgesinde Devrez Çayı İle Kızılırmak Nehri Arasında Kalan Bölgenin Vejetasyonu [The Vegetation of the Region Between the Devrez Stream and the Kızılırmak River in the Central Anatolian-Western Black Sea Transition Region], Doğa Bilimleri Dergisi, 2(9): 315-337.

Kılınç, M. (2011). Bitki Sosyolojisi [Plant Sociology], Palme Yayıncılık, 2. Baskı, (ISBN:978-975-8982-28-8), Ankara.



# The Role of Active Microorganisms to the Seed Germination in Calabrian Pine

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Abstract: Calabrian pine (*Pinus brutia* Ten.) is commonly found in ecosystems of the Eastern Mediterranean, Aegean, Marmara region and along Black Sea coastal area as small population. Turkey's geography, consist of the about 5.5 million hectares of Calabrian pine forest. Since seed is the most important element in the regeneration of forests, activating seed viability and germination has been the subject of many studies in recent years. One of the materials of these researches is the active microorganisms (EM). EM has been used for many applications such as agriculture, livestock, horticulture and landscape. The EM application was developed by Professor Teuro Higa in 1970's. The aim of this study was to determine the effect of EM on the seed germinations of Calabrian pine. In the study, four different EM type (EM-A, EM-5, EM-Gold and EM-FPE) and three different concentrations (5%, 15%, 25%) solutions were tried. Before sowing seeds were treated the 8 hour with the water. Each consisting of 20 seeds total 60 seed of the species were placed in petri dishes. About 5 ml EM solutions was added on the seeds. For the control treatments were given only water. It was allowed to germinate in the air-conditioning cabinet with a moisture content of 45% at 24 degrees. Germinated seeds were recorded daily during the experiment. As a result, some EM ratios were found to be more effective than the control group seeds.

Keywords: Seed, effective microorganisms, germinations, Pinus brutia, Turkey







# **ORAL PRESENTATION**

# Functional Uses of Hemp (Cannabis sativa L.)

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Abstract: Hemp, which has been cultivated for a long time by mankind, is a C3 plant which has 2n = 20 chromosome, annual, strong and long fibres. It is grown for its fibers that are strong and long and its seeds. The plant is considered to be native of Western and Central Asia (Russia, China, India, Pakistan, and Iran). It is known that hemp has been cultivated in Anatolia since M.Ö 1500. However, the cultivation area of hemp has been decreased in Turkey, recently. Moreover, Kastamonu, in Turkey, had come to the forefront in hemp growing. Particularly, Taşköprü province in Kastamonu had intensively produced industrial hemp. Furthermore, the Kastamonu climate meets the ecological requirements of Cannabis such as moisture, temperature, precipitation. Unfortunately, hemp has not been produced in Kastamonu for the last decades due to the use in sedative and narcotic drugs and also lowing economic prospects. In addition to this, another reason for the decrease in hemp production is that synthetic fibers become widespread. Cannabis is classified into the family Cannabaceae and has three main types: C. sativa, C. indica, and C. ruderalis. Beside this, there are of a large number of subspecies too for Cannabis. Hemp which is grown for the industrial purpose is C. sativa L. spp. vulgaris. Generally, C. sativa L. spp. indica is used for medical and narcotic aim because of containing THC (Tetrahydrocannabinol). C. sativa L. spp. ruderalis, called wild hemp, grows naturally in nature. The aim of this review, to reveal multi-purpose properties of hemp and to determine to possibilities the develop of hemp in agroforestry. Hemp has lots of functional usage areas, because of its unique properties. It is cultivated to produce a vast variety of products such as hemp seeds, hemp oil, clothing, rope, paper, insulation, cosmetics, biodegradable plastics, construction material, resin, pulp, animal bedding and feeding, fuel, drug. If used as fuel, the calorie value is half the amount of coal. Likewise, it might be using an alternative usage of hemp in agroforestry too. These are carbon dioxide absorption and more oxygen production. For instance, carbon dioxide absorption of hemp is fourfold higher than carbon dioxide absorption which has attain in agroforestry practices. Firewood production is also important for agricultural forestry. Likewise, 6-7 tones firewood is obtained from 1-hectare hemp field. Besides its use for the alkaloid industry is also very important due to its medical properties. In terms of medical use, it is used to treat many diseases ranging from anxiety to cancer diseases. Briefly, thanks to multipurpose uses, hemp produce areas should be expanded.

Keywords: Agroforestry, Cannabis, Hemp, Industrial, Medical

# **1. INTRODUCTION**

*Cannabis* is a member of the Cannabaceae family. Cannabis is a member of the Cannabaceae family. the best important types of Cannabaceae family are *Cannabis sativa, Cannabis indica* and *Cannabis ruderalis*. *C.sativa* and *C. indica* become prominent among these types. Typical features of *C.sativa* (called hemp) are which have pointed leaves, thin flowers and long.In spite of this, typical features of *C.indica* (called Marijuana) are which have large leaflet, close branches. Also C.indica has a dark green colour. These species tend to be shorter and bushier, producing fuller, denser flower buds (URL, 2019). Generally, whereas marijuana is prefered for medicinal purpose, hemp is preferred functional usages from nutritional to textiles (CRS REPORT, 2018).

The plant is thought to come from Western and Central Asia (Anwar *et.al.*, 2006). Hemp is of economic and pharmaceutical importance throughout the world. Pharmaceutical importance is stem from its 's THC ratio. THC ratio has to low for industrial purpose production legally. This ratio is 0.3% and 0.2% for ABD (Oomah, 2002).

Another product obtained from cannabis is essential oils. These oils include seconder compounds such as monoterpenes, sesquiterpenes, and other terpenoid-like compounds. Beside, essential oils of hemp are used cosmetics and aromatherapy (Turner *et.al.*, 1980). When evaluated as food, it points out nutritional value especially vitamins, minerals and b-carotene.

Hemp seed involves 25–35% oil, 20–30% carbohydrates, 20-25% protein, 10–15% insoluble fiber (Deferne *et.al.*, 1996). Another notable feature of hemp is which have all-purpose. So many different areas offer the possibility to use. All parts of hemp from fibre to shive can be used different areas. The aim of this review, to reveal multi-purpose properties of hemp.

## 2. INDUSTRIAL CANNABIS (HEMP) USE AREAS

Hemp naturally contains THC substance in its structure. Industrial Hemp naturally contains in small quantities THC substance in its structure. These quantities quite low in comparison with medical hemps. Also THC ratio in plants change based on a lot factors. Hemp is very sensitive temperature and other environmental conditions. Therefore, it shows different growth under different climatic conditions. One of the environmental conditions is day length. In a study conducted in Mexico, it was observed that day length-dependent illumination duration on cannabis seedlings affected THC ratio (Gönen, 2009). Generally, low THC ratio is significant for industrial production. It is a unique plant where every part of the *Cannabis* plant can be evaluated from seed to shivs. From this point of view, it can be used in many different fields in the industry.

#### **Usage for Fibres**

Hemp fibres have some of the best mechanical properties of all natural fibres. Commonly, they are applied for biocomposites in automotive sector. They are mainly used for insulation material and for bio-composites in automotive applications. Before the 1990s, fibres of industrial hemp had largely used in specialty paper and pulp. However, hemp pulp was very high price and consequently hemp pulp usage was restricted for only cigarette and bank notes. Today, besides the production of twine, rope and sack, it has a lot of usage in interior equipment applications in the automobile industry (Merdan *et al.*, 2013).

Bast fibers are generally used for textiles (including carpets and garments) and for industrial uses such as geotextiles, erosion control blankets and composite reinforcements and fillers. The high strength and economic properties of the hemp fiber make it possible to use a number of molded composites for glass pieces and synthetics after they are put in place: car parts, building materials as well as consumer products. The economic reality of cannabis is that the price of cannabis cannot compete with waste products (wood, straw, stove, etc.). It is technically possible to produce products such as biofuel or medium density chipboard, but there are difficulties in providing a competitive cost (ORAN,2019).

#### **Usage for Food**

Industrial hemp seeds have quite high nutritional content and they have rich fatty acid content too. Therefore, they have been consuming as food for long times by humans and animals. Hemp seeds are defined as a traditional food in the world (Carus, 2017). Moreover, hemp seed is a quite good herbal nutritional source in terms of has a protein source and also it contains omega-3 and omega-6 fatty acids and linoleic acid, which are important for health. Beside this it is source of vitamin E (Koçak, 2019). Industrial cannabis seeds can be consumed in our country by mixing with wheat seeds (called *Çetene*) or consumed by frying without mixing *Cannabis sativa* seed is also used as a food in bird feeding. The balanced amino acids, vitamins and fatty acids in the seeds allow birds to be fed more healthily. Thus, there is no deficiency of vitamin E in birds fed hemp seeds (Aytaç *et al.*, 2018). So, it can be say that *Cannabis sativa* seed is quite important for human and animal diet. Also it can be offered use as food supplement too.

#### **Usage for Shivs**

Shives that is one of evaluable part of hemp have a steady market too for trade. This market is mostly based on the use of hemp shivs as animal bedding materials for pets and horses. According to a hemp fibre producer's, hemp shivs production is very important economically. 1 kg hemp fiber consist of by product 1.7 kg of hemp shivs. In this context shivs obtain from hemp as waste product very important for hemp functional using. Moreover, they have a property that can absorb moisture up to four times their dry weight. After bedding rots, they turn into an excellent compost. Commonly, they use for horse bedding (Carus *et.al.*,2013).

#### **Usage for Medical**

The use of medical purpose of cannabis began in the mid-19th century (Kalant, 2001). It was described that cannabis extractions were effective especially on some diseases (O'Shaughnessy, 1839). And then different studies on effectiveness of hemp showed that it can be treated such as sleeplessness, pain, anxiety diseases (Kalant, 1972).

*Cannabis indica* is preferred than *Cannabis sativa* for medicine industry. Because percentage of THC ( $\Delta$ 9-tetrahydrocannabinol) much more in *Cannabis indica*. So, it is suitable for medical aim. *C.indica* involves chemicals (more than 460). More than the 60 of these are cannabinoids (Ben Amar, 2006). Some of cannabinoids find in the plant naturally. THC can be given as an example for this situation. Another natural cannabinoid is cannabidiol, which has some of the physiological actions of THC but does not produce the high that comes from smoking. Other cannabinoids are synthetic (i.e., made in a laboratory); these are functionally similar to THC. Some of these (e.g., Spice, K2) have been used recreationally and other synthetic cannabinoids, including dronabinol and nabilone are used therapeutically (Kalant & Porath-Waller, 2016).

Cannabinoids are considered effective in patients suffering from somatic diseases such as chronic pain (related to e.g. cancer or in terminally-ill patients), respiratory system disorders (asthma), eye disorders (glaucoma) or multiple sclerosis. Clinically, the most convincing results of using cannabinoids are found in providing relief for patients suffering from high muscle tone, adverse effects of chemotherapy (e.g. nausea) or support in AIDS treatment (Jablonski, 2015).

#### **Other Usage**

In addition to the traditional use of cannabis as medicine and food, it has different functional uses as a result of many alternatives it provides. It provides erosion control because of its deep roots (Amaducci *et al.*, 2008). Furthermore, hemp cultivation is 400% more efficient at  $CO_2$  absorption than agro-forestry per land use. Its rapid growth rate means it can provide the industrial quantities of biomass required in our modern society (Aytaç *et. al.*, 2018).

Hemp is a plant with a high oil content, so it can be used as a biodiesel source. But hemp as a biodiesel source cannot compete with other oil plants in Turkey (Gizlenci et al., 2019). Also hemp cultivation is 400% more efficient at CO<sub>2</sub> absorption than agro-forestry per land use. Its rapid growth rate means it can provide the industrial quantities of biomass required in our modern society (Aytaç *et. al.*, 2018).

#### 4. CONCLUSION

Hemp has become a popular plant for the last few years. Cannabis sativa take an important place in hemp species with different usage possibilities in industrial area. The use of industrial seed as a food is common in the world. But the consume of industrial hemp seeds as a food are not widespread in Turkey because of different reasons. There have been new use possibilities for insulation material and bio-composites recently. After *C. sativa* plant was separated from fiber, remaining part of plant can be used as fuel. Also it is an important bio-mass plant and its calorific value is very high. It has pesticidal applications as well. It can say that Modern uses are textiles, paper industry, building materials, personal hygiene (soap, shampoo, balms, lotions etc.). If the plant is evaluated in terms of these features, it can provide value added for different industries.

Marijuana is derived from the hemp plant *Cannabis sativa*. Tetrahydrocannabinol (THC) is the major psychoactive constituent of *Cannabis*, while cannabidiol (CBD) is the major non psychoactive constituent. The *Cannabis* plant contains many biologically active chemicals, including ~60 cannabinoids. THC is the primary psychoactive component found within *Cannabis*, and has been shown to have analgesic effects. Increasing evidence has highlighted numerous roles for other phytocannabinoids, particularly Cannabidiol (CBD), a non-psychoactive component with anti-inflammatory, analgesic, and antipsychotic properties. Other uses are epilepsy-especially in children with dravet syndrome, chronic pain, multiple sclerosis, psychosis or schizophrenia. Medical cannabis is also used for some cancer patients to relieve symptoms including nausea and vomiting loss of appetite, and pain. Briefly, thanks to multipurpose uses, hemp produce areas should be expanded.

#### REFERENCES

Amaducci, S., Zatta A., Raffanin, M., Venturi, G., 2008. Plant and Soil, 313, 1-2: 227-235.

- Anwar, F., Latif, S., Ashraf, M., 2006, Analytical characterization of hemp (*Cannabis sativa*) seed oil from different agro-ecological zones of Pakistan, J11190 in *JAOCS* 83, 323–329.
- Aytaç, S., Ayan, A.K., Arslanoğlu Ş.F., 2018.Suçlu olarak bilinen bitki: Kenevir. Ziraat Bilimlerinde Güncel Akademik Çalışmalar, 445-455.

Ben Amar, M. 2006. Cannabinoids in medicine: A review of their therapeutic potential. Journal of Ethnopharmacology, 105,1-25.

- Carus, M., Karst, S., Kauffmann, A., Hobson, J., Bertucelli S., 2013. The European Hemp Industry: Cultivation, processing and applications for fibres, shivs, seeds and flowers, European Industrial Hemp Association, 1-9.
- CRS REPORT (Congressional Research Service), 2018. Hemp as an agricultural commodity.1-43.
- Deferne, J. L., & Pate, D. W. 1996. Hemp seed oil: a source of valuable essential fattyacids. *Journal of the International Hemp* Association, 3, 4–7.
- Gizlenci, Ş., Acar, M., Yiğen, Ç., Aytaç, S., 2019. Kenevir tarımı. Karadeniz Tarımsal Araştırma Enstitüsü Müdürlüğü, Samsun
- Jablonski P., 2015. Medyczne Używanie Konopi [Medical use of Cannabis]. Serwis Informacyjny Narkomania; 3(71) (on Polish).
- URL, 2019. Kenevirturk: Industrial hemp, Retrieved in April 12, 2019 from www.kenevirturk.com.
- Kalant, H., Porath-Waller, A.J., 2016. Medical use of Cannabis and cannabinoids, Canadian Centre on Substance Abuse, 5, 1-20.
- Kalant, O.J. 1972. Report of the Indian Hemp Drugs Commission, 1893–94: A critical review. *International Journal of the Addictions*, 7, 77–96.
- Kalant, H. 2001. Medicinal use of cannabis: History and current status. Pain Research & Management, 6, 80-91.
- Koçak, Y., 2019. Sanayicinin yeni cevheri... Endüstriyel kenevir, Kenevir Çalıştayı, Kastamonu.
- Merdan, N., Koçak, D., Acar, K., 2013. Kenevir liflerinin konvansiyonel ve mikrodalga yöntemine göre maleik anhidrit ile yüzey modifikasyonu
- Oomah, B.D., Bussonb, M., Godfreya, D.V., Drovera, J. C.G. 2002. Characteristics of hemp (Cannabis sativa L.) seed oil, Food Chemistry 76 ; 33–43.
- O'Shaughnessy, W.B.,1839. On the preparations of the Indian Hemp, or Gunjah (Cannabis indica): Their effects on the animal system in health, and their utility in the treatment of tetanus and other convulsive diseases. *Transactions of the Medical and Physical Society of Bengal, 1838–1840*, 421–461.
- Orta Anadolu Kalkınma Ajansı (ORAN), 2019. Kenevir yetiştiriciliği.1-34.
- Turner E., C. M. A. Elsohly, E. G. Boeren 1980. Constituents of Cannabis sativa L. XVII. A review of the natural constituents. Journal of Natural Products 43 (2): 169-243



# Some Characteristics of Bare Rooted Seedlings in Different Eastern Beech Populations (*Fagus* orientalis Lipsky.) of Turkey

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Abstract: The nurseries' ecological conditions where the saplings are cultivated can affect the characteristics of the saplings and therefore their development. Furthermore, the genetic characteristics of the populations, where seeds are collected for saplings production and cultivation conditions of these stands can affect the development of saplings. In this study, the seeds obtained from different locations of the natural spreading area of Eastern beech were cultivated in the same ecological conditions; the genetic characteristics of the populations and effects of the characteristics of the cultivation environment of these populations on the morphological and physiological characteristics of the saplings have been investigated. For this study, seeds were obtained from Bursa-İnegöl, Balıkesir-Dursunbey, Sakarya-Akyazı, Kastamonu-Çatalzeytin, Zonguldak-Devrek-Tefen, Zonguldak-Devrek-Akçasu, and Bartın-Yenihan populations, and they were grown in the Gökçebey State Forest Nursery of Zonguldak Forest Regional Directorate. The physiological and morphological characteristics of 2+0 bare root saplings have been measured at the end of vegetation season. The morphological characteristics were as follows: height, root-collar diameter, number of branches, branch thickness, sapling shoot and root fresh weights, total sapling weight, sapling shoot and root dry-weights, total seedling dry-weight, shootroot ratio, sapling height to root collar diameter ratio, and Dickson quality index. The physiological characteristics were as follows: chlorophyll a, chlorophyll b, total chlorophyll, relative humidity, and transpiration rate. As a result; it has been determined that the population difference is a significant variation on all morphological characters except for shoot-root ratio and seedling height to root collar diameter ratios. In addition, significant differences have been found among populations on chlorophyll b and sapling relative ratio. In terms of transpiration, Bursa-İnegöl population (243.59) has shown the highest value; whereas, Bartin-Yenihan population (231.75) has shown the lowest value.

Key Words: Eastern beech, quality, seedling, population, nursery, Turkey



# An Instrument to Promote Sustainable Forest Management: Forest Certification

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Abstract: Forest certification is an institutional innovation created to implement sustainable forest management that emerged in the early 1990s. The concept of certification, which initially emerged from the influence of environmental groups, has soon become a potential tool for promoting sustainable forest management. Forest certification aims promoting sustainable forest management with specific criteria and indicators. These criteria and indicators are developed for the national level to describe and monitor status and trends in forest and forest management. Until today, 124 million ha of forest, corresponding to %3,2 of world forest, have been certified by different certification bodies established in last decade. After ten years of implementation, it was observed that the aim of conservation of biodiversity has not been achieved in tropical forests, which is the starting point of the certification concept. Most of certified areas are in the temperate and boreal zone and Europe is the continent with the most of certified forests. Only %10 of certified forest is located in tropical countries. Although the quality of audits of the standarts are varying in quality, there are indications that independent audits are encouraging improvement of forest management. Forest certification has been very successful in raising awareness and disseminating knowledge on a holistic sustainable forest management concept, embracing economic, environmental and social issues worldwide. This study aims to provide the contribution of our country' forestry management certification efforts for the development of applications in Turkey aimed to put forward. Study will provide what are the pros and cons of the certification systems, and what should be done to improve the deficiencies of those certification schemas (FSC, PEFC) which world are using.

Keywords: forest certification, sustainable management, FSC, PEFC, Turkey



# Investigation of Root Morphology and Development Potential of Wood Species in Windthrow Fields

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Abstract: Roots are part of the plant under the soil and they undertake four important tasks for plant development: 1)allows the plant to adhere to the soil, 2)-It takes water and water-soluble substances, 3)- in some plants the root nutrients are stored, 4)-plant hormones and other organic compounds synthesizes. In addition, the roots of plants constitute a significant amount of organic matter and carbon reservoirs, especially in forest ecosystems. In terms of these characteristics, the roots were among the research subjects of many researchers. Tree roots are divided into capillary (<2 mm), fine root (2-5 mm) and coarse root (5-10 mm) and are distinguished by their functional roles. Fine roots support the network of capillary roots. In addition, nutrients and water transmission to shoots, in supporting the plant structure play an active role. The capillary roots which absorb water and nutrients are important structural and functional components of the forest ecosystems between the tree and the soil. Fine roots reflect the above ground biomass. Coarse roots indicate the attachment of the plant to the soil and the durability of the species. The methods of examining the root systems of forest trees vary according to the purpose of the research. This study was carried out in the windthrown forest areas of Kastamonu and Sinop provinces in order to determine the root systems of Scotch pine, Black pine and fir trees exposed to severe wind or hurricane. According to the results, the highest root weight and diameter were noted for the fir growing on the flysch bedrock, while the highest root length was measured for the black pine growing on the schist bedrock. In conclusion, the results have indicated that there are some correlations between root properties (diameter, lenght and weight) of the windthrow trees, and tree species and bedrock types.

Keywords: Windthrow, root morphology, root classification, tree species, bedrock

#### **1. INTRODUCTION**

Windthrows have a large negative impact on the world's natural forests (Brázdil et al., 2004). The gaps in the forest created by the wind are important for the structural dynamics of the forest and greatly affect the tree structure, age and diversity (White, 1979; Pickett and White, 1985; Pontailler et al., 1997; Ulanova, 2000; Linke et al., 2007). However, windthrows reduce the profitability of forest areas managed for timber production with the timber economic value of trees (Savill 1983; Nieuwenhuis and Fitzpatrick 2002) and increased production costs (Quine et al., 1995). In addition, stands exposed to the wind are collected before the optimum administrative period to reduce the risk of winding in general (Gardiner and Quine 2000). Windthrows in a forest in particular has a wide presence in the Black Sea region of Western Turkey. The windthrows are very high and increased the wind speed continues to be one of the most important issues to deal the forest industry. This problem often leads to an increase in freeze-thawing events on the sloping hills, particularly in high altitude areas, that is to say, to limit the formation of soil and to the open winds of the species along the slopes.

To estimate and reduce forest losses due to wind; the characteristics of the tree, the habitat, the climate, the soil and the stability of the tree related to the forest management techniques should be accurately demonstrated. Wind damage in forest stands belongs to the real problems in forest conservation. In general, the tree crown sizes and characteristics (width, length, shape), tree trunk (height, diameter, strength) and tree root system (depth, width, length, root type), which are the parameters of tree growth, have the greatest effect on tree stability (Kônopka, 1978). The most important of these parameters is the morphology of the existing root system (Cucchi et al., 2004; Dupuy et al., 2005). Tree roots play a very important role. Because it contributes to both physical and biochemical aeration.

The reaction of the tree roots to the wind load, especially above the ground, is studied in less detail, and the work is very demanding and complicated (Reubens et al., 2009). The relationship between the architecture of tree root systems and the

biomechanics of tree stability was investigated by Coutts (1983, 1986) and the stability of the relatively mature trees, root architecture, root depth and root-soil plate condition were examined by Ray and Nicoll (1998). In fact, root responses (i) the overall morphology of the root system, which indicates how the roots are distributed along the length and width of the roots separately; (ii) the morphology and absolute magnitude of subsoil plant organs which determine how mechanical forces are transmitted to the root system; (iii) factors such as soil and weather conditions affecting the ability of roots to attach to plants, and (iv) lengths and deviations of dynamic forces applied to leaves and bodies. However, there are significant differences in the details of changes in root systems between species (Reubens et al., 2009). In general, plant root morphology provides an answer to perceived wind load. Plants may be shallow or poorly draining effect of increasing the number and length of root surfaces with well-drained soils or more excavating roots and tree rooting in the soil improving the durability of lateral roots in soil. The influence of wind on the stability of spruce trees in Slovakia was examined by Kodrík and Konôpka. In general, the majority of the studies examined the above-ground parts of the trees and the problem of root systems was less studied. Also, in areas with poorly drained soil characteristics specific problems affecting root growth were studied less. Stokes et al. (1996) stated that the root depth and length of the soil were the dominant factors affecting the tensile strength. Trees with deep and broadly spreading root systems are better kept than only those with superficial roots (Stokes, 2002). The shape and size of a root system is influenced by its immediate environment as well as its specific species (Köstler et al., 1968). Therefore, trees growing on thin, rocky soils encountered on mountain slopes may have different rooting types depending on the species. The morphology of the root system may be different in the same species that grows in a deep soil on flat ground (Köstler et al., 1968). Trees constantly change their morphology in response to changes in wind exposure (Nicoll and Ray, 1996). Wilson (1975) found an increase in both lower trunk and growth ring width in structural roots of *Pinus strobus* L. trees exposed to wind.

The development of tree root architecture is influenced by both tree species and various soil conditions (Toumey, 1929). The initial root systems of all seedlings develop along a single axis and form the pile root to form a broadly branched system of taproots (Coutts, 1989). However, the dominance of taproot in most species, decrease in very early development and begin to form secondary roots instead (Sutton, 1980). For example, after the storm of October 1987, only 2.4% of 4511 overturned trees investigated by the Royal Botanic Gardens in Kew were found to have taproots (Cutler et al., 1990). Another example, oak, pine and fir species have shown a taproot that continues until maturity (Büsgen et al., 1929). Although the root of the tree has important effects on the properties of the soil, there is also the interaction of the roots with the bedrock, which directly facilitates the deepening of the biomechanics of the soil (Fraser and Gardiner, 1967). In addition, the downward movement of the overturned trees (seasonal biogenic shift) (Kirkby, 2004; Wilkinson et al., 2009) contributes to the rock cover and a complete erosion may locally begin (Wilkinson et al., 2009) Results from 0.02 to 1.3 t/ha/year indicate that tree roots should be seen as an important factor affecting sediment transport in the water basin (Schaetzl et al., 1990).

This study was carried out in the windthrown forest areas of Kastamonu and Sinop provinces in order to determine the root systems of Scotch pine, Black pine and fir trees exposed to severe wind or hurricane. Firstly, the roots of the windthrow trees growing on different bedrock were collected and separated as capillary roots (<0.2 cm), fine roots (0.2 cm-0.5 cm) and coarse roots (0.5 cm - 2 cm). Their root length, root weight and root diameters were determined. It was then discussed by comparing the results of statistical analysis with the finding in of other studies.

# 2. MATERIAL AND METHODS

#### Study area

This study was conducted in Kastamonu (Tekçam and Bayam territorial division of Taşköprü Forest Enterprise) and Sinop (Sakız territorial division of Boyabat Forest Enterprise and Suludüz territorial division of Gerze Forest Enterprise) provinces which are located in the middle of Black Sea Region (Figure 1). Detailed information and map coordinates of the study areas are given in Table 1.

The strong wind and hurricanes that occurred in Tekçam study area in 2016 and Bayam in 2013 caused many black pine and Scotch pine trees to fall down and their roots were seen on the ground (Figure 2a, b). Similarly, in 2012 in the Sakız and Suludüz study areas, fir trees were overturned by wind and hurricanes and their roots were up on the ground too.



Figure 1. The location of the study area and satellite images



Figure 2. Field view overturned tree roots of study areas. a) Tekçam, black pine roots b) Suludüz, fir roots c) Bayam, Scotch pine roots d) Sakız, fir roots

The average age of black pine trees in Tekçam region was 65, with an average height of 15 m and the stand was 3 closed (0,71-1,0). The average age of the Scotch pine trees in Bayam region was 76, the average tree height is 16 m and the stand was 3 closed (between 0.71-1.0). The average age of fir trees in the Sakız region was 60, the average length was 19 m and the stand was 3 closed (0.71-1.0). The average age of the fir trees in Suludüz region was 65, the average height was 22 m and the stand was 3 closed (between 0.71-1.0).

#### Table 1. The location of the study areas and some soil properties

| Province /<br>Enterprise                  | District<br>Directorate       | Division<br>number | Coordinate                 | Tree<br>species | Aspect           | Elevation<br>(m) | Slope<br>(%) | Bedrock   | Soil<br>Type             |
|---|-------------------------------|--------------------|----------------------------|-----------------|------------------|------------------|--------------|---|--------------------------|
| Kastamonu,                                | Bayam<br>Forest<br>District   | 82                 | 41°23'02" N<br>34°22'46" E | Scotch pine     | South<br>western | 1200             | 10           | Meta Ultra<br>Alkaline<br>Komatiite                   | Brown<br>Forest<br>Soils |
| Forest<br>Enterprise                      | Tekçam<br>Forest<br>District  | 147                | 41°31'44" N<br>34°21'07" E | Black<br>pine   | South            | 1170             | 15           | Triassic-Lower<br>Jurassic<br>schist                  | Brown<br>Forest<br>Soils |
| Sinop,<br>Boyabat<br>Forest<br>Enterprise | Sakız<br>Forest<br>District   | 98                 | 41°38'51" N<br>34°50'10" E | Fir             | South            | 1300             | 10           | Middle Jurassic<br>Cretaceous<br>neritic limestone    | Brown<br>Forest<br>Soils |
| Sinop,<br>Gerze<br>Forest<br>Enterprise   | Suludüz<br>Forest<br>District | 159                | 41°41'27" N<br>34°58'10" E | Fir             | South            | 1300             | 10           | Upper Mionian<br>clasts and flysch<br>from carbonates | Brown<br>Forest<br>Soils |

#### Root samples collection and analysis

In each study area, 3 sampling areas (20 m x 20 m =  $400 \text{ m}^2$ ) were selected and all the exposed roots of the overturned trees were collected. Root samples were taken in the laboratory and they were classified according to the diameter as; capillary roots (<0.2 cm), thin roots (0.2 cm-0.5 cm) and coarse roots (0.5 cm-2 cm) (Sarıyıldız, 2015). The root diameter was measured in cm with the help of the MAHR mechanical caliper. The wet roots were put into paper bags and stored in the drying cabinet at 65°C for 24 hours and the rood dry weight was determined in grams by weighing in precision balance (Zengin, 2010). In the field, the root lengths of the overturned trees were measured in cm with a ruler.

#### Statistical analysis

One-way analysis of variance (ANOVA) was performed with SPSS (Version 22) program in order to evaluate the effects of bedrock and tree species on tree root length, diameter and weight values obtained from 4 different study areas. According to the ANOVA results, the significance of the difference was determined with the Tukey HSD test.

#### **3. RESULTS AND DISCUSSION**

#### Results

Table 2 shows the average diameter, length and weight of black pine, Scotch pine and fir tree according to root diameter classes. The average root diameter, length and weight of each tree species showed significant differences with respect to root diameter classes (p<0.05). The average root diameter and weight tends to increase from capillary roots to coarse roots in all 3 species (Table 2). The longest roots were capillary roots in all 3 species, the shortest roots were coarse roots in black pine trees and fine roots in Scotch pine and fir trees (Table 2). While there were no significant differences between root diameter classes of tree species, significant differences were determined among species in terms of root lengths and weights. The average capillary and fine root length was highest (respectively, 183 cm and 157 cm) in black pine and the lowest (respectively, 64 cm and 50 cm) value was determined in fir species. In the coarse roots, while the longest roots were Scotch pine (0.73 g), while the highest value was found in blcak pine. The lowest capillary root weight was determined in Scotch pine (0.73 g), while the highest value was found in fir capillary roots (3.82 g). Fine root weight was determined the highest in black pine (4.61 g), the lowest in fir (2.75 g), while the highest coarse root weight was determined in the fir (15.7 g) and the lowest in black pine (3.33 g).

**Table 2.** The average diameter, length and weight of black pine, Scotch pine and fir tree turnings according to root diameter classes

| Trees   | Root Properties            |                    |                               |                       |                       |                      |                     |                    |                          |
|---------|----------------------------|--------------------|-------------------------------|-----------------------|-----------------------|----------------------|---------------------|--------------------|--------------------------|
| Tree    | I                          | Diameter (m        | Length (cm)                   |                       |                       | Weight (gr)          |                     |                    |                          |
| species | Capillary                  | Fine               | Coarse                        | Capillary             | Fine                  | Coarse               | Capillary           | Fine               | Coarse                   |
| Black   | $0.10^{a}\pm0.04$          | $0.31^{ab}\pm0.08$ | $0.72^{ac} \pm 0.21$          | 183 <sup>b</sup> ±115 | 157 <sup>b</sup> ±119 | 29 <sup>ab</sup> ±33 | $1.69^{a}\pm1.28$   | 4.61ª±2.85         | 3.33 <sup>ab</sup> ±1.95 |
| pine    |                            |                    |                               |                       |                       |                      |                     |                    |                          |
| Scotch  | $0.13^a \pm 0.04$          | $0.3^{ab}\pm 0.08$ | $0.70^{\mathrm{ac}}{\pm}0.22$ | $79^{ab}\pm 24$       | 64 <sup>ab</sup> ±44  | 70 <sup>a</sup> ±59  | $0.73^a\!\pm\!0.16$ | 3.01ª±2.24         | $7.02^{ab}\pm 2.93$      |
| pine    |                            |                    |                               |                       |                       |                      |                     |                    |                          |
| Fir     | $0.11^{ab}\!\!\pm\!\!0.05$ | $0.3^{b}\pm 0.08$  | $0.96^{bc}{\pm}0.37$          | 64 <sup>ab</sup> ±29  | 50 <sup>ab</sup> ±36  | 53ª±49               | $3.82^{ab}\pm 6.8$  | $2.75^{ab}\pm2.73$ | 15.7 <sup>b</sup> ±12.5  |

Statistically, it was determined that the root diameter, length and weight of the trees were showed a significant relationship (p < 0.05) between the tree species on which the trees were grown on different bedrock (Table 3). The lowest average root weight and root diameter were determined on the tree roots on the schist bedrock, while the highest average root weight

and root diameter were determined on the trees on the flysch bedrock. The roots of trees on the schist bedrock had the longest roots, while the roots of trees on the neritic limestone bedrock had the lowest length (Table 3).

| Root          |                          |            |                         |                          |       |        |
|---------------|--------------------------|------------|-------------------------|--------------------------|-------|--------|
| properties    | Schist                   | Komatiite  | Neritic Limestone       | Flysch                   | F     | p*     |
| Weight (gr)   | 3.49ª±2.56               | 4.06ª±3.29 | 6.72 <sup>a</sup> ±7.79 | 13.5 <sup>b</sup> ±13.83 | 9.054 | 0.000* |
| Length (cm)   | 130 <sup>b</sup> ±117.82 | 69ª±43.72  | 50ª±28.44               | 61ª±53.27                | 8.338 | 0.000* |
| Diameter (mm) | 0.36ª±0.26               | 0.41ª±0.27 | $0.54^{ab}\pm 0.43$     | $0.75^{b}\pm0.45$        | 6.825 | 0.000* |

Table 3. The average diameter, length and weight of black pine, scotch pine and fir tree turnings on different bedrocks

\*p<0.05

The average weight, length and diameter of the deposed black pine, Scotch pine and fir roots are given in Table 4. In terms of these values, there were significant differences in root weight, length and diameter among tree species (p < 0.05). The lowest values were in black pine roots and the highest root weight and root diameter were in the fir roots. The highest root length were in black pine roots; the lowest was determined in fir roots (Table 4).

**Table 4.** The average diameter, length and weight of overturned black pine, scotch pine and fir tree

| Scotch pine<br>4.0 a±3.29 | <b>Fir</b><br>9.88 <sup>b</sup> ±11.49 | 7 930                                  | 0.001*   |
|---------------------------|--|--|--|
| $4.0^{a}\pm 3.29$         | $9.88^{b} \pm 11.49$                   | 7 930                                  | 0.001*   |
|                           | J.00 -11.17                            | 1.750                                  | 0.001  |
| 69ª±43.72                 | 53ª±42.36                              | 12.324                                 | 0.000*   |
| $0.41^{ab}\pm 0.27$       | 0.63 <sup>b</sup> ±0.45                | 7.480                                  | 0.001*   |
|                           | $0.41^{ab}\pm 0.27$                    | $0.41^{ab}\pm 0.27$ $0.63^{b}\pm 0.45$ | $0.41^{ab}\pm 0.27$ $0.63^{b}\pm 0.45$ $7.480$ |

\*p<0.05

The average root length and weight values according to root diameter classification are given in Table 5. In terms of these values, it was found that there was a significant difference between root properties and root diameter classification (p < 0.05). Root weight was observed the lowest fort he capillary roots, the highest was determined in the coarse roots (Table 5). Root length was highest capillary roots; lowest was determined in the coarse roots (Table 5).

**Table 5**. The average root weight and lengths according to root diameter classification

| Root        |                          | Root Type              |                          | Б      | **          |
|-------------|--------------------------|------------------------|--------------------------|--------|-------------|
| Properties  | Capillary Root           | Fine Root              | Coarse Root              | г      | h.          |
| Weight (gr) | 2.31ª±4,01               | 3.59ª±2.83             | 12.1 <sup>b</sup> ±11.74 | 17.917 | $0.000^{*}$ |
| Length (cm) | 134 <sup>b</sup> ±105.03 | 98 <sup>b</sup> ±97.42 | 49ª±47.02                | 9.254  | $0.000^{*}$ |
|             |                          |                        |                          |        |             |

\*p<0.05

The chi-square test results of the relationship between root diameter classes of tree species and bedrock type are given in Table 6 and Table 7. The Pearson Chi square value between the tree type and the root class was calculated as 9.327 and its p value (Asymp Sig 2 sided) was calculated as 0.053. According to the results obtained; There was no relationship between tree species and root diameter classes (p > 0.05), these two variables were independent of each other (Tablo 6).

**Table 6.** Chi-square results of the relationship between tree type and root diameter classes

| Tree species*Root<br>classes |             | Root diameter classes |              |                |       |            |    |       |
|------------------------------|-------------|-----------------------|--------------|----------------|-------|------------|----|-------|
|                              |             | Capillary<br>Root     | Fine<br>Root | Coarse<br>Root | Total | Chi-square | df | р     |
| Trace                        | Black pine  | 12                    | 21           | 12             | 45    |            |    |       |
| Species                      | Scotch pine | 2                     | 5            | 4              | 11    | 9.327ª     | 4  | 0.053 |
|                              | Fir         | 7                     | 22           | 34             | 63    |            |    |       |
| r                            | Fotal       | 21                    | 48           | 50             | 119   |            |    |       |

<sup>a</sup> 3 cells (33.3%) less than expected number. The expected minimum number is 1.94.

The Pearson Chi square value was calculated as 13.133 between the bedrock and root diameter classes and p value of was 0.041. A significant level of  $\alpha = 0.05$  (P <0.05) showed that there was a significant relationship between root classes and bedrock type (Table 7).

| Table 7. | Chi-square | results between  | bedrock type | and root cla | ss(p < 0.05) |
|----------|------------|------------------|--------------|--------------|--------------|
| Lable /  | on square  | results set ween | ocaroon type | una root ena | bb (p \0.05) |

|              | Doduo de Trus e*  |                   | Root diameter classes |                |       |                     |    |             |
|--------------|-------------------|-------------------|-----------------------|----------------|-------|---------------------|----|-------------|
| Root classes |                   | Capillary<br>Root | Fine<br>Root          | Coarse<br>Root | Total | Chi-square          | df | р           |
|              | Schist            | 12                | 21                    | 12             | 45    |                     |    |             |
| Bedro        | ck Komatiite      | 2                 | 5                     | 4              | 11    |                     |    |             |
| Туре         | Neritic Limestone | 5                 | 14                    | 14             | 33    | 13.133 <sup>a</sup> | 6  | $0.041^{*}$ |
|              | Flysch            | 2                 | 8                     | 20             | 30    |                     |    |             |
|              | Total             | 21                | 48                    | 50             | 119   |                     |    |             |

<sup>a</sup>3 cells (25.0%) less than expected number. The expected minimum number is 1.94.

#### Discussion

The average weight, length and diameters of the overturned tree roots studied show significant changes depending on the bedrock and tree species (Table 2, 3 and 4). The root length is the highest in the black pine roots on the schist bedrock and is the lowest in the fir roots on the neritic limestone bedrock (Table 3 ve 4). The differences in these forms of rooting can have an important effect on how tree species interact with the bedrock surface, how well they grow on shallow soils and how deep their roots are. Atalay (2015) has stated that fertile forests and dense grass cover are located on impermeable lands such as schist, gneiss, where water holding capacity is high. Yılmaz and Kaymak (2018) have shown that they have a good growing habitat for species such as black pine (*Pinus nigra*) and Taurus fir (*Abies cilicica*) at levels higher than 1500 m. in the lands composed of schist, gneissic etc. impermeable rocks. Rudolph (1985) reported that the black pine (Pinus banksiana L.) root penetrated to a depth of 3 m in deep, well-drained soils and generally had side roots that rotated downwards and grown as they approached other trees. Berndt and Gibbons (1958) have found that the roots of the same tree species growing on different bedrock have different root systems. In addition, due to the fact that black pine roots are a taproot system, it is possible that the root length is too high (Shouse and Phillips, 2016) (Table 2 and 4). Kodrík and Kodrík (2002) have noted that the number of roots with a diameter of 3-10 cm is an important factor for tree static stability and the roots with a diameter of 10 cm are most important for tree stability. Kodrík (2002) found that the relative root amount below the diameter of 3 cm was 59.5%, the diameter of 3.1-9.0 cm was 28% and the root diameter above 10 cm, and the total number of roots in the overturned spruce was only 12.5%. The results of the comparison of root and branch structure show that the number and length of root branches up to 6 cm root branch diameter has a great effect on the stability of the roots grown in badly drained areas (Stofko and Kodrik, 2008).

The roots of the fir trees are small, thick and large, and can penetrate between the bedrock cracks (Stokes et al., 2005). Raftoyannis et al. (2008) have stated that there is a shallow and splayed root system in rocky areas, although the root system of the Greek fir (*Abies cephalonica* Loudon) is generally rather shallow, deeper and in deep soils according to growing environment. However, root growth under adverse conditions may cause a shallow root system that increases the probability of water stress (Ulrich 1989; Gruber 1994). During drought, fine root growth is strongly reduced (Blanck et al., 1995). The structural properties of the the less deep soil that neritic limestones with more rigorous decomposition (chemical and mineralogical composition) is low root length of fir species, show a shallow root growth rather than taproot (Akbulut, 1980). In addition, neritic limestones differ from other rocks in terms of plant root development due to their chemical and lithological properties. Kantarci (1982) explaines that forest trees develop shallow and inadequate root systems in shallow or stony soils. Due to the fact that the soil is shallow or stony, inadequate water and nutrient capacity cause slow growth of the plants, thus decreasing the resistance to storm and the emergence of windthrows.

Root weight and root diameter on the flysch bedrock were the highest in the roots of the fir species and the lowest in the black pine roots on the schist bedrock (Table 3). It is noted low in the schist bedrock for black pine species, the mass distribution of the root system, substantially the upper 45 cm of soil and the effect of lateral roots often is limited in the first 3 cm (Witty et al., 2003). Flysches bedrock consists of alternation of variegated, greenish gray colored marl, sandstone and limestone layers (Kavak and Toprak, 2011). On a horizontally layered flysch, the soil is usually shallow and prevents the roots from reaching the depths (Kantarcı, 1982). In this study, the fir species flysches film on an inclined ground deeper roots with a taproot system has evolved in loose flysch layers. Since fir roots can easily reach water and nutrients in soil depths, root diameter and root weight can be expected to be high.

The root length and root diameter of the Scotch pine, which grows on the komatiit bedrock which is one of the ultramafic rocks, has higher value than the fir species grown on the neritic limestone (Table 3). In general, all types of Scotch pine, which grow on all types of parent materials, develop a taproot on well undifferentiated main materials (such as komatiit) in horizontal direction (Atalay and Efe, 2012). It is also known that the roots of Scotch pine have spread to 0.125 ha (Laitakarai, 1927). The ratio of capillary and fine roots is higher than that of coarse roots as it develops the root system

in the horizontal direction of the Scotch pine. The soil formation is very low due to the difficult decomposition of the severe alkaline komatiit bedrock and Scotch pine is creating shallow roots may be seen in the field of windthrows.

A positive relationship was observed between the root class and the bedrock types with the chi-square test (Table 7) and no relation was found between root class and tree species (Table 6). Biomechanical interactions between tree roots and bedrock types are more evident when the growing roots meet the gaps in the rock or in the weaker parts (Witty et al., 2003). The root comparison between species is not easy, but depends on the physical properties of the soil and the depth of rooting (Nicoll et al., 2006).

#### 4. CONCLUSION

The work presented here, the biomechanical effects of overturned trees of relatively different bedrock in the forest with local characteristics, these points-based pedalogical effect was conducted to further emphasize more role and impact of the root system. The results of the study have shown that bedrock and tree species have an effect on some root characteristics and development potentials of overturned trees and the results of the study here will also be the basis for the next biogeomorphological studies.

The results have indicated that the root characteristics have an effect on different species growing on schist, neritic limestone, komatiite and flysch bedrock. Factors such as the width and surface of the root plate of the species spreading on the bedrock vary depending on the growing environment changes, and they are a very important factor in the stability of the trees. In general, the windthrow has a negative effect on the depth of the root system, especially on the tree species growing at high altitudes and no matter how shallow the roots and root system is determined to be susceptible to so much rooting.

Many climatic zones that may affect the tree rooting anchorage are silvicultural and biological factors. However, the rooting environment (soil, geology and hydrology) probably has the greatest restrictions. The published data on root characteristics for overturned trees containing observations on this rooting environment are quite limited. Therefore, for all tree species, it should be emphasized in the next scientific studies that the vast majority of the roots will occur in the uppermost soil meter.

As a result, our studies have shown that the state of the wind transitions are reflected in the root state. This study revealed the well-defined differences in diminished and apparently overturned trees in the thin-root state and the correlations between the bedrock and a set of root parameters. The key causal factor of the increase of tree turnovers may increase the conditions such as predicted climate change, soil formation limitation (soil depth), water holding capacity, drainage status, irregularity of precipitation and other abiotic factors.

#### REFERENCES

- Acar M (2009) Kök Ayrışma Seyri Üzerinde Ağaç Türü, Bakı ve Yükseltinin Etkisi. Yüksek Lisans Tezi Artvin Çoruh Üniversitesi Fen Bilimleri Enstitüsü, Artvin.
- Akbaş B, Akdeniz N, Aksay A, Altun İ, Balcı V, Bilginer E, Bilgiç T, Duru M, Ercan T, Gedik İ, Günay Y, Güven İ H, Hakyemez H Y, Konak N, Papak İ, Pehlivan Ş, Sevin M, Şenel M, Tarhan N, Turhan N, Türkecan A, Ulu Ü, Uğuz M F, Yurtsever A vd. (2016) Türkiye Jeoloji Haritası. Maden Tetkik ve Arama Genel Müdürlüğü Yayını, Ankara.
- Akbulut A (1980) Eğridir Gölü Güneyinde Çandır [Sütçüler, Isparta] Yöresindeki Batı Torosların Jeolojisi. Türkiye Jeoloji Kurumu Bülteni, 23: 1-9.
- Atalay İ (2015) Türkiye Vejetasyon Coğrafyası, Meta Basım, İzmir.
- Atalay İ & Efe R (2012) Sarıçam [*Pinus sylvestris* var. *sylvestris*] Ormanlarının Ekolojisi ve Tohum Nakli Açısından Bölgelere Ayrılması. T C Orman ve Su İşleri Bakanlığı, Orman Ağaçları ve Tohumları Islah Araştırma Enstitüsü Müdürlüğü, Meta Basım Matbaacılık, İzmir.
- Berndt H W & Gibbons R D (1958) Root Distribution of Some Native Trees and Understory Plants Growing on Three Sites within Ponderosa Pine Watersheds in Colorado. US Department of Agriculture, Forest Service, Rocky Mountain Forest and Range Experiment Station. Fort Collins, Colo., Station Paper 37.
- Blanck K, Lamersdorf N, Dohrenbusch A & Murach D (1995) Response of a Norway Spruce Forest Ecosystem to Drought/Rewetting Experiments at Solling, Germany. Water Air Soil Pollut., 85: 1251–1256.
- Brázdil R, Dobrovolný P, Štekl J, Kotysa O, Valášek H & Jež J (2004) History of Weather and Climate in the Czech Lands VI: Strong Winds. Masaryk University, Brno, Czech Republic.

Büsgen M, Münch E & Thomsom T (1929) The Structure and Life of Forest Trees. Chapman and Hall, London.

- Coutand C, Dupraz C, Jaouen G, Ploquin S & Adam B (2008) Mechanical Stimuli Regulate the Allocation of Biomass in Trees: Demonstration with Young *Prunus avium* Trees. Annals of botany, *101*(9): 1421-1432.
- Coutts M P (1983) Root Architecture and Tree Stability. Plant Soil, 71: 171-188.
- Coutts M P (1986) Components of Tree Stability in Sitka Spruce on Peaty Gley Soil. Forestry, 59: 173-197.
- Coutts M P (1989) Factors Affecting the Direction of Growth of Tree Roots. Annales des Sciences Forestières 46: 277-287.
- Cucchi V, Meredieu C, Stokes A, Berthier S, Bert D, Najar M & Lastennet R (2004) Root Anchorage of Inner and Edge Trees in Stands of Maritime Pine [*Pinus pinaster* Ait.] Growing in Different Podzolic Soil Conditions. Trees, 18(4): 460-466.
- Cutler D F, Gasson P E & Farmer M C (1990) The Wind Blown Tree Survey: Analysis of Results. Arboricultural Journal 14: 265-286.
- Dupuy L, Fourcaud T, Stokes A & Danjon F (2005) A Density-based Approach for the Modelling of Root Architecture: Application to Maritime pine [*Pinus pinaster* Ait.] Root Systems. Journal of Theoretical Biology, *236*(3): 323-334.
- Fraser A I & Gardiner J B H (1967) Rooting and Stability in Sitka Spruce. For. Comm. Bull. 40, HMSO, London.
- Gardiner B A & Quine C P (2000) Management of Forests to Reduce the Risk of Abiotic Damage a Review with Particular Reference to the Effects of Strong Winds. For. Ecol. Manage. 135: 261-277.
- Gruber F (1994) Morphology of Coniferous Trees: Possible Effects of Soil Acidification on the Morphology of Norway Spruce and Silver Fir. In: Godbold D L, Huttermann A (Eds.). Effects of Acid Rain on Forest Processes. 265–324, New York: Wiley.
- Huggett R J (1995) Geoecology. An Evolutionary Approach. Routledge, London and New York.
- Kantarcı D (1982) Ağaçlandırma Alanlarında Arazi Hazırlığı ve Toprak İşlemesinin Orman Yetişme Ortamı Üzerindeki Etkileri. İstanbul Üniversitesi Orman Fakültesi Dergisi, 32(2): 52-93.
- Kavak O & Toprak S (2011) Gölbaşı Harmanlı [Adıyaman] Kömürlerinin Organik Jeokimyasal ve Petrografik Özellikleri. Jeoloji Mühendisliği Dergisi, 35(1): 43-78.
- Kirkby M (2004) In: Goudie A S (Ed.), Soil Creep. Encyclopedia of Geomorphology, 2: 974-977 IAG, Routledge, London and New York.
- Kodrík J (2002) Výskum Koreňových Sústav Hlavných Lesných Drevín Vzhľadom na Statickú Stabilitu Voči Vetru. Zprávy lesnického výzkumu, 47: 208–213.
- Kodrík J & Kodrík M (2002) Root Biomass of Beech as a Factor Influencing the Wind Tree Stability. Journal of Forest Science, 48: 549-564.
- Kônopka J (1978) Optimálna a Krajná Hranica Rastových Vlastností Stromov Smreka Z hľadiska Odolnosti Lesných Porastov Proti Vetru v oblasti Nízkych Tatier. In: HEŠKOVÁ A. a kol., Poznatky z ochrany lesov. Vedecké práce VÚLH 26: 13–41, Bratislava, Príroda.
- Köstler J N, Brückner E & Bibelriether H (1968) Die Wurzeln der Waldbäume. Untersuchungen zur Morphologie der Waldbäumein Mitteleuropa. Paul Parey, Hamburg and Berlin, Germany.
- Laitakari E (1927) The Root System of Pine, a Morphological Investigation. Acta Forestalia Fennica, 33: 1-380.
- Linke J, Betts M G, Lavigne M B & Franklin S E (2007) Introduction: Structure, Function and Change of Forest Landscape. In: Wulder M A, Franklin S E (Eds.) Understanding Forest Disturbance and Spatial Pattern. Remote Sensing and GIS Approaches. Taylor & Francis, Boca Raton, London, New York, pp. 1-29.
- Mickovski S B & Ennos A R (2003) The Effect of Unidirectional Stem Flexing on Shoot and Root Morphology and Architecture in Young *Pinus sylvestris* Trees. Canadian Journal of Forest Research, *33*(11): 2202-2209.
- Nicoll B C & Ray D (1996) Adaptive Growth of Tree Root Systems in Response to Wind Action and Site Conditions. Tree physiology, *16* (11-12): 891-898.
- Nicoll B C, Gardiner B A, Rayner B & Peace A J (2006) Anchorage of Coniferous Trees in Relation to Species, Soil Type, and Rooting Depth. Canadian Journal of Forest Research, *36*(7): 1871-1883.
- Nieuwenhuis M & Fitzpatrick P J (2002) An Assessment of Stem Breakage and the Reduction in Timber Volume and Value Recovery Resulting from a Catastrophic Storm: an Irish Case Study. Forestry, 75: 513-523.
- Pickett S T A & White P S (1985) The Ecology of Natural Disturbance and Patch Dynamics. Academic Press, Inc., Orlando, USA.
- Pontailler J Y, Faille A & Lemée G (1997) Storms Drive Successional Dynamics in Natural Forests: a Case Study in Fontainebleau Forest [France]. For. Ecol. Manage., 98: 1-15.
- Quine C P, Coutts M P, Gardiner B A & Pyatt D G (1995) Forests and Wind: Management to Minimise Damage. For. Comm. Bull. 114, HMSO, London.
- Raftoyannis Y, Spanos I & Radoglou K (2008) The Decline of Greek Fir [*Abies cephalonica* Loudon]: Relationships with Root Condition. Plant Biosystems, 142(2): 386-390.
- Ray D & Nicoll B C (1998) The Effect of Soil Water-table Depth on Root-Plate Development and Stability of Sitka Spruce. Forestry, 71: 169-182.

- Reubens B, Pannemans B, Danjon F, De Proft M, De Baets S, De Baerdemaeker J, ... & Muys B (2009) The Effect of Mechanical Stimulation on Root and Shoot Development of Young Containerised *Quercus robur* and *Robinia pseudoacacia* Trees. Trees, 23(6): 1213-1228.
- Rudolph T D (1985) Jack pine [Pinus banksiana Lamb.]. FS-252.
- Sariyildiz T (2015) Effects of Tree Species and Topography on Fine and Small Root Decomposition Rates of Three Common Tree Species [*Alnus glutinosa, Picea orientalis* and *Pinus sylvestris*] in Turkey. Forest Ecology and Management, 335: 71-86.
- Savill P S (1983) Silviculture in Windy Climates. For. Abstr. 44: 473-488.
- Schaetzl R J, Burns S F, Small T W & Johnson D L (1990) Tree Uprooting: Review of Types and Patterns of Soil Disturbance. Phys. Geogr., 11: 277-291.
- Shouse M & Phillips J (2016) Soil Deepening by Trees and the Effects of Parent Material. Geomorphology, 269: 1-7.
- Stofko P & Kodrik M (2008) Comparison of the Root System Architecture between Windthrown and Undamaged Spruces Growing in Poorly Drained Sites. J. For. Sci, 54: 150-160.
- Stokes A, Ball J, Fitter A H, Brain P & Coutts M P (1996) An Experimental Investigation of the Resistance of Model Root System to Uprooting. Annals of Botany, 78: 415-421.
- Stokes A (2002) Biomechanics of Tree Root Anchorage. In Plant Roots, CRC Press, 297-314.
- Stokes A, Salin F, Kokutse A D, Berthier S, Jeannin H, Mochan S & Fourcaud T (2005) Mechanical Resistance of Different Tree Species to Rockfall in the French Alps. Plant and soil, 278(1-2): 107-117.
- Sutton R F (1980) Root System Morphogenesis. New Zealand Journal of Forest Science 10: 265-292.
- Toumey J W (1929) Initial Root Habit in American Trees and its Bearing on Regeneration. In: Proceedings of the International Congress of Plant Sciences, vol. 1, ed. B M Duggar, George Banta, Ithaca, New York, 713-728.
- Ulanova N G (2000) The Effects of Windthrow on Forests at Different Spatial Scale: a Review. Forest Ecology and Management,135: 155-167.
- Ulrich B (1989) Effects of Acidic Precipitation on Forest Ecosystems in Europe. In: Adriano D C, Johnson A H (Eds.). Acidic Precipitation, Biological and Ecological Effects. 2: 189–272, New York: Springer.
- White P S (1979) Patterns, Process, and Natural Disturbance in Vegetation. The Botanical Review 45, 229-299.
- Wilkinson M T, Richards P J & Humphreys G S (2009) Breaking Ground: Pedological, Geological, and Ecological Implications of Soil Bioturbation. Earth-Science Reviews 97: 257–272.
- Wilson B F (1975) Distribution of Secondary Thickening in Tree Root Systems. In The Development and Function of Roots. Eds. J G Torrey & D T Clarkson. Academic Press, 197-219, London.
- Witty J H, Graham R C, Hubbert K R, Doolittle J A, & Wald J A (2003). Contributions of Water Supply from the Weathered Bedrock Zone to Forest Soil Quality. Geoderma 114: 389-400.
- Yılmaz F K & Kaymak H (2018) Şist ve Gnayslar Üzerindeki Arazi Kullanımına Alanya'nın Doğusu Örneği. Atatürk Üniversitesi Sosyal Bilimler Enstitüsü Dergisi, 22 (3): 2745-2773.
- Zengin O (2010) Giresun ili Alucra Yöresi Saf ve Karışık Sarıçam Meşçerelerinde Kalın Kök Kütlesinin Belirlenmesi. Yüksek Lisans Tezi, Artvin Çoruh Üniversitesi Fen Bilimleri Enstitüsü, Artvin.



# Antimicrobial Activity and GC-MS Analysis of Extracts of *Achillea millefolium* L. and *Achillea biserrata* M. Bieb. Grown in Turkey

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Abstract: In this study, the antimicrobial activities of the ethanolic extracts of Achillea millefolium L. (Yarrow/Civanperçemi) and Achillea biserrata M. Bieb. (Aksırıkotu), used in the folk-medicine, were tested against gram-negative and gram positive bacteria as well as yeast-like fungi by the agar diffusion method. 19 microorganisms namely, Enterobacter aerogenes ATCC 13048, Salmonella infantis, Listeria monocytogenes ATCC 7644, Klebsiella pneumoniae, Pseudomonas aeruginosa DSMZ 50071, Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis ATCC 29212, Listeria innocua, Salmonella enteritidis ATCC 13075, Enterococcus durans, Salmonella typhimurium SL 1344, Candida ablicans ATCC 10231, Enterococcus faecium ATCC 25923, Staphylococcus aureus, Staphylococcus epidermidis DSMZ 20044, Bacillus subtilis DSMZ 1971, Escherichia coli ATCC 25922 and Serratia marcescens were used. The extracts of Achillea millefolium and Achillea biserrata showed antibacterial activity against tested microorganisms at different levels.

Keywords: Achillea millefolium, Yarrow, Civanperçemi, Achillea biserrata, Aksırıkotu, disc diffusion, GC-MS

# **1. INTRODUCTION**

Plants and their essential oils, have been used since the beginning of human history. The essential oils are typically liquid, clear and unusually colored, complex and the present compounds are volatile, characterized by a strong odor and synthesized by aromatic plants during secondary metabolites, which act to protect the plant against microorganisms and insects. They can be synthesized in several plant organs such as buds, flowers, leaves, stems, branches, seeds, berries, roots, wood or bark, being stored in secretory cells, cavities, channels, epidermal cells or trichomes. Industrial production of essential oils are obtained bysteam distillation, which is on the rise in food and pharmaceutical applications, and pressurized supercritical carbon dioxide extraction. Essential oils have several biological properties, such as larvicidal action, antioxidant, analgesic and anti-inflammatory, fungicide and antitumor activity. This paper is concerned with antimicrobial activity of ethanol (65%) extracts of *Achillea millefolium* and *Achillea biserrata* against 19 microorganisms by using the disk diffusion method, and volatile components of *Achillea millefolium* and *Achillea biserrata* were determined by GC-MS analysis (Gas chromatography-Mass spectrometry).

# 2. MATERIAL AND METHODS

# **Plant Samples**

In this study, *Achillea millefolium* and *Achillea biserrata* were collected from Yaralıgöz Gate/Küre Mountain (Kastamonu/Turkey) at 1330 m. *Achillea millefolium and Achillea biserrata* leaves and flowers were used. Plant materials were dried at room temperature and on draft for three weeks. All the plants was identified by Assist. Prof. Dr. Kerim GÜNEY, Department of Forest Engineering, Faculty of Forestry, Kastamonu University.

# **Extraction method**

The plants were washed thoroughly 2-3 times with water, and then they were air dried under shade. Afterwards, the dried plant materials were ground in a mixer, the powder was kept in the amber glass bottle. About 10-30 g ground plant samples were extracted with 250 mL of ethanol (%65) in a Soxhlet apparatus by continuous heat extraction for 24 hours. All extract solutions were filtered through Whatman No.1 paper. Then, filtrates were evaporated with a rotary evaporator. The filtrates were freeze dried and stored in refrigerator at about 4°C after sealed with paraffin for further studies.

# **Determination of Antimicrobial Activities**

## **Preparation of Extract Stock**

Extract stocks to test the antimicrobial activity were prepared by dissolving 1 mg of extract in each 3 mL of ethanol for disk diffusion test.

# Strains

In order to analyse the antimicrobial activity of plants extracts, 19 microorganisms namely, *Enterobacter aerogenes* ATCC 13048, *Salmonella infantis, Listeria monocytogenes* ATCC 7644, *Klebsiella pneumoniae, Pseudomonas aeruginosa* DSMZ 50071, *Pseudomonas fluorescens, Salmonella kentucky, Enterococcus faecalis* ATCC 29212, *Listeria innocua, Salmonella enteritidis* ATCC 13075, *Enterococcus durans, Salmonella typhimurium* SL 1344, *Candida albicans* ATCC 10231, *Enterococcus faecium* ATCC 25923, *Staphylococcus aureus, Staphylococcus epidermidis* DSMZ 20044, *Bacillus subtilis* DSMZ 1971, *Escherichia coli* ATCC 25922 and *Serratia marcescens* were used.

## **Preparation of Innocula**

All strains were incubated according to their requirements as it was previously mentioned by Altuner and Canlı (2012) and Canlı et al (2015). For the inocula, morphologically similar colonies of each organism were transferred into physiological saline (Canlı et al, 2016a and b, Onbasli, 2013) and to adjust equal the number of the colonies in the solution, 0.5 McFarland standard was used (Hammer et al., 1999; Altuner et al., 2012a and b). Thus standard inocula is adjusted to contain approximately  $10^8$  cfu/mL for bacteria and  $10^7$  cfu/mL for *C. albicans* (Canlı et al, 2016c and d).

## **Disk Diffusion Test**

The disk diffusion test was applied as it was mentioned in the previous studies (Andrews, 2003; Canlı et al, 2014). Petri dishes containing Mueller Hinton Agar were used for disk diffusion test (Ilhan et al., 2006; Canlı et al, 2016e). 20, 40 and 80  $\mu$ L of extracts were loaded on empty sterile antibiotic disks (SAD). Disks were kept at 40°C for 24 h in aseptic conditions (Altuner et al., 2010). Microorganism suspensions were inoculated on the surfaces of the Mueller Hinton plates and left in aseptic conditions for 2-3 minutes before applying disks as described in the previous studies (Altuner and Akata, 2010). Inhibition zones were defined in mm by the method mentioned by Altuner et al. (2014).

#### Controls

Empty SAD was used as negative controls for disk diffusion test, where broth medium inoculated with each microorganism was used to control microorganisms.

#### **Statistical Analysis**

All tests were done in triplicates. All the results given were mentioned as the mean values for these three parallel studies. The statistical analysis was performed using a non-parametric method Kruskal-Wallis one-way analysis of variance. Significance level for p was accepted as 0.05. A value of p < 0.05 was considered statistically significant (Altuner, 2011).

#### GC-MS (Gas Chromatography-Mass Spectrometry) Analysis

GC-MS analysis was performed using Shimadzu GCMS QP 2010 ULTRA series device. Samples were passed through RTX-5MS capillary column (30 m x 0.25 mm x 0.25 µm). Helium gas was used as carrier gas. The injection temperature was maintained at 250° C. GC-MS analysis was performed according to the procedure in the literature. The oven is heated to 40° C and is allowed to stand at this temperature for 3 minutes. Then the temperature is increased by 4 degrees per minute to 240° C and waited for 10 minutes. Finally, the temperature is increased by 4 degrees per minute to 260° C and kept for 65 minutes and is completed to a total of 78 minutes. The injection temperature was maintained at 250° C and the injection volume was determined as 1 µl. The intermittent temperature is 250° C and the ion boiling temperature is 200°C. It was treated with hexane.

# **3. RESULT AND DISCUSSION**

The GC-MS results of the extracts are presented in Table 1, and the antimicrobial activity test results are presented in Table 2.

Disk diffusion test results showed that Achillea millefolium is active against S. infantis, K. pneumoniae, P. fluorescens, S. kentucky, E. faecalis L. innocua, E. durans, E. faecium, S. ayreus and E. coli with inhibition zones between 7-10 mm, where Achillea biserrata is active against K. pneumoniae, S. kentucky, E. faecalis, L. innocua, S. enteritidis, S. typhimurium, E. faecium, S. ayreus, S. epidermidis and S. marcescens with inhibition zones between 7-9 mm.

## 4. CONCLUSION

Plants are natural reservoirs of various phytonutrients and compounds which are urgent and essential to life in general. The phytochemical component of ethanol extract of *Achillea millefolium and Achillea biserrata* by GC-MS analysis shows its significance in phytopharmaceuticals, cosmetic and food industries. On the basis of antimicrobial activity, it was monitored that the ethanol extracts were effective only for few of the tested species.

#### Acknowledgements

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|      |         | Α     | chillea millefolium                     | Achillea biserrata |       |   |  |
|------|---------|-------|---|--------------------|-------|---|--|
| Peak | R. Time | Area% | Component Name                          | R. Time            | Area% | Component Name                          |  |
| 1    | 41.695  | 1.31  | Hexadecanoic acid, methyl ester         | 65.420             | 1.12  | -                                       |  |
| 2    | 45.970  | 1.27  | 9-Octadecenoic acid, methyl ester, (E)- | 66.984             | 5.85  | .betaAmyrin                             |  |
| 3    | 50.398  | 6.26  | Hexacosane                              | 68.477             | 1.60  | Lup-20(29)-en-3-ol, acetate, (3.beta.)- |  |
| 4    | 54.137  | 1.06  | 9,12-Octadecadienoyl chloride, (Z,Z)-   | 69.728             | 3.21  | Lup-20(29)-en-3-ol, acetate, (3.beta.)- |  |
| 5    | 54.232  | 2.88  | DI-(9-Octadecenoyl)-Glycerol            | 70.489             | 22.69 | .alphaAmyrin                            |  |
| 6    | 54.690  | 10.28 | Pentacosane                             | 71.215             | 55.38 | Lupeol                                  |  |
| 7    | 60.678  | 12.04 | Pentacosane                             | 73.528             | 1.86  | Phytol, acetate                         |  |
| 8    | 64.054  | 1.84  | 13-Docosenamide, (Z)-                   | 75.947             | 1.62  | Flavone 4'-OH,5-OH,7-DI-O-Glucoside     |  |
| 9    | 64.729  | 1.53  | Pentacosane                             |                    |       |   |  |
| 10   | 68.132  | 35.22 | Hexatriacontane                         |                    |       |   |  |
| 11   | 71.471  | 1.63  | Hexatriacontane                         |                    |       |   |  |
| 12   | 75.593  | 7.19  | Hexatriacontane                         |                    |       |   |  |
| 13   | 75.927  | 2.10  | Oleic Acid, Propyl Ester                |                    |       |   |  |

Table 1. GC-MS analysis of Achillea millefolium and Achillea biserrata

| Table 2. | Disk Diffusion | Test Results of e | xtracts at the | Achillea millefoli | um and Achillea | biserrata | concentration |
|----------|----------------|-------------------|----------------|--------------------|-----------------|-----------|---------------|
|----------|----------------|-------------------|----------------|--------------------|-----------------|-----------|---------------|

|                  | I     | Achillea millefoliu | m     | Achillea biserrata |       |       |  |
|------------------|-------|---------------------|-------|--------------------|-------|-------|--|
| Microorganisms   | 20 µL | 40 µL               | 80 μL | 20 µL              | 40 µL | 80 µL |  |
| E. aerogenes     | -     | -                   | -     | -                  | -     | -     |  |
| S. infantis      | -     | -                   | 8     | -                  | -     | -     |  |
| L. monocytogenes | -     | -                   | -     | -                  | -     | -     |  |
| K. pneumoniae    | -     | 8                   | 8     | 8                  | 8     | 8     |  |
| P. aeruginosa    | -     | -                   | -     | -                  | -     | -     |  |
| P. fluorescens   | -     | -                   | 7     | -                  | -     | -     |  |
| S. kentucky      | -     | 7                   | 7     | -                  | 7     | 7     |  |
| E. faecalis      | -     | 8                   | 10    | 8                  | 9     | 9     |  |
| L. innocua       | -     | -                   | 7     | -                  | 7     | 7     |  |
| S. enteritidis   | -     | -                   | -     | 7                  | 7     | 7     |  |
| E. durans        | -     | 7                   | 9     | -                  | -     | -     |  |
| S. typhimurium   | -     | -                   | -     | -                  | -     | 7     |  |
| C. ablicans      | -     | -                   | -     | -                  | -     | -     |  |
| E. faecium       | 7     | 7                   | 7     | 7                  | 7     | 7     |  |
| S. aureus        | -     | 9                   | 10    | _                  | -     | 7     |  |
| S. epidermidis   | -     | -                   | -     | -                  | -     | 7     |  |
| B. subtilis   | - | - | - | - | - | - |
|---------------|---|---|---|---|---|---|
| E. coli       | - | - | 7 | - | - | - |
| S. marcescens | _ | _ | _ | _ | _ | 7 |

#### REFERENCES

- Altuner, E.M., I. Akata and K. Canlı. 2012a. *In vitro* antimicrobial screening of *Bovista nigrescens* (Pers.). Kastamonu U. J. For. Fac. 12:90-96.
- Altuner, E.M., I. Akata and K. Canlı. 2012b. In vitro antimicrobial screening of Cerena unicolor (Bull.) Murrill (Polyporaceae Fr. Ex Corda). Fresen. Environ. Bullet. 21:3704-3710.
- Altuner, E.M. and I. Akata. 2010. Antimicrobial activity of some macrofungi extracts. Sakarya Üniversitesi Fen Bilimleri Enstitüsü Dergisi. 14(1):45-49.
- Altuner, E.M., K. Canlı and I. Akata. 2014. Antimicrobial screening of *Calliergonella cuspidata*, *Dicranum polysetum* and *Hypnum cupressiforme*. Journal of Pure and Applied Microbiology. 8(1):539-545.
- Altuner, E.M. and K. Canlı. 2012. In vitro antimicrobial screening of Hypnum andoi A.J.E. Sm. Kastamonu U. J. For. Fac. 12:97-101.
- Altuner, E.M. 2011. Investigation of antimicrobial activity of *Punica granatum* L. fruit peel ash used for protective against skin infections as folk remedies especially after male circumcision. African Journal of Microbiology Research. 5(20):3339-3342.
- Andrews, J.M. 2003. BSAC standardized disc susceptibility testing method (version 6). Journal of Antimicrobial Chemotherapy. 60:20-41.
- Canlı, K., I. Akata and E.M. Altuner. 2016c. *In vitro* antimicrobial activity screening of *Xylaria hypoxylon*. African Journal of Traditional, Complementary and Alternative Medicines. 13(4):42-46.
- Canlı, K., E.M. Altuner, I. Akata, Y. Türkmen and U. Üzek. 2016a. *In vitro* antimicrobial screening of *Lycoperdon lividium* and determination of the ethanol extract composition by gas chromatography/mass spectrometry. Bangladesh Journal of Pharmacology. 11(2):389-394.
- Canlı, K., E.M. Altuner and I. Akata. 2015. Antimicrobial screening of *Mnium stellare*. Bangladesh Journal of Pharmacology. 10:321-325.
- Canlı, K., B. Çetin, E.M. Altuner, Y. Türkmen, U. Üzek and H. Dursun. 2014. *In vitro* antimicrobial screening of *Hedwigia ciliata* var. *leucophaea* and determination of the ethanol extract composition by gas chromatography/mass spectrometry (GC/MS). Journal of Pure and Applied Microbiology. 8(4):2987-2998.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016d. *In vitro* antimicrobial screening of *Aquilaria agallocha* roots. African Journal of Traditional, Complementary and Alternative Medicines. 13(5):178-181.
- Canlı, K., A. Yetgin, I. Akata and E.M. Altuner. 2016e. *In vitro* antimicrobial activity screening of *Rheum rhabarbarum* roots. International Journal of Pharmaceutical Sciences Invention, 5(2):1-4.
- Hammer, K.A., C.F. Carson and T.V. Riley. 1999. Antimicrobial activity of essential oils and other plant extracts. Journal of Applied Microbiology. 86:985-990.
- Ilhan, S., F. Savaroğlu, F. Çolak, C.F. Iscen and F.Z. Erdemgil. 2006. Antimicrobial activity of *Palustriella commutata* (Hedw.) Ochyra extracts (Bryophyta). Turk. J. Biol, 30:149-152.
- Kartal, M. (2004). Avrupa birliği ülkelerinde tıbbi bitkisel ürünlerin ruhsatlandırılması, Ankara Üniversitesi Eczacılık Fakültesi Farmakognozi Anabilim Dalı.
- Nychas, G. J. E. (1995). Natural antimicrobials from plants. In: Gould, G. W. (Ed.), New Methods of Food Preservation. (58pp) London, Blackie: *Academic Profesional*.
- Onbasli, D., G. Yuvali Celik, E.M. Altuner, B. Altinsoy and B. Aslim. 2013. *In vitro* antimicrobial, antioxidant, and antibiofilm activities of *Bryum capillare*, a bryophyte sample. Current Opinion in Biotechnology, 24 (Supplement 1): 113.
- Özcan, M. and Sağdıç, O. (2003). Antibacterial activity of Turkish spice hydrosols. Food Control.
- Rabe, T. and Van Staden, J. (1997). Antibacterial activity of South African plants used for medicinal purposes. *Journal of Ethnopharmacology*.



# The Effects of Different Aspect and Altitude Gradient on the Tolerate Time Against Shelter Effects in Pure Oriental Beech (*Fagus Orientalis* Lipsky.) Stands in Yenice-Kızılkaya Region

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Abstract: Conservation of forest resources, ensuring continuity and increasing the quality of these resources is very important in terms of not interrupting the products and services provided from these sources. Our country has forest resources with different species and establishment characteristics with the effect of different ecological conditions. One of these forest species is the pure and mixed forests formed by the oriental beech (Fagus orientalis Lipsky.). Oriental beech is a species that is difficult to be rejuvenated by natural regeneration because of its slow growth in youth, early and late frosts and damage from the weed. In addition, due to the high elasticity of the top structure, the need for light from early ages disrupts the structure of the hill and this leads to the occurrence of poor quality individuals in advanced growth stages. Determining the duration of the shelter is important for determining the time of the lighting cuts. In this research, in the Yenice-Kızılkaya region. In the three different elevation levels in the III. site class conditions, the duration of the seeding of the seedlings of seed origin in the shaded and sunny aspects were determined. For this purpose, the individual who started to spread 3 tops of each elevation and maintenance condition was selected and repeated. Variance analysis was applied to determine whether there was a statistically significant difference between aspect and the elevation conditions and Duncan range test was used in the groupings based on the differences resulting from the analysis of variance. In the research, the age of beech individuals was determined at the lowest error level by scanning the section taken from the soil surface on the Tree-Scanner equipment, and the number of years was no longer prevented from causing the error. As a result of the evaluations made with the help of variance analysis; A significant difference was found between P < 0.01 statistical significance level. The results of the Duncan range test performed at this level of P < 0.05significance level were found to be 10.3 years in the second altitude gradient (1000-1200m) and the first group in the first group. On the other hand the average shelter duration period was 7,2 years in the first altitude gradient (800-1000m) group and this reason this altitude group in the second group acoording to Duncan range test results. According to the results of variance analysis, a statistically significant difference was found in P < 0.05 significance level. Therefore, according to the Duncan range test performed at the same level of significance, the average duration of shelter effects of the individuals in both altitude levels with shaded aspects (North, northeast and eastern aspects) is 9.4 years and therefore they are in the first group, southwest and west aspects; the duration of shelter was 6.8 years and it was found in the second group. According to these results, in Yenice-Kızılkaya region, depending on the altitude gradient stage and aspect variables, it can be suggested to perform lighting cuts after the average seeding cutting time in beech natural regeneration applications after an average period of 6,4-10,2 years.

Keywords: Tolerate Time, Shelter, Oriental Beech, Altitude, Aspect.

# **1. INTRODUCTION**

The forests that meet the many needs of the society with the products and collective services they provide are the most important sources that can renew themselves naturally. In addition, the sustainable management and continuity of these important resources, which fully fulfill the carbon storage task, is the primary responsibility of all countries and nations in the long and short term, in order to eliminate the negative effects of global climate change. In this respect, it is necessary to determine the effects of all forest resources, especially the natural, and the ecological conditions where the forests are shaped for the new forest area gains, both at the general forest area level and at the species level. The ecological conditions and the effects of these species on the change of these conditions should be elaborated with full details, especially because of the slow growth in the youth and the susceptibility to changing ecological conditions. Due to different ecological conditions, our country is home to many species of wood and pure and mixed forestry. The silvicultural and ecological characteristics and desires of these species are absolutely different from each other. One of these species is the oriental beech (*Fagus orientalis* Lipsky.) widely used in our country because of its precious wood. 1.4 million hectares of natural

propagation arises when the species establishes precious and precious forests along the entire Marmara and Black Sea strip (ÖZEL, 2007). However, owing to its ability to withstand the shadow for a long time, the oriental beech has a very high peak flexibility (SAATÇİOĞLU, 1969; ATA, 1995). Therefore, in the ecological areas which are shaped by different elevation and maintenance conditions, the determination of the duration of the natural and young people of the eastern beech to the upper and side walls is determined both in the natural rejuvenation studies of the species with the Uniform Shelterwood Method and in the determination of the time of the light cuts after the seeding and the healthy peak and seed characteristics. It will be very useful for the establishment of new and valuable forests (PETERS, 1992).

In this research, which was carried out in 2 different elevation levels and 2 different conditions in Yenice-Kızılkaya Forest Range District, the natural youths found in natural rejuvenation areas were followed closely and the peak index was used to determine the peak times of the peaks and upper and side shield pressures. Differences in the duration of resistance of natural youths were determined.

# 2. MATERIAL AND METHODS

This research was carried out in 8 different compartments in the Yenikaya Forestry Directorate under the Forestry Directorate of Yenice. The research area generally lies within the Euro-Siberian forest belt and the soil is deep in sandy-loam and sandy-clayey-slime formation and in the clastic structure. However, the organic matter content in the soil is sufficient and these organic compounds are generally in the form of mull type humus form. The average temperature in the research area is 18.7°C, the average rainfall is 1126.2mm and the vegetation period is 6 months. There is not water deficit period in the research area. In general, there is a surplus period especially during the winter months (ANONIM, 2015). The natural rejuvenation areas of beech, which are evaluated in the research, are generally the areas intervened at the beginning of the previous plan period. In the areas where the determinations are made in natural juvenility, the general stand type is Knd<sub>1</sub>/a<sub>2</sub> and the site condition is low in almost all areas.

The research was carried out in two different elevation conditions: 800-1000m and 1000-1200m with 2 different elevation levels, shaded and sunny. Sampling was performed randomly in each elevation and maintenance condition. The youth peak status index shown in Figure 1 was used in the realization of these samples.



Figure 1. Index of Top State of Natural Oriental Beech Juvenilities (ÖZEL, 2007).

Descriptive information about the samples taken with the help of this index is given in Table 1. According to this, measurements and determinations were made in 137 natural beech youths of different size natural rejuvenation areas in 8 compartments.

| Division<br>No | Altitude<br>Gradient (m) | Stand<br>Type | Area<br>(Ha) | Sample<br>Items |
|----------------|--------------------------|---------------|--------------|-----------------|
| 29             | Ι                        | $Knd_1/a_2$   | 3.5          | 12              |
| 34             | Ι                        | $Knd_1/a_2$   | 4.2          | 20              |
| 41             | Ι                        | $Knd_1/a_2$   | 1.8          | 10              |
| 46             | Ι                        | $Knd_1/a_2$   | 2.4          | 15              |
| 63             | II                       | $Knd_1/a_2$   | 5.5          | 25              |
| 78             | II                       | $Knd_1/a_2$   | 2.2          | 15              |
| 105            | II                       | $Knd_1/a_2$   | 3.6          | 18              |
| 118            | II                       | $Knd_1/a_2$   | 4.8          | 22              |

Table 1. Description informations of research area and natural juvenility samples.

In order to determine the durations of durability of the natural beech youth according to the changing peak conditions, the body sections were cut by the soil surface and scanned at the Trees Scanner.

Variance analysis (ANOVA) was used to determine whether there was a difference between the duration of uplift of the natural eastern beech youth and the height of the upper and the side guard in terms of elevation levels and maintenance conditions. SPSS package statistical program was used to perform these analyzes.

# 3. RESULTS AND DISCUSSION

#### Results

As a result of the analysis of variance (ANOVA) applied to the age determinations of the stem sections taken from the soil surface from the natural juvenilities samples determined according to the peak state index shown in Figure 1 from the natural rejuvenation areas in the scope of the research; A significant difference was found between P < 0.01 levels. The results of the Duncan test performed at this level of P < 0.05 confidence level were found to be 10.3 years in the second peak (1000-1200m) and the first group in the first group (800-1000m). while the average duration of durability was found to be 7.2 years and the second group was found to be in the area. According to the results of variance analysis, a statistically significant difference was found in P < 0.05 confidence level. Therefore, according to the Duncan test performed at the same level of confidence, the average duration of stopping of the individuals in both elevation levels with shaded views (North, northeast and eastern aspects) is 9.4 years and therefore they are in the first group, southwest and west views; the duration of stopping was 6.8 years and it was found in the second group.

#### Discussion

According to the findings obtained from the research, oriental beech can withstand much longer periods of 800-1000m than the lower zones without spreading its top to the effect of the top and side shield in the 1000-1200m altitude gradient, which is dominated by optimal growth conditions. This situation is particularly related to the formation of optimal conditions in the soil moisture and organic matter content. As a matter of fact, in the determination of the European beech forests which have similar growth conditions with the eastern beech, 15-year durations of beech individuals to the upper and side walls due to the sufficient amount of moisture content in the soil and air in the soil and air in 1000-1500m elevation It is reported that it can go up (BRADSHAW ET AL., 2002). However, in this research carried out in the areas of natural regeneration of oriental beech in the K121kaya region, it was determined that the duration of resistance of the beech was longer in shaded areas than in the sunny ones. In another study, it was found that radioactive cooling was late and the moisture value was high and the longer time to resist the top and side shields in natural ecological conditions were found to be longer (DIACI, 2002; PODLASKI, 2002). In the light of these comparative data, it can be said that the duration of the upper and side shield of the eastern beech does not contain the ecological conditions close to the conditions of optimal natural distribution, but it can be said that it is at the highest level possible in the research area.

#### 4. CONCLUSION

According to the data obtained from this research, especially due to the fact that the duration of the trench in the 1000-1200m elevation of the eastern beech is about 10 years, it can be suggested that especially the lighting and even the proper biological independence can be discharged if the youth have the youth. However, these periods can be re-evaluated by taking into consideration the burning and drying temperature effect and frost danger which will occur in the area. On the other hand, in light of the 10 years of shelters, the lighting and discharging applications to be performed in the eastern beech stands can be planned with priority. However, since the research in question covers a maximum period of 10 years, it is useful to re-evaluate the research type considering the need for light at a later age.

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#### REFERENCES

Anonim (2015). Kızılkaya Orman İşletme Şefliği Silvikültür Planı [Silviculture Plan of Kızılkaya Forest Range District], Yenice-Karabük.

Ata, C. (1995) Silvikültür Tekniği [Silviculture Techniques], Z.K.Ü Bartın Orman Fakültesi, Üniversite Yayın No: 4, Fakülte Yayın No: 3, Bartın, 453 s.

- Bradshaw, R.H.W. and Mountford, E.P. (2002) Report to accompany maps of past European Fagus Forests, Nature-Based Management of Beech in Europe Project (NAT-MAN), Working Report 4, Denmark, 8 p.
- Özel, H.B. (2007). Bartın ve Devrek Doğu Kayını (Fagus orientalis Lipsky.) Ormanlarında Meşcere Kuruluşları ve Grup Gençleştirme Uygulamalarının Başarısını Etkileyen Faktörler. [Stand Structures and The Factors Affecting The Success of Group Natural Regeneration Practices in Bartın and Devrek Oriental Beech (Fagus Orientalis Lipsky.) Forests], ZKÜ Fen Bilimleri Enstitüsü Doktora Tezi (yayımlanmamış).

Peters, R. (1992) Ecology of Beech Forests in The Northern Hemisphere, Wageningen, Nederlands, 125 p.

- Podlaski, R. (2002) Relationship between crown characteristics and the radial increment of beech (Fagus sylvatica L.) the Swietokrzyski National Park (Poland), Journal of Forest Science (48), pp. 93-99.
- Saatçioğlu, F. (1969) Silvikültürün Biyolojik Esasları ve Prensipleri, [Biological Principles and Basic Concepts of Silviculture], İ.Ü Orman Fakültesi, İ.Ü Yayın No: 1429, O.F Yayın No: 138, İstanbul, 323 s.



# Variation in Needles Nutrient Concentrations Of Cedar (*Cedrus Libani* A. Rich) with Height And Diamater

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**Abstract:** The main aim of this present study was to determine the relationship between needle nutrient concentrations of Cedar trees (Cedrus spss.) and the different heights and diameters of trees growing in the city center of Kastamonu province. For this purpose, needles of different heights (between 8 m and 21 m) and diameters (between 22 cm and 104 cm) cedar trees were sampled and analysed for macro (Ca, Mg, P, K and S) and micro (Fe, Mn, Na, Cu, Zn, Cl, Al, Co) nutrients using atomic absorption spectrophotometry in Kastamonu University Central Research Laboratory. In addition, some soil properties of the cedar trees and the silvicultural characteristics of the trees were determined. According to the results, it was seen that there was a statistically significant relationship between the macro and micro nutrient concentrations of the needles and the diameter and height of the trees. However, this relationship was clearly observed to be among the trees with the lowest height and diameter, while P and K showed an increase. Similar relationships were determined between height and macro nutrients. With the exception of the Na concentration of the needles, the other micronutrient elements showed a decrease in height and diameter of the trees. In general, it is thought that the decrease in macro and micro nutrients observed in the needles of cedar trees depending on the height and diameter of the cedar trees is caused by the changes in the soil properties of cedar trees.

Key words: Nutrients, tree height, tree diameter, cedar needle, Kastamonu

# **1. INTRODUCTION**

The amount of nutrients varies greatly between ecosystems ((Binkly and Vitousek, 1989). This causes differences in the structure and production of the plant community. Soil profile, soil pH and nutrient cycle between soil and trees are significant in determining the quality field (Moore et al., 2004; Sheikh and Kumar, 2010). The amount of nutrients accumulated in a forest ecosystem varies depending on the species, frequency, age, basal area, altitude, climatic conditions, soil conditions and relative humidity conditions of existing forest trees (Sprugel, 1984; Ruark and Bockheim, 1988; Raison et al., 1992; Mitchell et al., 1996; Wang et al., 1996). It can also be measured by tree genetic diversity in nutrient concentration and efficiency in trees (Oleksyn et al. 2002; Raitio and Sarjala 2000). In trees with other structural differences with which are seen in plant characteristics such as tree morphology, physiology and biomass can be greatly affected nutrient cycle and other ecosystem functions. In previous studies, spruce trees of different ages, diameters and heights have been shown to differ in macro and micro nutrient concentrations in needles (Sasser and Binkley, 1989; Wang and Klinka, 1997). Researchers generally agree that leaves provide the best index for nutrient status in a tree (Howlett and Cahoon, 1964; Mead, 1984; Powers, 1984). Some researchers have found that nutrient ratios are better than nutrient levels of forest trees (Hellman and Gessel, 1963; Ingestad, 1967).

In this study, we aimed at determining the variation in macro (Ca, Mg, P, K, S) and micro (Fe, Mn, Na, Cu, Zn, Ci, Al, Co) nutrients in the needles of different aged cedar trees (*Cedrus libani* A. Rich.) with height and diamater located in Cumhuriyet Square Park at Kastamonu province.

# 2. MATERIAL AND METHODS

# **Study Area**

The study area was located in Turkey's western Black Sea region at Kastamonu city center which located between 41°22'35 N and 33°46'36 E in Republic Square Park (Figure 1).



Figure 1. General view of the study area

There are two different climate types in Kastamonu province. While the Black Sea climate is dominant in the northern part of the province, the influence of the Central Anatolia climate is observed in the south. Winters are cold and snowy and summers are mild and rainy. The average annual temperature is 9.8° C. The coldest months are January and February, while the hottest months are July and August. The distribution of precipitation is quite regular in Kastamonu and winter precipitation accounts for 18% of the annual precipitation and 28% for summer precipitations. Most of the precipitation falls in the spring months. The annual precipitation fell by 18% in May compared to a 6% in January. The average annual rainfall is 449.7 mm. The average wind speed is 14 m/sn in the province, the prevailing wind, in the southwest direction 3524 times during the year (URL-1).

Cedar trees (3 or 4 trees) in the Kastamonu province Cumhuriyet Square Park were chosen to examine. They were quite mature and at least 100 or 150 years old (Figure 2a, b, ve c).



(a)

(b)



**Figure 2.** Sampling of cedar trees **a**) 1<sup>st</sup> cedar tree **b**) 3<sup>rd</sup> cedar tree **c**) 5<sup>th</sup> cedar tree **d**) Measuring some soil properties of cedar samples

# Determination of soil and silvicultural characteristics

The soil temperature of the soil surface was measured by a digital thermometer with a stainless steel probe (Checktemp1 Hanna) (Karelin et al., 2015; Figure 2d). A tree diameter meter was used to measure the diameter of the tree on the breast height (130 cm) (Avsar, 2004). TruPulse Laser Rangefinder was used to measure the height of the trees (Unger et al., 2014). In addition, soil sensors were used to measure the pH (Digital Soil pH Meter) and moisture content (20 cm long Digital Soil Moisture Meter) of the soils under the cedar tree (Milosavljevic et al., 2016; Figure 2d).

#### Needle sampling and chemical analysis

The cedar tree is one of the dominant tree species in the green area of the Square park. Therefore, the needles of cedar trees were chosen for this study. Needle sampling in cedar trees was carried out on 7<sup>th</sup> of December in 2018. Approximately 60 g needles were collected from the 11 different aged cedar trees on each area. In each tree, at least two branches of the tree top as 1/2-1/4 were collected (Wang and Klinka, 1997; Kiser et al., 2013). The needle samples were dried in the oven at 60° C for a day (Kiser et al., 2013). The dried leaves were ground for chemical analysis using a Bosh type MKM6000 coffee grinder (Selcuk et al., 2011). The elements of macro nutrients (Ca, Mg, P, K and S) and micronutrients (Fe, Mn, Na, Cu, Zn, Cl, Al, Co) were determined by atomic absorption spectrophotometer in Kastamonu University Central Research Laboratory. Chemical results analysis nutrient concentrations are expressed in ppm.

#### Statistical analysis

Variance analysis (ANOVA) was performed by using SPSS program on whether there is any significant difference between some macro and micro nutrient elements of different diameter and size cedar needles. In accordance with ANOVA results, the significance of the difference was determined with Duncan test.

# 3. RESULTS AND DISCUSSION

#### Results

#### Some silvicultural and soil characteristics of the study area

The height, diameter, circumference of the cedar trees together with the soil reaction (pH), temperature (°C) and humidity (%) details of the soil properties are shown in Table 1. Since the second and third cedar trees were approximately 100-150 years, the tree diameter was quite high compared to the other young cedar trees (Table 1). While the highest tree height was 21 m (6th cedar tree), the lowest tree height was found for 8th and 10th cedar trees. The highest tree diameter (104 cm) and the circumference (300 cm) were observed in 3rd cedar tree, the lowest tree diameter (20 cm) and tree circumference (52 cm) were observed in 10th cedar tree. The average soil pH was 7.3 and slightly alkaline soil in 11th cedar trees. The average soil temperature of the cedar tree was 5.05° C and the average soil moisture was 6.5% (Table 1).

| Tree<br>No | Tree<br>Height<br>(m) | Tree<br>Diameter<br>(cm) | Tree<br>Circumference<br>(cm) | рН  | Soil<br>Temperature<br>(°C) | Moisture<br>(%) |
|------------|-----------------------|--------------------------|-------------------------------|-----|-----------------------------|-----------------|
| 1          | 20                    | 65,5                     | 190                           | 7.9 | 5.5                         | 7.5             |
| 2          | 18                    | 101                      | 258                           | 7.9 | 4.7                         | 2.1             |
| 3          | 18                    | 104                      | 300                           | 7.1 | 4.4                         | 8.5             |
| 4          | 14                    | 33                       | 100                           | 6.9 | 5.0                         | 9               |
| 5          | 15                    | 30                       | 102                           | 6.9 | 5.4                         | 10              |
| 6          | 21                    | 53                       | 156                           | 7.8 | 4.2                         | 5               |
| 7          | 12                    | 67                       | 138                           | 7.7 | 4.0                         | 3               |
| 8          | 15                    | 46                       | 98                            | 6.1 | 3.8                         | 10              |
| 9          | 9                     | 22                       | 62                            | 6.9 | 5.9                         | 8               |
| 10         | 8                     | 20                       | 52                            | 7.1 | 6.6                         | 5.5             |
| 11         | 10                    | 30                       | 75                            | 7.5 | 6.1                         | 2.5             |

Table 1. Changes in silvicultural and soil properties of cedar species of different diameters and heights

# Macro nutrient concentrations

Macronutrients (Ca, Mg, P, K and S) of cedar needles of different tree heights and diameters are given in Table 2 and Table 3. Statistically, the macronutrients in the neddles differed significantly between tree height and tree diameters (p<0.05).

Ca and S concentrations were highest for the 12 m height and 67 cm diameter in cedar needles, whereas Ca and S concentrations were lowest for 18 m height and 101cm and 104 cm diameters in cedar needles. The highest Mg concentrations were found for the 14 m height and 33 cm diameter in cedar needles, and the lowest concentrations was for the 21 m height and 53 cm diameter. The highest P concentrations were for the 20 m height and 65.5 cm in cedar needles while the lowest for the 12 m height and 46 cm diameter in cedar needles. The lowest K concentrations were for the 15 m height and 46 cm diameter while the highest K concentrations were for the 21 m height and 53 cm diameter (Table 2 and 3).

Table 2. Mean values of macro nutrients and Duncan test results on tree height basis

| Tree          | MACRO NUTRIENTS (ppm)   |                           |                         |                           |                         |       |  |  |
|---------------|-------------------------|---------------------------|-------------------------|---------------------------|-------------------------|-------|--|--|
| Height<br>(m) | Ca                      | Mg                        | Р                       | K                         | S                       | p*    |  |  |
| 8             | 21150 <sup>cd</sup> ±20 | 3718.0 <sup>de</sup> ±36  | 2201.0°±4               | 12190.0 <sup>ab</sup> ±20 | 4583.0 <sup>cd</sup> ±5 | 0.000 |  |  |
| 9             | 20610 <sup>cd</sup> ±20 | 3152.0 <sup>bcd</sup> ±33 | 2254.0°±4               | 13620.0bc±20              | 4717.0 <sup>d</sup> ±5  | 0.000 |  |  |
| 10            | 22190 <sup>d</sup> ±30  | 3560.0 <sup>cde</sup> ±34 | 2075.0 <sup>b</sup> ±4  | 15100.0bc±20              | 4662.0 <sup>cd</sup> ±5 | 0.000 |  |  |
| 12            | 26470 <sup>e</sup> ±30  | 3232.0 <sup>bcd</sup> ±33 | 1929.0ª±4               | 15520.0°±20               | 6021.0 <sup>f</sup> ±6  | 0.000 |  |  |
| 14            | 22570 <sup>d</sup> ±30  | 4514.0 <sup>f</sup> ±39   | 2010.0 <sup>ab</sup> ±4 | 10160.0ª±20               | 3450.0 <sup>a</sup> ±4  | 0.000 |  |  |
| 15            | 25125e±21               | 4271.5 <sup>ef</sup> ±37  | $2000.5^{ab}\pm4$       | 10050.5ª±20               | 4085.0 <sup>b</sup> ±4  | 0.000 |  |  |
| 18            | 13525ª±22               | 2749.5 <sup>abc</sup> ±36 | 2317.5°±4               | 14720.0bc±30              | 3411.5 <sup>a</sup> ±5  | 0.000 |  |  |
| 20            | 19030°±20               | 2528.0 <sup>ab</sup> ±31  | 2436.0 <sup>d</sup> ±5  | 12140.0 <sup>ab</sup> ±20 | 5026.0°±5               | 0.000 |  |  |
| 21            | 16480 <sup>b</sup> ±20  | 2110.0 <sup>a</sup> ±28   | 2307.0°±4               | 23050.0 <sup>d</sup> ±30  | 4439.0°±5               | 0.000 |  |  |

Table 3. Mean values of macro nutrient and Duncan test results on tree diameter basis

| Tree             | MAKRONUTRIENTS (ppm)    |                          |                        |                        |                       |            |  |  |  |
|------------------|-------------------------|--------------------------|------------------------|------------------------|-----------------------|------------|--|--|--|
| Diameter<br>(cm) | Ca                      | Mg                       | Р                      | К                      | S                     | <b>p</b> * |  |  |  |
| 20               | 21150 <sup>d</sup> ±20  | 3718 <sup>cd</sup> ±36   | 2201.0 <sup>e</sup> ±4 | 12190°±20              | 4583 <sup>bc</sup> ±5 | 0.000      |  |  |  |
| 22               | 20610 <sup>cd</sup> ±20 | 3152 <sup>bc</sup> ±33   | 2254.0 <sup>g</sup> ±4 | 13620 <sup>d</sup> ±20 | 4717 <sup>cd</sup> ±5 | 0.000      |  |  |  |
| 30               | 24580 <sup>ef</sup> ±30 | 4320.5 <sup>de</sup> ±33 | 2083.5 <sup>d</sup> ±5 | 13635 <sup>d</sup> ±20 | 4248 <sup>b</sup> ±5  | 0.000      |  |  |  |
| 33               | 22570 <sup>de</sup> ±30 | 4514 <sup>e</sup> ±39    | 2010.0°±4              | 10160 <sup>b</sup> ±20 | 3450ª±4               | 0.000      |  |  |  |
| 46               | 23280e±30               | 3462°±34                 | 1909.0ª±4              | 7931ª±18               | 4336 <sup>b</sup> ±5  | 0.000      |  |  |  |
| 53               | $16480^{b}\pm 20$       | 2110 <sup>a</sup> ±28    | 2307.0 <sup>h</sup> ±4 | 23050g±30              | 4439 <sup>bc</sup> ±5 | 0.000      |  |  |  |
| 65,5             | 19030°±20               | 2528 <sup>ab</sup> ±31   | 2436.0 <sup>j</sup> ±5 | 12140°±20              | 5026 <sup>d</sup> ±5  | 0.000      |  |  |  |
| 67               | $26470^{f}\pm 30$       | 3232°±33                 | 1929.0 <sup>b</sup> ±4 | 15520 <sup>e</sup> ±20 | 6021°±6               | 0.000      |  |  |  |
| 101              | 15800 <sup>b</sup> ±20  | 3229°±33                 | 2216.0 <sup>f</sup> ±4 | 11970°±20              | 3521ª±4               | 0.000      |  |  |  |
| 104              | $11250^{a}\pm 20$       | 2270ª±30                 | 2419.0 <sup>i</sup> ±5 | $17470^{f}\pm 30$      | 3302ª±4               | 0.000      |  |  |  |

The concentrations of Fe, Mn, Na, Cu, Zn, Cl, Al and Co in the needles of cedar trees of different heights are given in Table 4 and Table 5. Statistically, the amounts of micronutrients contained in the needles showed significant differences between tree height and tree diameter (p<0.05). While Fe and Co concentrations were highest for the 14 m height and 33 cm diameter in cedar needles, Fe concentrations were lowest for the 20 m height and 65,5 cm diameter in cedar needles and Co concentrations were lowest for the 9 m height and 22 cm diameter in cedar needles. The highest Mn concentrations were for the 9 m height and 22 cm diameter in cedar needles of 21 m height and

53 cm diameter. While the highest Na concentration was for the 21 m height and 53 cm diameter in cedar needles, the lowest Cu, Zn, Cl concentrations were for the 12 m height and 67 cm diameter cedar needles. Al concentrations of 20 m long and 53 cm diameter cedar needles were low values while Al concentrations were highest in cedar meedles of 15 m height and 30 cm diameter (Table 4 and 5). When tree heights were grouped between 8-15 m and 18-21 m and tree diameters between 20-46 cm and 53-104 cm, the average of Ca, Mg, Fe, Mn, Cu, Zn and Al concentrations decreased with increasing tree height and diameter. P, K and Na concentrations increased with increasing tree height and diameter. As the tree diameter increased, concentration of S increased and Cl and Co concentrations decreased. As the tree height increased, concentration of S decreased.

| Tree          | MICRO NUTRIENTS (ppm)      |                            |                        |                          |                         |                            |                         |                           |       |
|---------------|----------------------------|----------------------------|------------------------|--------------------------|-------------------------|----------------------------|-------------------------|---------------------------|-------|
| Height<br>(m) | Fe                         | Mn                         | Na                     | Cu                       | Zn                      | Cl                         | Al                      | Со                        | p*    |
| 8             | $681.40^{abc}{\pm}2.4$     | 72.40°±0.5                 | $1220^{bc} \pm 127$    | $9.3^{cd}\pm0.3$         | 35.40 <sup>g</sup> ±0.3 | 2745.0°±3                  | $1158^{abc}\pm 8$       | 20.70 <sup>abc</sup> ±2.1 | 0.000 |
| 9             | 594.00 <sup>abc</sup> ±2.2 | 109.70 <sup>g</sup> ±0.6   | 900 <sup>ab</sup> ±121 | 10.1 <sup>de</sup> ±0.3  | 25.70°±0.3              | 1757.0 <sup>abc</sup> ±2   | $1100^{abc} \pm 7$      | 17.70 <sup>a</sup> ±1.9   | 0.000 |
| 10            | 702.00 <sup>abc</sup> ±2.4 | $92.00^{\rm f}{\pm}0.6$    | 830 <sup>a</sup> ±115  | 11.2°±0.3                | 26.90 <sup>f</sup> ±0.3 | 1316.0 <sup>ab</sup> ±2    | $1036^{abc}\pm7$        | 21.60 <sup>abc</sup> ±2.1 | 0.000 |
| 12            | 481.10 <sup>ab</sup> ±2.0  | $50.60^d \pm 0.4$          | 1310 <sup>c</sup> ±121 | 5.7ª±0.3                 | 18.10ª±0.2              | 961.57ª±4                  | 793 <sup>ab</sup> ±6    | 19.10 <sup>ab</sup> ±1.9  | 0.000 |
| 14            | 848.40°±2.7                | 49.60 <sup>cd</sup> ±0.4   | $2040^d\pm132$         | $8.8^{cd}\!\!\pm\!\!0.3$ | 22.00°±0.3              | 2673.0°±3                  | $1439^{bc}\!\!\pm\!\!9$ | 25.00°±2.1                | 0.000 |
| 15            | 789.58 <sup>bc</sup> ±2.6  | $42.15^{bc}\pm 0.8$        | 1510°±126              | $9.0^{cd}\pm0.3$         | $24.60^{d}\pm0.3$       | $1364.5^{ab}\pm 3$         | 1557°±5                 | 23.43 <sup>bc</sup> ±2.5  | 0.000 |
| 18            | 517.15 <sup>ab</sup> ±2.5  | $36.50^{ab}\!\!\pm\!\!0.5$ | 2275 <sup>d</sup> ±126 | 7.95 <sup>bc</sup> ±0.3  | 20.60 <sup>b</sup> ±0.3 | $2385.0^{bc}\!\!\pm\!\!3$  | 795.15 <sup>ab</sup> ±5 | 23.25 <sup>bc</sup> ±2.5  | 0.000 |
| 20            | 403.40 <sup>a</sup> ±1.8   | 43.90 <sup>bcd</sup> ±0.4  | $1160^{abc}{\pm}127$   | $8.3^{bc}\!\!\pm\!\!0.3$ | 25.60°±0.3              | $1884.0^{abc}\!\!\pm\!\!2$ | 682.4ª±5                | 18.90 <sup>ab</sup> ±1.9  | 0.000 |
| 21            | 615.77 <sup>abc</sup> ±2.8 | 32.80ª±0.4                 | 3430 <sup>e</sup> ±127 | $7.1^{ab}\!\!\pm\!\!0.3$ | 22.00°±0.3              | 1375.0 <sup>ab</sup> ±2    | 933.6 <sup>abc</sup> ±6 | $23.90^{bc}\pm 2.0$       | 0.000 |

Tablo 4. Mean values of micro nutrients and Duncan test results on tree height basis

| Table 5. Mean values of micro nutrient and Duncan test results on tree dian | neter basis |
|---|-------------|
|---|-------------|

| MICRO NUTRIENTS (ppm)               |  |   |   |   |   |  |   |   |
|-------------------------------------|--|---|---|---|---|--|---|---|
| Fe                                  | Mn   | Na  | Cu  | Zn  | Cl  | Al   | Со  | p*  |
| $681.4^{\text{cd}}{\pm}2.4$         | 72.4 <sup>b</sup> ±0.5   | 1220ª±127   | 9.3 <sup>g</sup> ±0.17  | $35.4^{f}\pm0.17$   | 2745 <sup>d</sup> ±3  | $1158^{abc}\!\!\pm\!\!8$   | 20.7 <sup>ab</sup> ±2.1   | 0.000   |
| 594.0 <sup>abc</sup> ±2.2           | 109.7°±0.6   | 900 <sup>a</sup> ±121   | 10.1 <sup>h</sup> ±0.17   | 25.7°±0.17  | 1757°±2   | $1100^{abc} \pm 7$   | 17.7 <sup>a</sup> ±1.9  | 0.000   |
| 921.33°±2.3                         | 71.55 <sup>b</sup> ±0.4  | 1210ª±126   | $10.95^{i}\pm0.16$  | 26.1°±0.17  | 1516,5 <sup>b</sup> ±2  | 1655°±8  | 20.6 <sup>ab</sup> ±2.1   | 0.000   |
| $848.4^{\text{de}} \!\!\pm\!\! 2.7$ | 49.6ª±0.4  | 2040 <sup>b</sup> ±132  | $8.8^{\text{fg}}{\pm}0.17$  | 22.0°±0.17  | 2673 <sup>d</sup> ±3  | $1439^{bc}\!\!\pm\!\!9$  | 25.0°±2.1   | 0.000   |
| 438.5 <sup>ab</sup> ±1.9            | 33.2ª±0.4  | 1430 <sup>a</sup> ±121  | $7.3^{bc} \pm 0.17$   | 23.9 <sup>d</sup> ±0.17   | 1012ª±1   | 840ª±6   | 27.3°±1.9   | 0.000   |
| 615.77 <sup>bc</sup> ±2.8           | 32.8ª±0.4  | 3430°±127   | 7.1 <sup>b</sup> ±0.17  | 22.0°±0.17  | 1375 <sup>b</sup> ±2  | 933.6 <sup>ab</sup> ±6   | 23.9 <sup>bc</sup> ±2.0   | 0.000   |
| 403.4ª±1.8                          | 43.9ª±0.4  | 1160 <sup>a</sup> ±127  | $8.3^{\text{ef}} \!\!\pm\! 0.17$  | 25.6 <sup>e</sup> ±0.17   | 1884°±2   | 682.4ª±5   | 18.9 <sup>a</sup> ±1.9  | 0.000   |
| 481.1 <sup>abc</sup> ±2.0           | 50.6ª±0.4  | 1310 <sup>a</sup> ±121  | 5.7ª±0.17   | 18.1ª±0.17  | 961.57ª±1   | 793ª±6   | 19.1ª±1.9   | 0.000   |
| 616.2 <sup>bc</sup> ±2.3            | 39.7ª±0.4  | 2380 <sup>b</sup> ±127  | $8.2^{de}\pm0.17$   | 19.9 <sup>b</sup> ±0.17   | 1089ª±2   | 927.6 <sup>ab</sup> ±6   | 26.0°±2.1   | 0.000   |
| 418.1 <sup>ab</sup> ±1.9            | 33.3ª±0.4  | 2170 <sup>b</sup> ±138  | $7.7^{cd} \pm 0.17$   | 21.3°±0.17  | 3681°±3   | 662.7ª±5   | 20.5 <sup>ab</sup> ±2.0   | 0.000   |
|                                     | $\begin{tabular}{ c c c c c } \hline Fe \\ \hline 681.4^{cd} \pm 2.4 \\ \hline 594.0^{abc} \pm 2.2 \\ 921.33^{e} \pm 2.3 \\ 848.4^{dc} \pm 2.7 \\ 438.5^{ab} \pm 1.9 \\ 615.77^{bc} \pm 2.8 \\ 403.4^{a} \pm 1.8 \\ 481.1^{abc} \pm 2.0 \\ 616.2^{bc} \pm 2.3 \\ 418.1^{ab} \pm 1.9 \\ \hline \end{tabular}$ | Fe         Mn           681.4 <sup>cd</sup> ±2.4         72.4 <sup>b</sup> ±0.5           594.0 <sup>abc</sup> ±2.2         109.7 <sup>c</sup> ±0.6           921.33 <sup>e</sup> ±2.3         71.55 <sup>b</sup> ±0.4           848.4 <sup>de</sup> ±2.7         49.6 <sup>a</sup> ±0.4           438.5 <sup>ab</sup> ±1.9         33.2 <sup>a</sup> ±0.4           615.77 <sup>bc</sup> ±2.8         32.8 <sup>a</sup> ±0.4           403.4 <sup>a</sup> ±1.8         43.9 <sup>a</sup> ±0.4           481.1 <sup>abc</sup> ±2.0         50.6 <sup>a</sup> ±0.4           616.2 <sup>bc</sup> ±2.3         39.7 <sup>a</sup> ±0.4           418.1 <sup>ab</sup> ±1.9         33.3 <sup>a</sup> ±0.4 | Fe         Mn         Na           681.4 <sup>cd</sup> ±2.4         72.4 <sup>b</sup> ±0.5         1220 <sup>a</sup> ±127           594.0 <sup>abc</sup> ±2.2         109.7 <sup>c</sup> ±0.6         900 <sup>a</sup> ±121           921.33 <sup>a</sup> ±2.3         71.55 <sup>b</sup> ±0.4         1210 <sup>a</sup> ±126           848.4 <sup>de</sup> ±2.7         49.6 <sup>a</sup> ±0.4         2040 <sup>b</sup> ±132           438.5 <sup>ab</sup> ±1.9         33.2 <sup>a</sup> ±0.4         1430 <sup>a</sup> ±121           615.77 <sup>bc</sup> ±2.8         32.8 <sup>a</sup> ±0.4         3430 <sup>c</sup> ±127           403.4 <sup>a</sup> ±1.8         43.9 <sup>a</sup> ±0.4         1160 <sup>a</sup> ±127           481.1 <sup>abc</sup> ±2.0         50.6 <sup>a</sup> ±0.4         1310 <sup>a</sup> ±121           616.2 <sup>bc</sup> ±2.3         39.7 <sup>a</sup> ±0.4         2380 <sup>b</sup> ±127           418.1 <sup>ab</sup> ±1.9         33.3 <sup>a</sup> ±0.4         2170 <sup>b</sup> ±138 | Fe         Mn         Na         Cu           681.4 <sup>cd</sup> ±2.4         72.4 <sup>b</sup> ±0.5         1220 <sup>a</sup> ±127         9.3 <sup>g</sup> ±0.17           594.0 <sup>abc</sup> ±2.2         109.7 <sup>c</sup> ±0.6         900 <sup>a</sup> ±121         10.1 <sup>h</sup> ±0.17           921.33 <sup>a</sup> ±2.3         71.55 <sup>b</sup> ±0.4         1210 <sup>a</sup> ±126         10.95 <sup>i</sup> ±0.16           848.4 <sup>de</sup> ±2.7         49.6 <sup>a</sup> ±0.4         2040 <sup>b</sup> ±132         8.8 <sup>fg</sup> ±0.17           438.5 <sup>ab</sup> ±1.9         33.2 <sup>a</sup> ±0.4         1430 <sup>a</sup> ±121         7.3 <sup>bc</sup> ±0.17           615.77 <sup>bc</sup> ±2.8         32.8 <sup>a</sup> ±0.4         3430 <sup>c</sup> ±127         7.1 <sup>b</sup> ±0.17           403.4 <sup>a</sup> ±1.8         43.9 <sup>a</sup> ±0.4         1160 <sup>a</sup> ±127         8.3 <sup>cf</sup> ±0.17           41.1 <sup>abc</sup> ±2.0         50.6 <sup>a</sup> ±0.4         1310 <sup>a</sup> ±121         5.7 <sup>a</sup> ±0.17           616.2 <sup>bc</sup> ±2.3         39.7 <sup>a</sup> ±0.4         2380 <sup>b</sup> ±127         8.2 <sup>dc</sup> ±0.17           418.1 <sup>ab</sup> ±1.9         33.3 <sup>a</sup> ±0.4         2170 <sup>b</sup> ±138         7.7 <sup>cd</sup> ±0.17 | Fe         Mn         Na         Cu         Zn           681.4 <sup>cd</sup> ±2.4         72.4 <sup>b</sup> ±0.5         1220 <sup>a</sup> ±127         9.3 <sup>s</sup> ±0.17         35.4 <sup>f</sup> ±0.17           594.0 <sup>abc</sup> ±2.2         109.7 <sup>c</sup> ±0.6         900 <sup>a</sup> ±121         10.1 <sup>b</sup> ±0.17         25.7 <sup>c</sup> ±0.17           921.33 <sup>a</sup> ±2.3         71.55 <sup>b</sup> ±0.4         1210 <sup>a</sup> ±126         10.95 <sup>i</sup> ±0.16         26.1 <sup>c</sup> ±0.17           848.4 <sup>de</sup> ±2.7         49.6 <sup>a</sup> ±0.4         2040 <sup>b</sup> ±132         8.8 <sup>fs</sup> ±0.17         22.0 <sup>c</sup> ±0.17           438.5 <sup>ab</sup> ±1.9         33.2 <sup>a</sup> ±0.4         1430 <sup>a</sup> ±121         7.3 <sup>bc</sup> ±0.17         23.9 <sup>d</sup> ±0.17           615.77 <sup>bc</sup> ±2.8         32.8 <sup>a</sup> ±0.4         3430 <sup>c</sup> ±127         7.1 <sup>b</sup> ±0.17         22.0 <sup>c</sup> ±0.17           403.4 <sup>a</sup> ±1.8         43.9 <sup>a</sup> ±0.4         160 <sup>a</sup> ±127         8.3 <sup>ef</sup> ±0.17         25.6 <sup>e</sup> ±0.17           481.1 <sup>abc</sup> ±2.0         50.6 <sup>a</sup> ±0.4         1310 <sup>a</sup> ±121         5.7 <sup>a</sup> ±0.17         18.1 <sup>a</sup> ±0.17           616.2 <sup>bc</sup> ±2.3         39.7 <sup>a</sup> ±0.4         2380 <sup>b</sup> ±127         8.2 <sup>de</sup> ±0.17         19.9 <sup>b</sup> ±0.17           418.1 <sup>ab±1.9</sup> 33.3 <sup>a</sup> ±0.4         2170 <sup>b</sup> ±138         7.7 <sup>cd</sup> ±0.17         21.3 <sup>c</sup> ±0.17 | Fe         Mn         Na         Cu         Zn         Cl           681.4 <sup>cd</sup> ±2.4         72.4 <sup>b</sup> ±0.5         1220 <sup>a</sup> ±127         9.3 <sup>g</sup> ±0.17         35.4 <sup>f</sup> ±0.17         2745 <sup>d</sup> ±3           594.0 <sup>abc</sup> ±2.2         109.7 <sup>c</sup> ±0.6         900 <sup>a</sup> ±121         10.1 <sup>h</sup> ±0.17         25.7 <sup>e</sup> ±0.17         1757 <sup>c</sup> ±2           921.33 <sup>e</sup> ±2.3         71.55 <sup>b</sup> ±0.4         1210 <sup>a</sup> ±126         10.95 <sup>i</sup> ±0.16         26.1 <sup>e</sup> ±0.17         1516,5 <sup>b</sup> ±2           848.4 <sup>de</sup> ±2.7         49.6 <sup>a</sup> ±0.4         2040 <sup>b</sup> ±132         8.8 <sup>fg</sup> ±0.17         22.0 <sup>c</sup> ±0.17         2673 <sup>d</sup> ±3           438.5 <sup>ab</sup> ±1.9         33.2 <sup>a</sup> ±0.4         1430 <sup>a</sup> ±121         7.3 <sup>bc</sup> ±0.17         23.9 <sup>d</sup> ±0.17         1012 <sup>a</sup> ±1           615.77 <sup>bc</sup> ±2.8         32.8 <sup>a</sup> ±0.4         3430 <sup>c</sup> ±127         7.1 <sup>b</sup> ±0.17         22.0 <sup>c</sup> ±0.17         1375 <sup>b</sup> ±2           403.4 <sup>a</sup> ±1.8         43.9 <sup>a</sup> ±0.4         1160 <sup>a</sup> ±127         8.3 <sup>ef</sup> ±0.17         25.6 <sup>e</sup> ±0.17         1884 <sup>c</sup> ±2           413.1 <sup>ab</sup> ±2.0         50.6 <sup>a</sup> ±0.4         1310 <sup>a</sup> ±121         5.7 <sup>a</sup> ±0.17         18.1 <sup>a</sup> ±0.17         961.57 <sup>a</sup> ±1           616.2 <sup>bc</sup> ±2.3         39.7 <sup>a</sup> ±0.4         2380 <sup>b</sup> ±127         8.2 <sup>de</sup> ±0.17         19.9 <sup>b</sup> ±0.17         1089 <sup>a</sup> ±2           418.1 <sup>ab</sup> ±1.9         33.3 <sup>a</sup> ±0.4         2170 <sup>b</sup> ±138         7.7 <sup>cd</sup> ±0.17         21.3 <sup>c</sup> | MICEO NUTRIENTS (ppm)           Fe         Mn         Na         Cu         Zn         Cl         Al           681.4 <sup>cd</sup> ±2.4         72.4 <sup>b</sup> ±0.5         1220 <sup>a</sup> ±127         9.3 <sup>s</sup> ±0.17         35.4 <sup>f</sup> ±0.17         2745 <sup>d</sup> ±3         1158 <sup>abc</sup> ±8           594.0 <sup>abc</sup> ±2.2         109.7 <sup>c</sup> ±0.6         900 <sup>a</sup> ±121         10.1 <sup>h</sup> ±0.17         25.7 <sup>e</sup> ±0.17         1757 <sup>c</sup> ±2         1100 <sup>abc</sup> ±7           921.33 <sup>a</sup> ±2.3         71.55 <sup>h</sup> ±0.4         1210 <sup>a</sup> ±126         10.95 <sup>i</sup> ±0.16         26.1 <sup>e</sup> ±0.17         1516,5 <sup>b</sup> ±2         1655 <sup>c</sup> ±8           848.4 <sup>de</sup> ±2.7         49.6 <sup>a</sup> ±0.4         2040 <sup>b</sup> ±132         8.8 <sup>fe</sup> ±0.17         22.0 <sup>c</sup> ±0.17         2673 <sup>d</sup> ±3         1439 <sup>bc</sup> ±9           438.5 <sup>ab</sup> ±1.9         33.2 <sup>a</sup> ±0.4         1430 <sup>a</sup> ±121         7.3 <sup>bc</sup> ±0.17         22.0 <sup>c</sup> ±0.17         1012 <sup>a</sup> ±1         840 <sup>a</sup> ±6           615.77 <sup>bc</sup> ±2.8         32.8 <sup>a</sup> ±0.4         3430 <sup>c</sup> ±127         7.1 <sup>b</sup> ±0.17         22.0 <sup>c</sup> ±0.17         1375 <sup>b</sup> ±2         93.6 <sup>ab</sup> ±6           403.4 <sup>a</sup> ±1.8         43.9 <sup>a</sup> ±0.4         160 <sup>a</sup> ±127         7.1 <sup>b</sup> ±0.17         22.0 <sup>c</sup> ±0.17         1884 <sup>c</sup> ±2         682.4 <sup>a</sup> ±5           481.1 <sup>abc</sup> ±2.0         50.6 <sup>a</sup> ±0.4         1310 <sup>a</sup> ±121         5.7 <sup>a</sup> ±0.17         18.1 <sup>a</sup> ±0.17         961.57 <sup>a±1</sup> 793 <sup>a</sup> ±6           616.2 <sup>bc</sup> ± | Fe         Mn         Na         Cu         Zn         Cl         Al         Co           681.4 <sup>cd</sup> ±2.4         72.4 <sup>b</sup> ±0.5         1220 <sup>a</sup> ±127         9.3 <sup>c</sup> ±0.17         35.4 <sup>f</sup> ±0.17         2745 <sup>d</sup> ±3         1158 <sup>abc</sup> ±8         20.7 <sup>ab</sup> ±2.1           594.0 <sup>abc</sup> ±2.2         109.7 <sup>c</sup> ±0.6         900 <sup>a</sup> ±121         10.1 <sup>h</sup> ±0.17         25.7 <sup>c</sup> ±0.17         1757 <sup>c</sup> ±2         1100 <sup>abc</sup> ±7         17.7 <sup>a</sup> ±1.9           921.33 <sup>a</sup> ±2.3         71.55 <sup>b</sup> ±0.4         1210 <sup>a</sup> ±126         10.95 <sup>i</sup> ±0.16         26.1 <sup>e</sup> ±0.17         1516.5 <sup>b</sup> ±2         1655 <sup>c</sup> ±8         20.6 <sup>ab</sup> ±2.1           848.4 <sup>de</sup> ±2.7         49.6 <sup>a</sup> ±0.4         2040 <sup>b</sup> ±132         8.8 <sup>f</sup> ±0.17         22.0 <sup>c</sup> ±0.17         2673 <sup>d</sup> ±3         1439 <sup>bc</sup> ±9         25.0 <sup>c</sup> ±2.1           438.5 <sup>ab</sup> ±1.9         33.2 <sup>a</sup> ±0.4         1430 <sup>a</sup> ±121         7.3 <sup>bc</sup> ±0.17         23.9 <sup>d</sup> ±0.17         1012 <sup>a</sup> ±1         840 <sup>a</sup> ±6         27.3 <sup>c</sup> ±1.9           615.77 <sup>bc</sup> ±2.8         32.8 <sup>a</sup> ±0.4         3430 <sup>c</sup> ±127         7.1 <sup>b</sup> ±0.17         22.0 <sup>c</sup> ±0.17         1375 <sup>b</sup> ±2         933.6 <sup>ab</sup> ±6         23.9 <sup>b</sup> ±2.0           403.4 <sup>a</sup> ±1.8         43.9 <sup>a</sup> ±0.4         160 <sup>a</sup> ±127         8.3 <sup>cf</sup> ±0.17         25.0 <sup>c</sup> ±0.17         1884 <sup>c</sup> ±2         682.4 <sup>a</sup> ±5         18.9 <sup>a</sup> ±1.9           433.4 <sup>a</sup> ±1.8         43.9 <sup>a</sup> ±0.4         1106 <sup>a</sup> ±127 |

#### Discussion

In our study, while Ca, Mg, Fe, Mn, Cu, Zn and Al concentrations decreased in cedar needles depending on tree height and diameter, P, K, Na concentrations increased (Table 3 and 4). Co and Cl concentrations decreased depending on tree height and increased with tree diameter (Table 4 and 5). Wang and Klinka (1997) found that the concentrations of N, P and K in spruce needle showed a negative correlation with stand age and indicated a positive correlation with tree height and diameter growth. Askew (1937) reported that the concentration of Ca, P, Na and Cl in the needles of P. radiata species decreased and the concentrations of N, Mg, K and Mn were higher in young needles. Emmert (1957) observed that the concentrations were observed. N, P, Ca, S and Fe concentrations decreased while K and Mn concentrations in *Agathis australis* species increased with age (Peterson 1961).

In this study, significant differences were observed between macro nutrients and micro nutrients in the needles due to the increase in tree height and tree diameter. Three factors could have been effective in these differences. First, the major change in the age of cedar trees can cause a large change in the rate of tree growth. Therefore, it affects the absorption of nutrients taken in the same soil. Secondly, the fact that approximately 3 cedar trees in the study area is dominated by trees with a diameter of 100-150 years can be effective. Depending on the progress of tree age, it is likely that both height and diameter growth rates will decrease. Wang and Klinka (1997) have stated that the annual height increase rate of spruce

trees decreases the tree diameter and height increase with the progress of tree age. Therefore, growth in old trees may not be as sensitive as nutrient demand in young trees. Third, the relationships between tree growth and leaf nutrient concentrations may be effective for other important growth-limiting factors (eg soil moisture, aeration, etc.).

When we look at the findings in our study for micro nutrients, Cu concentration decreased with tree age and tree height. Cu concentration is a small trace metal found in chloroplast of plants and containing about 70% copper in leaves (Wilkinson, 1994). Kabata-Pendias and Piotrowska (1984) reported that the normal Cu content in the plants ranged from 2 to 20 ppm. However, in most plants normal Cu contents are in the range of 4-12 ppm. Our results showed that the Cu content was in the specified range. Zn concentration is an important element in all organisms and plays an important role in the synthesis of enzymes, auxins and some proteins. Plants with symptoms of Zn deficiency cause prolonged delayed cells. A critical Zn toxic level in the leaves is about 100 ppm in the dry plant material ((Allen et al., 1974; Yılmaz and Zengin, 2003). The cobalt content of the plant is related to the beneficial cobalt compound in the soil, plant species and plant tissue (Güneş et al., 2004). The toxic limit of the Co concentration for the leaves is about 0.5 ppm (Pendias and Pendias, 1992). In our study, Co content has more than 0.5 ppm and limit overrun levels. Güneş et al. (2004) reported that the co concentration was washed in the winter between autumn and spring.

#### 4. CONCLUSION

The examination of the accumulation of nutrients in forest ecosystems will be very useful for gathering information on the nutrient dynamics of the ecosystem. It can also provide information on the effects of forest management practices and trees against the physiological status of abiotic and biotic stress factors on the efficiency of forest ecosystem balance and forest soils quality. In particular, it is necessary to determine the content of leaf/needle and subsoil nutrient content among other tree species and in cedar trees growing in water and nutrient competition between plant and soil.

#### REFERENCES

- Allen S E, Grimshow H M, Parkinson J A & Quarmby C (1974) Chemical Analysis of Ecological Materials. Blackwell Scientific Publications, Osney Mead, Oxford, UK.
- Askew H O (1937) The Chemical Composition of Pinus radiata Needles. N. Z. J. Sci. Technol. 18: 651-655.
- Avsar M D (2004) The Relationships between Diameter at Breast Height, Tree Height and Crown Diameter in Calabrian pines [*Pinus brutia* Ten.] of Baskonus Mountain, Kahramanmaras, Turkey. Journal of Biological Sciences, 4(4): 437-440.
- Binkl D & Vitousek P M (1989) Soil Nutrient Availability. In: (Eds.): Pearey R W, J Ehleringer, N A Mooney, P W Rundel. Plant Physiological Field Methods and Instrumentation London; Chapman and Hall, 75-96.
- Emmert F H (1957) A Comparison of Different Leaf Samples and the Total and Soluble Tests as Indicators of Apple Tree Nitrogen, Potassium and Magnesium Nutrition. In Proc. Am. Soc. Hort. Sci 69: 1-12.
- Güneş A, Alpaslan M & Inal A (2004) Plant Nutrition and Fertilizer. Ankara University, Agriculture Publication No: 1539, Ankara, Turkey.
- Heilman P E & Gessel S P (1963) The Effect of Nitrogen Fertilization on the Concentration and Weight of Nitrogen, Phosphorus and Potassium in Douglas-fir Trees. Soil Sci. Soc. Amer. Proc. 27: 102-105.
- Howlett F S & Cahoon G A (1964) Leaf Analysis. Ohio Report on Res. and Dev. 49: 67-68.
- Ingestad T (1967) Methods for Uniform Optimum Fertilization of Forest Tree Plants. Proc. 14th IUFRO Congress, 3: 265-269.
- Kabata-Pendias A, & Piotrowska M (1984) Zanieczyszczenie Glebi Roslin Uprawnych Pierwiastkami Sladowymi. CBRopracowanie problemowe, Warszawa, Poland.
- Karelin D V, Lyuri D I, Goryachkin S V, Lunin V N & Kudikov A V (2015) Changes in the Carbon dioxide Emission from Soils in the Course of Postagrogenic Succession in the Chernozems Forest-steppe. Eurasian soil science, *48*(11): 1229-1241.
- Kiser L, Fox T & Carlson C (2013) Foliage and Litter Chemistry, Decomposition and Nutrient Release in *Pinus taeda*. Forests, 4(3): 595-612.
- Mead D J (1984) Diagnosis of Nutrient Deficiencies in Plantations. 259-291. In G D Bowen & E K S Nambiar (eds.) Nutrition of Plantation Forests. Academic Press, London, England.
- Milosavljevic I, Esser A D & Crowder D W (2016) Effects of Environmental and Agronomic Factors on Soil-dwelling Pest Communities in Cereal Crops. Agriculture, Ecosystems and Environment, 225: 192-198.
- Mitchell A K, Barclay H J, Brix H, Pollard D F W, Benton R & DeJong R (1996) Biomass and Nutrient Element Dynamics in Douglasfir: Effects of Thinning and Nitrogen Fertilization over 18 Years. Canadian Journal of Forest Research, 26(3): 376-388.
- Moore J A, Mika P G, Shaw T M & Garrison-Johnston M I (2004) Foliar Nutrient Characteristics of Four Conifer Species in the Interior Northwest United States. West. J. Appl. For., 19: 13-24.

- Oleksyn J, Reich P B, Zytkowiak R, Karolewski P & Tjoelker M G (2002) Needle Nutrients in Geographically Diverse *Pinus sylvestris* populations. Ann For Sci 59: 1-18.
- Pendias K & Pendias H (1992) Trace Elements in Soil and Plant. CRC Press, Boca Raton.
- Peterson P J (1961) Variation in the Mineral Content of kauri (*Agathis australis* Salisb.) Leaves with Respect to Leaf Age, Leaf Position and Tree Age. *NZJ Sci*, 4: 669-678.
- Powers R F (1984) Estimating Soil Nitrogen Availability Through Soil and Foliar Analysis. 353-379. In E. L. Stone (ed.) Forest Soils & Treatment Impacts. Sixth North American Forest Soils Conference Proceedings, Knoxville, TN, June 1983. Univ. of Tenn., Knoxville, TN. 454 p.
- Raison R J, Myers B J & Benson M L (1992) Dynamics of *Pinus radiata* Foliage in Relation to Water and Nitrogen Stress: I. Needle Production and Properties. Forest Ecology and Management, 52(1-4): 139-158.
- Raitio H & Sarjala T (2000) Effect of Provenance on Free Amino Acid and Chemical Composition of Scots pine Needles. Planta 221: 231-238
- Ruark G A & Bockheim J G (1988) Biomass, Net Primary Production and Nutrient Distribution for an Age Sequence of *Populus tremuloides* Ecosystems. Canadian Journal of Forest Research, 18(4): 435-443.
- Sasser C L & Binkley D (1989) Nitrogen Mineralization in High-elevation Forests of the Appalachians. II. Patterns with stand development in fir waves. Biogeochemistry, 7(2): 147-156.
- Selcuk A R, Demiray E & Yilmaz Y (2011) Antioxidant Activity of Grape Seeds Obtained from Molasses [Pekmez] and Winery Production. Academic Food Journal/Akademik GIDA.
- Sheikh M A & Kumar M (2010) Nutrient Status and Economic Analysis in Oak and Pine Forests in Garhwal Himalaya. J. Amer. Sci., 6: 117-122.
- Sprugel D G (1984) Density, Biomass, Productivity and Nutrient-cycling Changes During Stand development in wave-regenerated balsam fir forests. Ecological Monographs, 54(2): 165-186.
- Unger D R, Hung I K, Brooks R & Williams H (2014) Estimating Number of Trees, Tree Height and Crown Width Using Lidar Data. GIScience & remote sensing, 51(3): 227-238.
- URL-1 (2019) http://www.kastamonukultur.gov.tr/TR-169990/iklim-ve-bitki-ortusu.html adresine 31/03/2019 tarihinde erişilmiştir.
- Wang G G & Klinka K (1997) White Spruce Foliar Nutrient Concentrations in Relation to Tree Growth and Soil Nutrient Amounts. Forest Ecology and Management, 98(1): 89-99.
- Wang J R, Zhong A L, Simard S W & Kimmins J P (1996) Aboveground Biomass and Nutrient Accumulation in an Age Sequence of Paper Birch [*Betula papyrifera*] in the Interior Cedar Hemlock Zone, British Columbia. Forest ecology and management, 83(1-2): 27-38.
- Wilkinson R E (1994) Plant–Environment Interaction. Marcel Dekker, New York, 559pp.
- Yılmaz S & Zengin M (2003) Monitoring Environmental Pollution in Erzurum by Chemical Analysis of Scots Pine [*Pinus sylvestris* L.] Needles. Environment International 1097: 1-7.



# Horticulture Practice of Medicinal Plants in Kastamonu

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**Abstract:** Alternative products are important sources of rural people income. In order to grow non-ordinary agricultural products, production conditions must be investigated. The results of these studies generate new sources of income for people living in the countryside. There are two ways to discovery the sources of unusual target products. The first way is the adaptation of the exotic species to the ecological conditions of our country, and the second way is the determination of the available resource values in our flora. During the climatic conditions in Kastamonu, medicinal plant production practice were carried out between 2002-2014 and some target species were identified. For mass production of target species, land was rented, seedling greenhouse was established, electricity and irrigation infrastructure was established. After planting, plant seedlings were produced. The study started in 2014 in Kastamonu Gölköy is still ongoing. The first three years were harvested by hand. After the fourth year, the method of harvesting with the machine was used. The harvested plants were dried by natural methods in 90% shading. Dried products are placed in cloth bags and stored in containers. Dried plants are marketed either in wholesale or retail market. In addition, some plants were extracted by volatilization by water vapor distillation method and analyzed by GC-MS. The species produced are *Salvia officinalis, Melissa officinalis, Origanum onites, Echinacea purpurea, Thymus vulgaris, Mentha piperita, Matricaria chamomilla, Rheum rhabarbatum, Hypericum perforatum.* 

**Keywords:** Horticulture, Medicinal Plants, Salvia officinalis, Melissa officinalis, Origanum onites, Echinacea purpurea, Thymus vulgaris, Mentha piperita, Matricaria chamomilla, Rheum rhabarbatum, Hypericum perforatum

# **1. INTRODUCTION**

In this study, the process of the transformation of ecological information into economic value is explained. This application is important in terms of creating a rural development model. Alternative plant products are important sources of rural people income. In order to grow non-ordinary agricultural products, production conditions must be investigated. The results of these studies generate new sources of income for people living in the countryside. There are two ways to discovery the sources of unusual target products. The first way is the adaptation of the exotic species to the ecological conditions of our country, and the second way is the determination of the available resource values in our flora.

# 2. MATERIAL AND METHODS

During the climatic conditions in Kastamonu, medicinal plant production practice were carried out between 2002-2014 and some target species were identified (Tekdemir R., 2003, Uzunoğlu Y. 2004, Güney K.B., 2005). The results of these studies revealed that the plant species of *Salvia officinalis, Melissa officinalis, Origanum onites, Echinacea purpurea, Thymus vulgaris, Mentha piperita, Matricaria chamomilla, Rheum rhabarbatum and Hypericum perforatum* were suitable for mass production. The suitable area for mass production was determined in Gölköy. All of the projects were financed by the authors except for GC-MS analysis. This research was planned in 2012 and implemented in 2013. Technical infrastructure has been established to implement the project. For mass production of target species, land was rented, seedling greenhouse was established, electricity and irrigation infrastructure was established. This business is registered with the farmer registration system. Then, the first stage of the production phase was prepared. The soil was plowed and cleaned from weeds. Leveling was made ready for planting seedlings. Certified seeds of the target species are provided. The seeds thrown into the germination peat mixed with perlite in the viols were ready to plant within 15-25 days of germination within 45 days. Plantation of seedlings ready for planting was given water. Care was taken to complete these procedures until the beginning of July. Considering the climatic conditions, the seedlings were allowed to grow by giving water when needed. No herbiside and insectide were used in the fight against weeds and other pathogens.

Hand-picked herbs were dried in a separate area and burned. The remaining ashes were still sprinkled on the planting area. At the end of the first planting year, the plant was harvested by hand once. In the second year, 2 times the 3rd year was harvested by hand 3 times. Regular water determines the increase in the number of harvests. The harvest is made from the 20 cm section of the hill exile. When irrigation is not done, the number of harvests is limited to one. The first three years were harvested by hand. After the fourth year, the method of harvesting with the machine was used. The harvested plants were dried by natural methods in 90 % shading. Dried products are placed in cloth bags and stored in containers (Photo 1-8).

# 3. RESULTS AND DISCUSSION

Dried plants are marketed either in wholesale or retail market. In addition, some plants were extracted by volatilization by water vapor distillation method and analyzed by GC-MS. These are sage, thyme and mint.

# 4. DISCUSSION

With this project, it has been observed that the medicinal plants that can be taken into domestic or exotic culture have an important economic potential. Furthermore, the fact that the exotic species, which have economic value and which are imported, can be cultured in our country will prevent the outflow of foreign countries.



Figure 1. Rented field in Gölköy



Figure 2. Medicinal plant seedlings



Figure 3. Irrigation system



Figure 4. Manuel harvest



Figure 5. Pre-harvest Sage

Figure 6. Drying in the greenhouse



Figure 7. Pre-harvest coneflower



Figure 8. Pre-harvest majoram

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# REFERENCES

- Tekdemir, R. Ilgaz Dağı Büyük Hacet Yüksek Dağ Florası. Yüksek lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 81 sayfa, Ankara, (2003).
- Uzunoğlu, Y. Ilgaz Dağı Küçük Hacet Yüksek Dağ Florası. Yüksek lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 100 sayfa, (2004).
- Güney, K.B. Ballıdağ Florası (Kastamonu/Daday). Yüksek Lisans Tezi. Ankara Üniversitesi Fen Bilimleri Enstisüsü. 108 sayfa, (2005).



# The Precautions for Conservation of Habitats Upon Flora and Wildlife and Effects of Marble Quarries in Kastamonu on Them

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Abstract: This study has been prepared by compiling the information on the fields where flora and fauna studies on Qualified and Protected Areas for the determination of biological diversity in Kastamonu for more than 30 years. The Republic of Turkey State Planning Organization, TUBITAK, Kastamonu University and the European Union has provided financial support for the detection of these sites. In 2019, Kastamonu University was accepted as a university of specialization by the Council of Higher Education within the scope of Forestry and Natural Tourism. More than 100 projects have been prepared recently aimed at regional development. However, the density of potential marble quarry sites prepared by the General Directorate of Mining Affairs and to be submitted to the tender process poses a great threat to protected areas. In this study, the negative effects of marble quarries on ecosystems and habitats were also explained. The digitized feature field map presented in this article has been obtained by considering flora and vegetation research, wildlife development site reports and biodiversity report.

Keywords: Marble Quarries, Biodiversity, Biologic Conservation, Flora, Fauna, Wildlife, Kastamonu

# **1. INTRODUCTION**

Kastamonu has two important National Parks in the protected areas such as Ilgaz and Küre Mountains. At the same time, these fields, which embrace the karstic belt with their important canyons such as Valla, Horma and Çatak, support their biodiversity with ecological corridors in neighboring areas. Within the scope of the Küre Mountains National Park Project completed in 2004, 89 families, 357 genera and 616 plant taxa were identified. The fauna consists of 48 mammals, 129 bird species, 113 invertebrates, 10 amphibians and 23 reptile taxa. Rapid Ecological Assessment of Küre Mountains National Park and Buffer Zone Project, the flora list was updated and 1014 plant taxa were determined. Of these taxa, 42 species are endemic. In addition, 7 of these species are endangered. In 2018, "The Terrestrial and Inland Water Ecosystems Biological Diversity Inventory and Monitoring Study of Kastamonu Province Project was completed. In this project, 1847 plant taxa were identified and 258 were endemic. In this biodiversity study conducted on a provincial basis, 29 habitats have been identified in 8 different ecosystems. These are given in Table 1 and Table 2.

| Ecosystem<br>Number | EUNIS<br>Code | Ecosystem<br>Type  |  |  |  |  |
|---------------------|---------------|--|--|--|--|--|
| 1.                  | В             | Coastal Ecosystem  |  |  |  |  |
| 2.                  | С             | Inland Surface Waters Ecosystem  |  |  |  |  |
| 3.                  | Е             | Grasslands and Lands Dominated by Forbs, Mosses or Lichens Ecosystem                 |  |  |  |  |
| 4.                  | F             | Heathland, Scrub and Tundra Ecosystem  |  |  |  |  |
| 5.                  | G             | Voodland, Forest and other wooded Land Ecosystem                                     |  |  |  |  |
| 6.                  | Н             | Basic and ultrabasic inland cliffs Ecosystem   |  |  |  |  |
| 7.                  | Ι             | Arable land with unmixed crops grown by low-intensity agricultural methods Ecosystem |  |  |  |  |
| 8.                  | J             | Residential buildings of city and town centres Ecosystem                             |  |  |  |  |

Table 1. EUNIS Ecosystem Type of Kastamonu Region

| Habitat<br>Number | EUNIS<br>Code | Habitat<br>Type  | Area<br>(Hectares) |
|-------------------|---------------|--|--------------------|
| 1.                | B1            | Coastal dunes and sandy shores   | 33,89              |
| 2.                | C1            | Surface standing waters  | 50,17              |
| 3.                | C2.3          | Permanent non-tidal, smooth-flowing watercourses                             | 692,77             |
| 4.                | E2.1          | Permanent mesotrophic pastures and after mathgrazed meadows                  | 4846,53            |
| 5.                | E2.7          | Unmanaged mesic grassland  | 262121,98          |
| 6.                | F5.3          | Pseudomaquis   | 66,09              |
| 7.                | F9            | Riverine and fen scrubs  | 25,33              |
| 8.                | G1.1          | Riparian and gallery woodland, with dominant Alnus, Betula, Populus or Salix | 3514,30            |
| 9.                | G1.6          | Fagus woodland   | 108127,27          |
| 10.               | G1.7          | Thermophilous deciduous woodland   | 22412,98           |
| 11.               | G1.8          | Acidophilous Quercus dominated woodland                                      | 81189,63           |
| 12.               | G1.A          | Meso- and eutrophic Quercus, Carpinus, Fraxinus, Acer, Tilia, Ulmus woodland | 8,11               |
| 13.               | G1.C          | Highly artificial broadleaved deciduous forestry plantations                 | 116,25             |
| 14.               | G3.1          | Abies and Picea woodland   | 84588,71           |
| 15.               | G3.5          | Pinus nigra woodland   | 226422,72          |
| 16.               | G3.6          | Subalpine mediterranean Pinus woodland                                       | 77512,06           |
| 17.               | G3.7          | Lowland to montane mediterranean Pinus woodland (excluding Pinus nigra)      | 5012,26            |
| 18.               | G3.9          | Coniferous woodland dominated by Cupressaceae or Taxaceae                    | 903,27             |
| 19.               | G3.F          | Highly artificial coniferous plantations                                     | 40616,24           |
| 20.               | G4.5          | Mixed Pinus sylvestris - Fagus woodland                                      | 6619,31            |
| 21.               | G4.6          | Mixed Abies - Picea -Fagus woodland  | 20350,49           |
| 22.               | G4.7          | Mixed Pinus sylvestris - acidophilous Quercus                                | 8381,34            |
| 23.               | H3.2          | Basic and ultrabasic inland cliffs   | 1602,14            |
| 24.               | I1.3          | Arable land with unmixed crops grown by low-intensity agricultural methods   | 314182,19          |
| 25.               | J1.1          | Residential buildings of city and town centres                               | 23320,62           |
| 26.               | J2.2          | Rural public buildings   | 202,65             |
| 27.               | J3.2          | Active opencast mineral extraction sites, including quarries                 | 620,89             |
| 28.               | J4            | Transport networks and other constructed hardsurfaced areas                  | 3461,41            |
| 29.               | J5            | Highly artificial man-made waters and associated structures                  | 380,28             |

# **Table 2.** EUNIS Habitat Type of Kastamonu Region

This study has been prepared by compiling the information on the fields where flora and fauna studies on Qualified and Protected Areas for the determination of biological diversity in Kastamonu for more than 30 years. The Republic of Turkey State Planning Organization, TUBITAK, Kastamonu University and the European Union has provided financial support for the detection of these sites. In 2019, Kastamonu University was accepted as a university of specialization by the Council of Higher Education within the scope of Forestry and Natural Tourism. More than 100 projects have been prepared recently aimed at regional development. However, the density of potential marble quarry sites prepared by the General Directorate of Mining Affairs and to be submitted to the tender process poses a great threat to protected areas.



Map 1. Potential marble quarry sites of Kastamonu Province (General Directorate of Mining Affairs)

# 2. MATERIAL AND METHODS

The digitized feature field map presented in this article has been obtained by considering flora and vegetation research, wildlife development site reports and biodiversity report. Each study is based on the results of the research conducted in a particular geography. The sites were investigated independently of each other. In each of these studies, a large number of plant and animal taxa protected by international conventions have been identified (Such as Bern Convention, IUCN, CITES, Ramsar).



Map 2. Red border (Special area - rich areas of biodiversity), Green Border (Ecological corridors between special areas) (1-54).

It is not possible to keep wildlife animal species in independent areas. The acquisition of mining operating licenses between flora and wildlife areas will lead to habitat fragmentation and genetic isolation. This mandatory boundary from condensed mine sites will cause a narrowing of the gene pool and pose a threat to that population. The solution of this problem is to establish the ecological corridors between these individual research areas and to provide the genetic fluidity between them. 13 ecological corridors were proposed in this study.

#### 3. RESULTS AND DISCUSSION

In this study, the negative effects of marble quarries on ecosystems and habitats were also explained. Marble quarries established amongst the core of biodiversity cause habitat fragmentation, while those built on ecological corridors show a barrier effect, causing the shrinkage of the gene pool on plant and animal species to increase the percentage of occurrence of genetic diseases. As a result, diseased populations will lose their assets. It has been demonstrated by the scientific studies that the marble quarries exhibit the following harms. In our country, more open pit operations are carried out. Open pit management covers the ongoing processes of removing the alteration level of soil and vegetation, stepping up and block production. The first phase of operation is the removal of soil and vegetation and / or alteration level (pickling) of the marble formation by excavations. This layer, which is formed in thousands of years and has a rich biodiversity, is irreversibly eliminated. After this excavation process, steps are taken to form an open hearth. With the stepping activities, the hob area is expanded both horizontally and vertically, and as the production of blocks increases, the space volume of the hearth area increases. These pits formed by the marble blocks are the leading factors that cause the most change on natural topography.

- Due to the very low block yield in marble quarries, large amounts of residues are formed. Disposal of wastes resulting from production into indiscriminate environment may cause the masses to shift.

- The process of soil formation on the bedrock and the remaining surface and the spontaneous formation of the vegetative tissue on it again require hundreds of years.

- The accumulation of residues due to increased production requires more storage space. The lack of commitment to the assessment of production in other industrial areas leads to image pollution.

- Another problem is the marble powder produced during production. The grains in the marble quarry are more than 50 microns in the atmosphere and they reach a distance of 800 meters. This value is a distance determined by the resumption of the shield dust with operating activity. In an open area to the wind this distance finds the miles. Although it is said that this spread can be solved by irrigation, it is a fact that the evaporation that occurred after it did not solve this event. In addition, photosynthesis slows down when the top of the leaves of the plants due to dust pollution is closed. After the stoma closure, the respiration stops and as a result, the plant is dried. This is a loss of productivity in agricultural areas.

- Sound noise pollution is generated by cutting and sanding machines and explosives used in the facility. Fauna elements have negative effects especially on reproductive times.

- In marble quarries, if the necessary measures are not taken according to the type of rock, it may occur in the effects such as pollution of the groundwater, noise pollution, soil loss / destruction caused by the fact that the top soil is not stored regularly in a separate place.

- An ecotourism visit to Küre Mountains National Park will not leave a good impression on Kastamonu, as it will not leave a good impression on the mines, noise, trucks and dust.

#### 4. CONCLUSION

In this study, it is recommended not to allow the investments in the ecologically important areas and corridors defined by the red and green boundaries to disrupt the structure of the soil, to harm the biodiversity and to prevent the flow of natural gene resources. In determining ecological corridors, forest areas, valleys, hill ridges and stream beds between the special fields are taken as basis. The proposed ecological corridors for genetic fluidity are given below.

- 1. Ecological corridor: Saka Mountain Germeçtepe Dam Line
- 2. Ecological corridor: Saka Mountain Karaçomak Dam Line
- 3. Ecological corridor: Karaçomak Dam Ilgaz Mountain Line
- 4. Ecological corridor: Ilgaz Mountain Köklüce Mountain Line
- 5. Ecological corridor: Saka Mountain Ballıdağ Line

- 6. Ecological corridor: Ballıdağ Küre Moutains National Park Line
- 7. Ecological corridor: Karyatağı Mountain Küre Moutains National Park Line
- 8. Ecological corridor: Karyatağı Mountain Kart Mountain Line
- 9. Ecological corridor: Ballıdağ Karyatağı Mountain Line
- 10. Ecological corridor: Kart Mountain Küre Ayancık Forest Line
- 11. Ecological corridor: Elekdağ Abana Ayancık Line
- 12. Ecological corridor: Oyrak Gate Ballıdağ Line
- 13. Ecological corridor: Kart Mountain West Küre Mountains

#### REFERENCES

1. Tekdemir, R. Ilgaz Dağı Büyük Hacet Yüksek Dağ Florası. Yüksek lisans Tezi. Gazi

Üniversitesi Fen Bilimleri Enstitüsü. 81 sayfa, Ankara, (2003).

- Uzunoğlu, Y. Ilgaz Dağı Küçük Hacet Yüksek Dağ Florası. Yüksek lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 100 sayfa, (2004).
- 3. Enez, Z., Kastamonu Germeçtepe Barajının Çevresinin Florası. Yüksek Lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 88 sayfa, (2004).
- 4. Köseoğlu, M. Küre Dağları Sorkun Yaylası Florası. Yüksek Lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 78 sayfa, (2005).
- 5. Özen, C. Kart Dağı Florası. Yüksek Lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 83 sayfa, (2008).
- 6. Karaburç, İ. Oyrak Geçidi ve Çevresi Florası. Yüksek Lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 75 sayfa, (2006).
- 7. Pehlivan, G. Ilgaz Dağı Milli Parkı Florası. Yüksek Lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 132 sayfa, (2007).
- Demirbaş, M. Küre Dağları Milli Parkı Armutluçayır Bölgesi Florası. Yüksek Lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstitüsü. 81 sayfa, (2004).
- 9. Özbek, M.U. Kurtgirmez Dağı ve Çatak Kanyonu (Küre Dağları-Kastamonu) Florası. Yüksek Lisans Tezi. Gazi Üniversitesi Fen Bilimleri Enstisüsü. 161 sayfa, (2004).
- Güney, K.B. Ballıdağ Florası (Kastamonu/Daday). Yüksek Lisans Tezi. Ankara Üniversitesi Fen Bilimleri Enstisüsü. 108 sayfa, (2005).
- 11. Kanoğlu, E. Abana (Kastamonu) ve Yöresinin Florası. Yüksek Lisans Tezi Karadeniz Teknik Üniversitesi Fen Bilimleri Enstisüsü. 104 sayfa, (2002).
- 12. Menteş, İ. Ilgaz Dağı Milli Parkının Korunan Alan Olarak İncelenmesi ve Yönetimi. Doktora Tezi. Karadeniz Teknik Üniversitesi Fen Bilimleri Enstisüsü. 227 sayfa, (2001).
- 13. Demirörs M. Saka Dağı Florası. Doktora Tezi. Ankara Üniversitesi Fen Bilimleri Enstisüsü. 165 sayfa (1982).
- Ketenoğlu, O., Güney, K. Batı Küre Dağları (Kastamonu-İnebolu-Cide) Florasına Katkılar. Ot Sistematik Botanik Dergisi. 4 (2): 39-60 Ankara, (1997).
- 15. Sakallıoğlu H. Karaçomak Barajı (Kastamonu) ve Civarı Florası. Ormancılık Araştırma Dergisi. 29(57): 23-54, Ankara, (1983).
- Demirörs, M. Saka Dağ Florasına Katkılar (Kastamonu/Türkiye). Gazi Üniversitesi Kastamonu Eğitim Fakültesi Dergisi. Mart 2006, Cilt:14, No:1, 241-244.
- 17. Çiçek, E. Bir Doğa Harikası Kızılcasu (Kastamonu). Florası ve Tali Türlerimiz. 1.Ulusal Ormancılık Kongresi, Türkiye Ormancılar Derneği, T.C Orman Bakanlığı, Ankara, 2001, 21-55.
- 18. Ketenoğlu K., New floristic records from Northwestern Anatolia (Kastamonu province) Candollea .1983, Vol. 38, No. 2, 465-476.
- Kılınç, M. New Floristic Records from A5 (Kastamonu-Çorum). Communications. De La Faculte Des Sciences De L'Universite D'Ankara. Serie C2, Tome 21, Anne 1977. Ankara.
- 20. Tütüncü M., Ilgaz Dağı Milli Parkı ve Yakın Çevresinin Eğrelti Otları (*Pteridophyta*) Florası. Yüksek Lisans Tezi. Ankara Üniversitesi Fen Bilimleri Enstisüsü, 53 sayfa (2006).
- 21. Vurdu H., Uslu N., Güney K., Ünal S., Ayan S., Sıvacıoğlu A., Gürel N., Küçük Ö., Ulusan D., Öztürk S., Türkyılmaz E., Küre Dağı Milli Parkı'nın Floristik Zenginliği ve Habitat Alanlarının Belirlenmesi. DPT Projesi 2002K120250, Sonuç Raporu, Ankara. 2004.

- 22. Vural, M. Flora Report. Protection of Biodiversity and collaborative protected area management at the Kure Mountains National park as a model for Turkey, 37 p., (PIMS: 1988 Enhancing Forest Protected Areas Management System-UNDP Project) Ankara. 2003.
- 23. Güney K., Benli M., Yiğit N., Geven F., Bingöl Ü. Karyatağı Dağı (Kastamonu/Azdavay)'ın florası ve endemik bitkilerinin antimikrobiyal etkilerinin araştırılması. Tübitak-TBAG-HD/16 (105T031) (2006).
- 24. Kurt, L., Köklüce Dağı (Kastamonu)'nın Bitki Sosyolojisi Yönünden Araştırılması Yüksek Lisans Tezi, Ankara Üniversitesi Fen Bilimleri Enstisüsü, 1-60, (1992).
- 25. Güney K., Geven F., Bingöl Ü. Kastamonu İli Ruderal Vejetasyonunun Sintaksonomik Analizi. Tübitak-TOVAG-1050022 (2006).
- 26. Yurdakulol E., Demirörs M., Yıldız A., 2002. A phytosociological study of vegetation of the Devrekani-İnebolu-Abana area (Kastamonu, Turkey) Israel Journal of Plant Sciences, Vol. 50, Sayfa 309.
- 27. Akman Y, Quezel P, Yurdakulol E, Ketenoğlu O, Demirors M (1987) La végétation des hauts sommets de l' Ilgaz Dağ. Ecologia Mediterranea 13, 1-2, 119-129.
- 28. Davies C. E., Moss D., Hill M. O. 2004. EUNIS Habitat Classification Revised 2004.
- 29. Davis, P.H., Flora of Turkey and The East Eagean Islands, 1965-1985. Edinburg University Press, Edinburgh, (1-9): 1-600.
- 30. Davis, P.H., Mill, R. R., Tan., Kit. 1988. Flora of Turkey and The East Eagean Islands, Suplement I. Edinburg University Press, Edinburg.
- 31. Güner, A., Özhatay, N., Ekim T., Başer, K.H.C., 2000. Flora of Turkey and The East Eagean Islands", Suplement II. Edinburg University Press, Edinburg.
- 32. Çırpıcı, A., Türkiyenin Flora ve Vejetasyonu Üzerindeki Çalışmalar, 1987. Doğa T.U. Botanik Dergisi, 11: 2. Ankara.
- 33. Davis, P.H., Hedge, I.C., 1975. The Flora of Turkey. Past, Present, and Future, Candollea, 30 (2): Edinburg, 331-351.
- 34. Güner, A., Aslan, S., Ekim, T., Vural, M., Babaç, M.T., (edlr.). 2012. Türkiye Bitkileri Listesi (Damarlı Bitkiler). Nezahat Gökyiğit Botanik Bahçesi ve Flora Araştırmaları Derneği Yayını. İstanbul.
- 35. Ekim, T., Koyuncu, M, Vural, M., Duman, H., Aytaç, Z., Adıgüzel, N. 2000. Türkiye Bitkileri Kırmızı Kitabı, Barışcan Ofset, Ankara, 5-15.
- 36. Özhatay N., Kültür Ş., Aksoy N. 1994. Check-List of Additional Taxa to the Supplement Flora of Turkey. T. Jr. of Botany. 18, 497-514.
- 37. Özhatay N. ve ark. 1999. Check-List of Additional Taxa to the Supplement Flora of Turkey II. T. Jr. of Botany. 23, 151-169.
- 38. Özhatay N. ve ark. 2006. Check-List of Additional Taxa to the Supplement Flora of Turkey III. T. Jr. of Botany. 30, 281-316.
- 39. Özhatay N. ve ark. 2009. Check-List of Additional Taxa to the Supplement Flora of Turkey IV T. Jr. of Botany. 33, 191-226
- 40. Özhatay N. ve ark. 2011. Check-List of Additional Taxa to the Supplement Flora of Turkey V. T. Jr. of Botany. 35, 589-624.
- 41. Özhatay N. ve ark. 2013. Check-List of Additional Taxa to the Supplement Flora of Turkey VI. Istanbul Ecz. Fak. Derg. 43(1) s.33-82.
- 42. Ilgaz Dağı Yaban Hayatı Geliştirme Sahası Raporu.
- 43. Kartdağı Yaban Hayatı Geliştirme Sahası Raporu.
- 44. Gavurdağı Yaban Hayatı Geliştirme Sahası Raporu.
- 45. Elekdağ Yaban Hayatı Geliştirme Sahası Raporu.
- 46. Özbek, U., Koç, M., Hamzaoğlu, E. (2018). *Rhaponticum pulchrum* (Asteraceae), a new record for the Turkish Flora. Gazi University Journal of Science (Kabul edildi).
- 47. Duran, A., Dinç M., Martin, E. (2008). A New Species of *Draba* (Brassicaceae) from North Anatolia, Turkey. Novon A Journal for Botanical Nomenclature 18 (4):464-468
- 48. Çetin, Ö., Şeker, M.Ö., Duran, A. (2015). A new subspecies of Seseli gummiferum (Apiaceae) from Ilgaz Mountain National Park,

northern Turkey. PhytoKeys 56: 99-110.

- 49. Duran, A., Çetin, Ö. (2016). New species and a synonym of the genus Hesperis (Brassicaceae) from Turkey. Turk J Bot 40: 87-96
- Hamzaoğlu, E., Koç M. (2016). A new gigantic species from Turkey, Angelica turcica (Umbelliferae). Phytotaxa 245 (1): 066– 070.
- 51. http://www.bizimbitkiler.org.tr/v2/index.php (01.03.2019)
- 52. http://www.tubives.com/ (01.03.2019)
- 53. http://www.cites.org/ (01.03.2019)
- 54. http://conventions.coe.int/Treaty/en/Treaties/Word/104.doc (01.03.2019)



# A Methodological Approach to the Determination of Soil Damage According to Logging Techniques in Coniferous Stands

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Abstract: Production of timber in forestry covers the whole of the activities carried out in the process of meeting the demands of them and transporting to storage by cutting planted trees reaching the cutting age in order to earn income for the forest enterprises. The production of timbers is generally divided into two separate phases. The first of these is logging operation process (primary transport), which involves transporting from the side of the tree to the nearest forest road and the second is far transportation (secondary transport), transporting of products by trucks and similar vehicles to forest sales depots. Logging operations that are the first and most difficult stage of producing the timbers are usually done by human and animal power and with partly mechanized in our country. The lack and not optimal usage of the logging methods with mechanization increases the cost of harvesting and can also cause damages on forest ecosystems (trees in the stands, youth, forest soil, etc.). For this reason, it is necessary not only to reduce the costs but also to use modern methods to minimize damages to the ecosystem during and after logging operations. Logging operations usually change the soil compaction, infiltration properties of the soil. In addition, the logging operations increase the degradation of the humus layer of the soil, the tearing of the soil surface, and finally change the erosion tendency values.

In this methodological approach, in coniferous stands, according to stand development stage (b-bc, c-cd), slope classes (0-20%, 20-33%,> 33% (according to IUFRO) the log passes on the skidding trail (5. log passes,10. log passes), it will be determined which of the mechanization methods will be suitable in terms of soil damage (in soil occurs due to the change in the soil compaction, bulk density, soil moisture, organic matter, pH, soil respiration, texture, electrical conductivity and TOC (total organic carbon-total nitrogen)) in which the methods of mechanization are skidding with tractor, skidding cable, skidding with portable winch (with skidding cone)

Keywords: A Methodological Approach, Logging Techniques, Soil Properties, Soil Damage, In Coniferous

# **1. INTRODUCTION**

In order to benefit from the forests rationally and continuously, while determination of logging techniques, the selection of the application which is sensitive to the ecosystem, economic, will be used in the shortest period of time to be used and which gives the least damage to the product should be paid attention. In our country, as a result of 95% of logging operation the is carried out traditional methods (animal power-man power), quality and amount losses on logs and various damages on ecosystem arisen. At the same time, the conditions for working workers are heavy and accidents can lead to death. For this reason, logging operations which constitutes the first and the most difficult stage of the harvesting are one of the subjects that should be developed with more modern and sensitive methods.

In recent years, it has gained great importance to determine and minimize the environmental impacts of all kinds of interferences to ecosystem with the increase of environmental sensitivity of the society (Ünver ve Acar, 2006). Precision forestry approach aims to provide optimum efficiency from forest resources and minimize environmental damage by using modern techniques and technological tools to make economic, environmental and sustainable decisions in forestry studies (Gülci, 2014).

Timber production methods cause damage on soil, residual stand and log (Bayoğlu 1972; Gürtan 1975; Akay ve ark., 2007 a,b; Makineci ve ark., 2007). Menemencioğlu (2006), indicateted in his study that on sloping land logging operation done with the manpower in the soil and residual trees led to more destruction. Turk and Gumus (2015) surveyed the damage of soil and seedlings using the agricultural tractors on skidding trail.

Soil compaction occurs when timber skid on ground. This reduces soil porosity on the forest floor and affects water infiltration, soil moisture, soil aeration and root volume. (Greacen and Sands, 1980). Landsberg (2003), determined that the depth of the trace after skidding on the forest floor was 15-25 cm and the average soil compaction was 500kP and over. According to (Virdine et al, 1999), it was determined that 11% of the area in which gap cutting was made was deteriorated, the volume weight increased by 21% and the 70% of cutting area was covered with the residues. Eroğlu et al. (2010), stated that logging operations with the tractor and manpower has a significant impact on forest soil permeability, volume weight and soil water balance and these impacts have negative effects on the soil organisms, root growth of plants, plant nutrients and plants water intake.

# 2. MATERIAL AND METHODS

This study will be carried out in Kastamonu Regional Directorate, Samatlar Forest Management Directorate.



Figure 1. Geographic location of the study area

1. . . . . . .

Different harvesting stands to be formed according to the method of logging tecniques, development stage, slope class and log passes. And in terms of stand development stage; the stage of "b" indicates pole stage; and the stage of "c" indicates small-size tea and the stage of "d" indicates the mid-size tree.

| Table 1. | The distribution of selected sample fields |
|----------|--|
|          |  |

| Logging techniques                                | Stands<br>stage | development | Slope classes(%) | Logging<br>passes |
|---|-----------------|-------------|------------------|-------------------|
| Skidding with tractor                             | b-bc /c-cd      |             | 0-20 / 20-33     | 5./10.            |
| Skidding cable                                    | b-bc /c-cd      |             | 20-33 / >33      | 5./10.            |
| Skidding with portable winch (with skidding cone) | b-bc/c-cd       |             | 0-20/20-33/>33   | 5./10.            |
| Control   | b-bc /c-cd      |             | 0-20/20-33/>33   |                   |

In the scope of the study, first of all, soil samples will be taken in order to reveal the state of the soil structure in the preproduction study areas before they are affected by the logging activities.

Soil samples will be taken from 2 different points and 2 different depth levels (0-5 cm, 5-10 cm) of each sample area and required measurements will be done with penetrometer and soil respiration devises on this points.

With the start of harvesting, 3 skidding trails will be determined in the area where 3 logging techniques will be used near the control points and haven't been any previous logging operation.

3 logging techniques will be carried out on the skidding trails by taking into account the log passes (5. passes and 10. passes).

Firstly, for 5. log passes, soil samples will be taken from 2 different points and 2 different depth (0-5 cm, 5-10 cm) and then penetrometer and soil respiration measurements will be done same points. The same procedures will be also applied after the 10. log passes on the skidding trails.

Chambers used to measure soil respiration will be left in the area after the first measurement. These measurements will be repeated in order to determine how change soil respiration and compaction in the soil 5 days and 10 days after.

Thus soil samples will be taken (from 24 control points, 32 log trail and 32-wheel trail for tractor, 32 cable with tractor, 48 portable winch) 168 in total.

#### **3. CONCLUSION**

The optimum primary transport plan will be developed to minimize the soil damages determined according to stage classes, slope classes and logging passes. With the creation of the models of these areas, the most appropriate decisions can be made to determine the logging techniques suitable for the area technical structure for future studies.

As a result of this study, it will be possible to make the most appropriate decisions about determining of the logging technique according to the technical structure of the land and important contribution will be made in conventionalization of the use of the most appropriate method in the harvesting for the forestry of our country. It will be a sample application for precision forestry studies reducing the damages to ecosystem works and determining appropriate methods for the land. In this study, the effect of the damage on soil in various areas will be demonstrated by applying different logging techniques and will contribute to the application of environmentally sensitive forestry operations.

#### REFERENCES

- Acar, H. H. ve Eroğlu, H., 2003. Dağlık Arazide Üretilen İnce Çaplı Odunların Fiberglass Oluk Yöntemi İle Bölmeden Çıkarılması İmkanları Üzerine Bir Araştırma. KTÜ Proje No: 22.113.001-2, 35s., Ocak 2003, Trabzon.
- Akay, A. E., Sessions, J. ve Aruga, K., 2007. Designing A Forwarder Operation Considering Tolerable Soil Disturbance and Minimum Total Cost. J. Terramechanics, 44, 187-195.
- Akay, A. E., Yüksel, A., Reis, M. ve Tutus, A., 2007. The Impacts of Ground-Based Logging Equipment On Forest Soil. Pol. J. Environ. Stud. 16, 371-376
- Bayoğlu S (1972) Türkiye'de Orman Nakliyatı ve Geliştirilmesi İmkanları Üzerine Bir Etüd, İ.Ü. Orman Fakültesi, İstanbul, 1747/185, 73.
- Eroğlu, H., Sarıyıldız, T., Küçük, M. ve Sancal, E. 2010. Doğu Ladini Meşcerelerinde Bölmeden Çıkarma Çalışmalarının Orman Toprağının Fiziksel Özellikleri Üzerine Etkileri. Süleyman Demirel Üniversitesi Orman Fakültesi Dergisi Seri: A. Sayı: 1, ISSN: 1302-7085, pp. 30-42.
- Greacen, E. L., & Sands, R. 1980. Compaction of forest soils. A review. Soil Research, 18(2), 163-189.
- Gülci, N., 2014. Üretim Planlamasında Hassas Ormancılık Üzerine Araştırmalar. Doktora Tezi, Kahramanmaraş Sütçü İmam Üniversitesi, Orman Fakültesi, Kahramanmaraş. 264 s.
- Gürtan H., Dağlık ve Sarp Arazili Ormanlarda Kesim ve Bölmeden Çıkarma İşlemlerinde Uğranılan Kayıpların Saptanması ve Bu İşlerin Rasyonalizasyonu Üzerine Araştırmalar, Tübitak Yayın No: 250, Proje No: Toag-81, Ankara.
- Landsberg, J.D., Miller, R.E., Anderson, H.W. ve Tepp, J.S., 2003. Bulk Density and Soil Resistance to Penetration As Affected by Commercial Thinning in Northeastern Washington, Res. Pap. Portland, or: U.S. Department of Agriculture, Forest Service, Pacific Northwest Research Station, Washington.
- Makineci, E., Demir, M. ve Yılmaz, E., 2007. Long-Term Harvesting Effects On Skid Trail Road In A Fir (Abies Bornmulleriana Mattf.) Plantation Forest. Build. Environ. 42, 1538-1543.
- Menemencioğlu K., 2006." Ilgaz Devrez Orman İşletme Sefliği'nde Cografi Bilgi Sistemi (Cbs) Yardımıyla Orman Hasat Zararlarını Azaltıcı Transport Planlaması" Doktora Tezi, BARTIN.

- Ünver, S. ve Acar, H. H. 2006. The Effects of Wood Raw Material Production Activities on Wood Quality Classes. Journal of Artvin Forestry Faculty. 6 (1/2). 128-134.
- Türk, Y., & Gümüş, S., 2015. Tarım traktörleriyle bölmeden çıkarmada meydana gelen toprak ve fidan zararlarının araştırılması. Artvin Çoruh Üniversitesi Orman Fakültesi Dergisi, 16(1), 55-64.
- Virdine, C.G., Dehoop, C. ve Lanford, B.L., 1999. Assessment of Site and Stand Disturbance from Cut-to-Length Harvesting, 10th Biennial Southern Silvicultural Research Conference, February 16-18 1999, Shreveport, La.



# Job Satisfaction of Employees of the Zonguldak Regional Directorate of Forestry and Its Relationship with Some Individual Features

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Abstract: This study was carried out to determine the job satisfaction of the employees in the Zonguldak Regional Directorate of Forestry and its affiliated units, to check the difference of job satisfaction according to some individual features (age, gender, marital status, working status of the spouse, place of birth, education, task unit, duty position, total service duration, duty duration, number of duty place, proxy duty) and to determine the relationships between individual features and job satisfaction. In order to obtain the research data, a questionnaire form consisting of two parts (Individual Features Section; Job Satisfaction Section) was prepared. This form has been applied to 348 people working in various positions in the Zonguldak Regional Directorate of Forestry and affiliated units in 2018 by sending to their e-mail addresses and face to face interviews. Descriptive statistics, the Kruskal-Wallis H-Test and Correlation Analysis were used to evaluate the data obtained. The total job satisfaction level of the employees was measured as Low (43-100 points), Medium (101-158 points) and High (159-215 points) according to the answers given to 43 questions with 5-point Likert scale. The total job satisfaction level of the subjects ranged from 71 to 210 points, and the overall average was 141.68 points and the employees had medium job satisfaction. Total job satisfaction showed a statistically significant difference according to the task unit, marital status, working status of the spouse and proxy duty. However, it was determined that total job satisfaction did not show a significant difference according to the position of duty, education, age, gender, place of birth, total service duration, duty duration and number of duty place. In additionally, it was found that total job satisfaction was negatively related to the marital status and proxy duty variables. At the end of the study, some evaluations and suggestions were made in order to increase job satisfaction of the employees of the Zonguldak Regional Directorate of Forestry and its affiliated units and to improve their performance.

**Keywords:** Job satisfaction, variables affecting job satisfaction, individual features, business success, Zonguldak Regional Directorate of Forestry

# **1. INTRODUCTION**

Knowing the level of satisfaction of the employees from their workplaces and works and the variables related to it is extremely important for the management to achieve its goals and to succeed. Job satisfaction is one of the most important subjects of working life. The success of the institutions depends on the employees who have high job satisfaction. For this reason, job satisfaction studies have gained value in all institutions today.

Job satisfaction is defined in different ways by behavior scientists and researchers (İncir, 1990; Eren, 1996; Aksu et al., 2002; Akkamış, 2010). Job satisfaction is generally defined as the level of satisfaction or dissatisfaction that people feel about their jobs (Davis, 1981; Çıtak et al., 2008). In this study, job satisfaction is defined as "the level of satisfaction from work and workplace, which occurs as a result of the integration of the employee into the job, loving his work, connecting to his job, self-sacrificing work, respecting the person, adequacy of work place and working conditions, wages and social rights, and on the one hand, improving the work efficiency on the other hand, providing benefits to the person."

Various studies are carried out on the job satisfaction of the employees in almost every organization. Studies are generally based on measuring the job satisfaction level of the person, determining the factors affecting job satisfaction and examining the relationships with personal characteristics. In forestry organization, it is important to measure the level of job satisfaction of the employees, to determine the factors affecting job satisfaction and to take measures to increase job satisfaction.

Some studies have been made on job satisfaction of employees in the forestry organizations in various regions of Turkey (Yılmaz and Koçak, 2008; Yılmaz et al., 2009; Akyüz et al., 2011; Çok et al., 2017; Bozkurt et al., 2018). However, such a study has not been done before in the Zonguldak Regional Directorate of Forestry and its affiliated units.

Therefore, this study was carried out to determine the job satisfaction of the employees in the Zonguldak Regional Directorate of Forestry and its affiliated units (Zonguldak, Devrek, Dirgine, Ereğli, Yenice, Bartın, Ulus, Karabük, Safranbolu Forest Enterprises, Gökçebey Nursery), to check the difference of job satisfaction according to some individual features (age, gender, marital status, working status of the spouse, place of birth, education, task unit, position of duty, total service duration-experience, duty duration, number of duty place, proxy duty) and to determine the relationships between individual features and job satisfaction. In this respect, the subject is original and useful. The results will provide positive contributions to the forest resources management process.

# 2. MATERIAL AND METHODS

This study was carried out in the Zonguldak Regional Directorate of Forestry (ZRDF) and its affiliated units (Zonguldak, Devrek, Dirgine, Ereğli, Yenice, Bartın, Ulus, Karabük, Safranbolu Forest Enterprises, Gökçebey Nursery) (Figure 1). ZRDF has a total area of 936,063.6 ha, of which 63% (592,017.9 ha) is forested and the remaining 37% (344,045.7 ha) is open area (ZRDF, 2017).



Figure 1. Study area of the Zonguldak Regional Directorate of Forestry (ZRDF, 2017)

In order to obtain the research data, a questionnaire form consisting of two parts (Individual Features Section-12 questions; Job Satisfaction Section-43 questions with 5-Likert Scale) was prepared. In the first section, there are 12 questions related to some individual features of employees (age, gender, marital status, working status of the spouse, place of birth, education, task unit, duty position, total service duration, duty duration, number of duty place, proxy duty), and in the second section, 43 questions prepared according to the 5-point Likert Scale (1-I am not satisfied at all, 2-I am less pleased, 3-I am medium satisfied, 4- I am more satisfied, 5-I am very much satisfied) that measures job satisfaction in various aspects.

In 2018, the number of people working in the Zonguldak Regional Directorate of Forestry and its affiliated units is 635. The survey was conducted in full field (on all staff) by e-mail sending and face-to-face interviews. However, the number of surveys realized and recycled was 348 (ie a sampling of 55%).

Total job satisfaction level was measured with a score of 43-215 by 43 questions with 5-Likert scale and defined as a variable for statistical analyses. Qualitative values of individual features have been digitized by order statistics method

and defined as variables. Individual features were evaluated with descriptive statistics (percentage, arithmetic mean and standard deviation). The Kruskal-Wallis (K-W) H-Test was used to check the difference between total job satisfaction and some individual features. The relationships between individual features and job satisfaction were investigated by Correlation Analysis. MS Excel and SPSS 22.0 programs were used to evaluate the data.

# 3. RESULTS AND DISCUSSION

# **Individual Features and Descriptive Statistics**

Findings and descriptive statistics on the individual features of 348 subjects surveyed are given in Table 1 and 2. According this results, 20% of the interviewees work in Bartın Forest Enterprise, 15% in Zonguldak (included center of regional directorate), 15% in Safranbolu, 11% in Karabük, 9% in Devrek (included Gökçebey Forest Nursery), 9% in Yenice, 8% in Ereğli, 8% in Ulus and%5 in Dirğine. Of these, 46% are office personnel, 27% are land staff, 15% are chief engineer and engineer, 9% is other employee and 3% is manager and assistant manager.

74% of them are university graduates. Their age varies between 22 and 64, and the average age is 41. In addition, 80% of the interviewees were male and the majorities (83%) were married. The spouse of 50% of the interviewees does not work. Majority (78%) work in the geographic region where the birthplace is located. The total service duration (experience) of the interviewees ranged from 1 to 44 years and the average was 16.5 years. The duration of duty is between 1-42 years and the average is 9.4 years. The number of duty place of the participants varied between 1 and 12, and the average was 2.3 times. This result shows that the employees of the Zonguldak Regional Directorate of Forestry continue to work for a long time. Likewise, the majority (56%) have no proxy duty. However, 44% have proxy duty time to time or every time.

| Features  | Groups   | Number | Percent | Mean      |  |  |
|---|--|--------|---------|-----------|--|--|
|   | 1.Zonguldak  | 51     | 15      |           |  |  |
|   | 2.Bartın   | 68     | 20      |           |  |  |
|   | 3.Devrek   | 31     | 9       |           |  |  |
|   | 4.Dirgine  | 17     | 5       |           |  |  |
| Task unit   | 5.Ereğli   | 28     | 8       | 4,6       |  |  |
|   | 6.Karabük  | 40     | 11      |           |  |  |
|   | 7.Safranbolu   | 54     | 15      |           |  |  |
|   | 8.Ulus   | 27     | 8       |           |  |  |
|   | 9.Yenice   | 32     | 9       |           |  |  |
|   | 1. Director, vice director                           | 12     | 3       |           |  |  |
|   | 2. Chief engineer, forest management chief, engineer | 52     | 15      |           |  |  |
| Duty position   | 3. Office staff                                      | 160    | 46      | 2,8       |  |  |
|   | 4. Land staff  | 93     | 27      |           |  |  |
|   | 5.Other  | 31     | 9       |           |  |  |
|   | 1.Primary education                                  | 29     | 8       |           |  |  |
|   | 2.High school  | 64     | 18      | 2.0       |  |  |
| Education   | 3.College  | 145    | 42      | (College) |  |  |
|   | 4.University   | 97     | 28      |           |  |  |
|   | 5.Postgraduate                                       | 13     | 4       |           |  |  |
|   | 20-30  | 54     | 16      |           |  |  |
|   | 31-40  | 122    | 35      | 10.9      |  |  |
| Task unit       5.Ereğli       28         6.Karabük       40         7.Safranbolu       54         8.Ulus       27         9.Yenice       32         1. Director, vice director       12         2. Chief engineer, forest management chief, engineer       52         3. Office staff       160         4. Land staff       93         5.Other       31         1.Primary education       29         2.High school       64         3.College       145         4.University       97         5.Postgraduate       13         20-30       54         31-40       122         41-50       108         ≥51       64         1.Male       280         2.Female       68         1.married       290         2.Single       58 | 31   | 40,8   |         |           |  |  |
|   | ≥51  | 64     | 18      |           |  |  |
| Gandar  | 1.Male   | 280    | 80      | 1.2       |  |  |
| Gender  | 2.Female   | 68     | 20      | 1,2       |  |  |
| Marrital status   | 1. married   | 290    | 83      | 1.2       |  |  |
| Marital status  | 2.Single   | 58     | 17      | 1,2       |  |  |
| Working status of the   | 1.Wife Working                                       | 116    | 33      |           |  |  |
| working status of the   | 2.Wife Not Working                                   | 174    | 50      | 1,3       |  |  |
| spouse  | 3.No Wife  | 58     | 17      |           |  |  |

Table 1. Some individual features of the interviewee

|                         | 1.Marmara Region                                 | 18  | 5  |      |  |  |  |
|-------------------------|--|-----|----|------|--|--|--|
|                         | 2. Aegean Region         3. Mediterranean Region |     | 2  |      |  |  |  |
|                         |  |     | 6  |      |  |  |  |
| Region of birth place   | 4. Southeastern Anatolia Region                  | 1   | 0  | 5,6  |  |  |  |
|                         | 5. Eastern Anatolia Region                       | 3   | 1  |      |  |  |  |
|                         | 6. Blacksea Region                               | 272 | 78 |      |  |  |  |
|                         | 7. Central Anatolia Region                       | 27  | 8  |      |  |  |  |
| Total service duration- | 0-10   | 116 | 33 |      |  |  |  |
|                         | 11-20  | 106 | 31 | 16,5 |  |  |  |
| experience (year)       | 21-30  | 91  | 26 | 1    |  |  |  |
|                         | ≥31  | 35  | 10 |      |  |  |  |
| _                       | 0-5  | 144 | 41 |      |  |  |  |
| Duty duration           | 6-10   | 101 | 29 |      |  |  |  |
| Duty duration           | 11-15  | 36  | 10 | 9,4  |  |  |  |
| (year)                  | 16-25  | 44  | 13 |      |  |  |  |
|                         | ≥26  | 23  | 7  |      |  |  |  |
| Duovy duty              | 1.No   | 195 | 56 |      |  |  |  |
| rioxy uniy              | 2.Time to time                                   | 135 | 39 | 1,5  |  |  |  |
|                         | 3.Every time                                     | 18  | 5  |      |  |  |  |

Table 2. Descriptive statistics on individual features

| Variables                           | Minimum | Maximum | Mean   | <b>Standard Deviation</b> |
|-------------------------------------|---------|---------|--------|---------------------------|
| Task unit                           | 1       | 9       | 4,6    | 2,7                       |
| Duty position                       | 1       | 5       | 2,8    | 0,93                      |
| Education                           | 1       | 5       | 3,0    | 0,97                      |
| Age                                 | 22      | 64      | 40,8   | 9,54                      |
| Gender                              | 1       | 2       | 1,2    | 0,39                      |
| Marital status                      | 1       | 2       | 1,2    | 0,37                      |
| Working status of the spouse        | 0       | 2       | 1,3    | 0,75                      |
| Region of birth place               | 1       | 7       | 5,6    | 1,43                      |
| Total service duration (experience) | 1       | 44      | 16,5   | 10,03                     |
| Duty duration                       | 1       | 42      | 9,4    | 8,33                      |
| Number of duty place                | 1       | 12      | 2,3    | 1,55                      |
| Proxy duty                          | 1       | 3       | 1,5    | 0,60                      |
| Total Job Satisfaction              | 71      | 210     | 141,68 | 26,27                     |

# **Measurement of Job Satisfaction**

Total job satisfaction was measured by 43 questions having Likert scale with 5 points as a score between 43 and 215, and it was determined as below three groups:

- 1) Low job satisfaction is between 43-100 points,
- 2) Medium job satisfaction is between 101-158 points,
- 3) High job satisfaction is between 159-215 points.

The employees' total job levels range from 71 to 210 points and the general mean is  $141.68 \pm 26.27$  points ( $\pm$ S). Accordingly, it was found that employees had medium job satisfaction.

The first three questions having highest score on the job score scale consisting of 43 questions are;

- 1) My work is meaningful and useful to humanity (question 4;  $\bar{x} = 3.99$ ),
- 2) Good communication and dialogue with my subordinates at my workplace (question 14;  $\bar{x} = 3.83$ ),
- 3) Since my job provides a guaranteed future (question 27;  $\overline{x} = 3.74$ ).

Therefore, these three questions should be given importance to increase total job satisfaction. The last three questions having lowest score are as follows;

- 1) The promotion and relocation criteria at my workplace are fair, reasonable and logical (question 17;  $\bar{x} = 2.69$ ),
- 2) From the reward-incentive and appreciation system in return for my work (question 30;  $\bar{x} = 2.71$ ),
- 3) From the change of place/task periodically made in the organization (question 34;  $\bar{x} = 2.79$ ).

# Controlling Difference of Total Job Satisfaction according to Individual Features

The difference of total job satisfaction according to task unit, duty position, education, age, gender, marital status, working status of the spouse, place of birth, total service duration-experience, duty duration, number of duty place, and proxy duty was controlled by the Kruskal-Wallis (K-W) H-Test, and Duncan Test was used to reveal different groups (Kalıpsız, 1988; Özdamar, 2002). According this results, it was determined that total job satisfaction showed a statistically significant difference according to the task unit, marital status, working status of the spouse and proxy duty. However, it was determined that total job satisfaction did not show a significant difference according to the position of duty, education, age, gender, place of birth, total service duration, duty duration and number of duty place (Table 3).

| Variables                                  | Kesuits of<br>Kruskal-Wallis<br>H-Test |    |             | Controlling difference by Duncan Test                              |                     |                            |                 |  |  |
|--|--|----|-------------|--|---------------------|----------------------------|-----------------|--|--|
| v ariables                                 | Chi-<br>Square<br>Value                | DF | No          | No Groups Elements Ja<br>Satisf                                    |                     | X                          | IN              |  |  |
| 1. Task Unit                               | 25,72**                                | 8  | 1 2         | Yenice<br>Ulus, Bartın, Zonguldak, Dirgine,<br>Safranbolu, Karabük | Low<br>Medium       | 131,75<br>141,74           | 32<br>257       |  |  |
| 2. Marital status                          | 5,72*                                  | 1  | 3<br>1<br>2 | Ereğli, Devrek<br>Single<br>Married                                | High<br>Low<br>High | 150,51<br>133,41<br>143,76 | 59<br>58<br>290 |  |  |
| 3. Working status of the spouse            | 8,27*                                  | 2  | 1 2         | No wife<br>Wife working or not working                             | Low<br>High         | 133,41<br>143,76           | 58<br>290       |  |  |
| 4. Proxy duty                              | 5,75*                                  | 2  | 1<br>2      | Every time<br>No or soma time                                      | Low<br>High         | 127,5<br>142,08            | 18<br>330       |  |  |
| 5. Duty position                           | 1,07                                   | 4  | T           | otal job satisfaction is not different acco                        | ording to duty.     |                            |                 |  |  |
|  | 3,39                                   | 4  |             | bial job satisfaction is not different acco                        | ording to educati   | on.                        |                 |  |  |
| 7. Age<br>8. Gender                        | 0.32                                   | 1  |             | tal job satisfaction is not different accord                       | ording to gender    |                            |                 |  |  |
| 9 Birth place                              | 3 33                                   | 6  | T           | tal job satisfaction is not different accord                       | ording to birth pl  | ace                        |                 |  |  |
| 10. Total service duration<br>(experience) | 4,66                                   | 3  | To          | Total job satisfaction is not different according to birth place.  |                     |                            |                 |  |  |
| 11. Duty duration                          | 3,09                                   | 4  | Τc          | otal job satisfaction is not different acco                        | ording to duty du   | ration                     |                 |  |  |
| 12. Number of duty place                   | 10,27                                  | 9  | To<br>pl    | otal job satisfaction is not different acco<br>ace.                | ording to number    | r of dut                   | у               |  |  |

Table 3. Controlling difference of total job satisfaction according to individual features

\* is significant at 0.05 confidence level; \*\* is significant at 0.01 confidence level; DF: Degree of freedom

According to Table 3, the total job satisfaction in the Yenice Forest Enterprise was *low*, and total job satisfaction was found as *medium* in Ulus, Bartın, Zonguldak, Dirgine, Safranbolu and Karabük Forest Enterprises and *high* in Ereğli and Devrek Forest Enterprises. It is thought that this situation was caused by factors such as Yenice is a relatively small district and deprivation zone, high workload, heavy working conditions and management approach, and Ereğli and Devrek Forest Enterprises are relatively on the developed districts and main roads according to the region average.

Additionally, married people, who have wife working or not, have a high level of job satisfaction, whereas singles have lower levels of job satisfaction. In other words, the job satisfaction level of married employees was found to be higher in the Zonguldak Regional Directorate of Forestry and its affiliated units. On the other hand, the fact that the employees

have proxy duties as well as their primary duties decreases the total job level. This result is very normal because of often no additional fees for proxy duty, an extra additional burden, stress and trouble on employee.

#### **Correlations between Individual Features and Job Satisfaction**

The correlation between some individual features (age, gender, marital status, working status of the spouse, place of birth, education, task unit, duty position, total service duration-experience, duty duration, number of duty place, proxy duty) of the participants and their total job satisfaction levels was investigated by Correlation Analysis (Table 4).

| Total Job Satisfaction |
|------------------------|
| -0,026                 |
| -0,019                 |
| -0,048                 |
| 0,019                  |
| 0,009                  |
| -0,141**               |
| 0,069                  |
| -0,016                 |
| -0,018                 |
| 0,014                  |
| -0,049                 |
| -0,135*                |
|                        |

Table 4. Results of Correlation Analysis

\* is significant at the 0.05 confidence level; \*\* is significant at the 0.01 confidence level.

According to the results of the correlation analysis, there was a significant negative correlation  $(r = -0.141^{**})$  between the total job satisfaction and marital status variables at the 0.01 confidence level. This means that if employees are married, their job satisfactions increase. However, in a study by Yılmaz et al. (2009) at the Mersin Regional Directorate of Forestry, it was found out that singles had the highest job satisfaction, while married people had lower job satisfaction. In a study conducted by Yılmaz and Koçak (2008) for the employees of the Eastern Mediterranean Forestry Research Directorate; there was no statistically significant difference between the job satisfaction levels of the employees in terms of marital status. Similarly, in a study conducted by Çok et al. (2017) at the Elazığ Regional Directorate of Forestry; there was no statistically significant difference in terms of job satisfaction scale scores between single and married. In a study by Bölüktepe (1993), it was found that married workers had higher job satisfaction than single workers; however, there was no statistically significant difference.

There is a significant negative correlation ( $r = -0,135^*$ ) at the 0.05 confidence level between job satisfaction and proxy duty variables. In other words, job satisfaction decreases when employees are assigned their duties as a proxy duty.

#### 4. CONCLUSIONS AND RECOMMENDATIONS

At the end of the study, the following assessments and recommendations were made in order to increase the job satisfaction of the employees, to work more efficiently and to be successful;

- 1) Workplace conditions and work-related facilities/equipment must be adequate and of high quality.
- 2) Management approach should be based on merit, and a performance measurement and compensation system based on objective criteria should be applied.
- 3) It should be ensured that the person's work is respected and feeling comfortable with self-conscience himself from the work.
- 4) There should be arrangements (appreciation, premium, promotion, etc.) that will increase employee performance.
- 5) An effective management information system should be established in the subordinate relations.
- 6) The morale and motivation of the employees should be kept high. For this, superiors should have a good dialogue with the employees and take care of their various problems (health, etc.).

- 7) Managers need to be fair, to be respectful to their subordinates, to have a management license and to understand human psychology.
- 8) Employees must have the authority to make and implement decisions related to their work.
- 9) Employees should feel free and employee's integration with work should be ensured.
- 10) Employees must have a workload to carry out their work and family life balanced.
- 11) A system based on merit and performance should be applied in the promotion and displacement.
- 12) The effectiveness of rotation applications should be analyzed and a new regulation should be made.
- 13) An organization culture based on human rights and law should be established.
- 14) Balance and justice should be provided between the work and the wage.
- 15) Employee rights should be improved.
- 16) Employees should not be given proxy duty, and additional fees should be paid in case of proxy duty.
- 17) Work distribution among employees should be done in a balanced manner.
- 18) Employees of the institution's social facilities should benefit equally.
- 19) Necessary attention must be paid to informing the employees and in-service training.

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#### REFERENCES

- Akkamış, O. 2010. İlköğretim I. ve II. Kademe Öğretmenlerin İş Tatmini Üzerine Bir Değerlendirme. Yeditepe Üniversitesi Sosyal Bilimler Enstitüsü İstanbul.
- Aksu, G., Acuner, A. M. and Tabak R. S. 2002. Sağlık Bakanlığı Merkez ve Taşra Teşkilatı Yöneticilerinin İş Doyumuna Yönelik Bir Araştırma (Ankara Örneği). Ankara Üniversitesi, Tıp Fakültesi Mecmuası, Cilt:53, Sayı:4, Ankara.
- Akyüz, K. C., Koçak, S., Balaban, Y., Yıldırım, İ. and Gedik, T. 2011. Çalışanların iş tatmin düzeylerinin incelenmesi (Muğla Orman Bölge Müdürlüğü örneği). SDÜ Orman Fakültesi Dergisi, 12: 20-26, Isparta.
- Bozkurt, A., Daşdemir, İ., Karakaya, S. and Şahin, H. A. 2018. Sakarya İli Kavak Üreticilerinin İş Doyumunu Etkileyen Faktörler. Journal of Bartın Faculty of Forestry, 20 (3): 609-617. DOI: 10.24011/barofd.461799.
- Bölüktepe, E. F. 1993. Kamu Örgütlerinde İş Tatmini. Atatürk Üniversitesi. Sosyal Bilimler Enstitüsü. İşletme Anabilim Dalı. Erzurum.
- Çıtak, M. A., Koldere, Y., Ünsar, S. and Ergin, G. 2008. İstanbul İlinde Görev Yapan Kamu ve Özel İlköğretim Öğretmenlerinin Tatmin Düzeylerinin Belirlenmesin Yönelik Bir Araştırma. Trakya Üniversitesi, Sosyal Bilimler Dergisi, Cilt:10, Sayı:2.
- Çok, N., Göksu, E., Doğaner, A., Kalkan, B. and Güneş, Ö. 2017. Elazığ Orman Bölge Müdürlüğü Çalışanlarının İş Doyumu ve Bazı Bireysel Özelliklerinin İş Doyumuna Etkisi. *Turkish Journal of Forest Science*, 1(2) 2017: 155-168.
- Daşdemir, İ. 2019. Bilimsel Araştırma Yöntemleri (2. Basım). Nobel Akademik Yayıncılık ve Danışmanlık Tic. Ltd. Şti., Yayın No: 1536, ISBN 978-605-320-442-8, 210 s., Ankara.
- Davis, K. 1981. Human Behovior at Work (Sixth Edition). New Delhi: TATA Mc Graw Hill Publishing Company Ltd.
- Eren, E. 1996. Yönetim ve Organizasyon. Beta Yayın No: 4, İstanbul, 621 s.
- İncir, G. 1990. Çalışanların İş Doyumu Üzerine Bir Araştırma. MPM Yayın No: 401, Ankara.
- Kalıpsız, A. 1988. İstatistik Yöntemler. İstanbul Üniversitesi Orman Fakültesi Yayın No: 3522/394, İstanbul, 558 s.
- Özdamar, K. 2002. Paket Programlar İle İstatistiksel Veri Analizleri (4. Baskı). ISBN 975-6786-00-7, Kaan Kitabevi, 686 s., Eskişehir.
- Yılmaz, E. and Koçak Z. 2008. Doğu Akdeniz Ormancılık Araştırma Müdürlüğü Çalışanlarına Yönelik İş Doyumu Araştırması. Çevre ve Orman Bakanlığı Yayın No: 364, DOA Yayın No: 48,Çeşitli Yayın No: 8, 56 s., Tarsus.
- Yılmaz, E., Daşdemir İ., Karabulut S., Koçak Z. and Polat O. 2009. Orman Genel Müdürlüğü Taşra Teşkilatı Çalışanlarının İş Doyumunu Etkileyen Faktörler, Mersin Orman Bölge Müdürlüğü ve Buna Bağlı Orman İşletme Müdürlükleri Örneği. Çevre ve Orman Bakanlığı Yayın No: 387, DOA Yayın No:52, Teknik Bülten No:30, 65 s., Tarsus.
- ZRDF, 2017. Zonguldak Orman Bölge Müdürlüğü 2017 Yılı Çalışma Programı. Zonguldak.



# **ORAL PRESENTATION**

# Socioeconomic Analysis of Forest Crimes in the Ulus Region of Bartin

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Abstract: This study was carried out to examine the forest crimes in the Bartin province-Ulus State Forest Enterprise in the last 10 years in terms of their various characteristics, to evaluate the damage and compensation calculations in practice from a scientific point of view so as to contribute to the reduction of forest crimes and to the sustainable forest management. The data were taken from the crime records of the Ulus Forest Enterprise and its affiliated forest management chiefs, and the case files and expert reports on forest crimes in the Ulus Courthouse. Various characteristics of forest crimes were examined by tables and percentage method. Compensation accounts regulated by the forest enterprise and compensation accounts in case files were compared, the reasons of the differences, sufficiency and suitability of the compensation accounts to the scientific literature were discussed. In the last 10 years, the wood cutting, opening-settling, occupying and benefiting and fire crimes were mostly committed and the number of crimes reduced in recent years. There is no difference between the amount of compensation calculated by the forest enterprise and the compensation calculated by the experts for the crimes of cutting, opening-settling, occupying and benefiting. In the case of fire crime, the compensation calculated by the expert is 15.3 times less than that of the forest enterprise and this difference is due to the calculation of reforestation cost. In addition, it was found that there was a calculation approach focusing on the damages of wood raw material and the cost of seedling and reforestation, all damages in the forest ecosystem integrity were not taken into account, the compensation amounts were insufficient and were not in accordance with the Appraisal Methods of the Forest Assets in the scientific literature. According to these results, it was revealed that the damages and compensation calculations which take into account all forest functions in forest crimes should be calculated in accordance with the laws and scientific principles, and in this context, some suggestions were developed for implementation and sustainable forest management.

**Keywords:** Forest protection, sustainable forest management, forest crimes, damage and compensation account, Ulus Forest Enterprise Directorate

# **1. INTRODUCTION**

The actions, such as unauthorized tree cutting, holding, consuming, transporting, forest opening-settling, occupying and benefiting, grazing fire etc., which are harmful to forests and prohibited by laws, are considered as forest crimes. Although a significant number of forest crimes remain confidential, the number of forest crimes is 10,829 units / year at the 2013-2015 period in Turkey. On the other hand, 4.51 million steres woods were annually produced from the state forests by illegal ways between 2010 and 2015 (OGM, 2016). The rules were set up for the protection and continuity of forests, and forest crimes and how to calculate the loss were explained. In addition, these issues were explained with some laws and regulations. Despite these, forest crimes and destroyed forested areas have continued to increase; it has been a major problem for Turkey. Therefore, forest crimes should be analyzed in terms of socioeconomics and the amount of damages and compensation should be calculated correctly.

In Turkey, there are 7,095,650 people living in forests and adjacent to 21,375 forest villages (URL-1, 2019). Forest villagers constitute approximately 9% of Turkey's population, the least developed and poorest part of Turkey as socioeconomic and live as depend on forest. Forest villagers having unfavorable conditions can damage forests by actions such as tree cutting, grazing, opening, occupying and benefiting. On the other hand, migration from rural to large cities, distorted urbanization, squatters etc. cause settlings in forest areas. In addition, some actions such as fire, grazing, unauthorized tree cutting, uncontrolled mining, stone and sand quarries also damage the forests. Here, forest crimes are formed as a result of such actions that damage forests. It is essential to investigate the causes of forest crimes, to conduct socio-economic analysis well and to prevent them before they occur. However, after the crime occurred; the amount of damage and compensation should be calculated in accordance with the law and scientific principles. The prevention of forest crimes and the correct calculation of the compensation amount will contribute to the conservation and sustainable management of forests.

Several studies were carried out on forest crimes in Turkey (Sekendiz, 1980; Ayanoğlu, 1987; Kapucu, 1987; Gümüş, 1992; Ayanoğlu and Güneş, 2003; Güneş, 2004; Şen and Ünal, 2003; Erbakıcı, 2007; Bozaran and Görücü, 2017; Ayaz and İnanç, 2018). Additionally there are some publications (Fırat, 1971; Acun, 1976; Fırat and Miraboğlu, 1977; Miraboğlu, 1979; Geray, 1987; Ünal, 1990; Türker, 2013; Daşdemir, 2018) on the calculation of amount of damage and compensation for forest crimes in Turkey. However, there is no study examining the socioeconomic analysis of forest crimes, the damages and compensation calculations in the Ulus Forest Enterprise which is important for forestry activities and forest crimes in the Bartın province.

Therefore, this study was carried out to examine the forest crimes in the Ulus State Forest Enterprise in the last 10 years in terms of their various characteristics, to evaluate the damage and compensation calculations in practice from a scientific point of view so as to contribute to the reduction of forest crimes and to the sustainable forest management.

# 2. MATERIAL AND METHOD

This study was carried out in the Ulus Forest Enterprise located in the Ulus District of Bartın. The study area of the Ulus Forest Enterprise includes the Ulus District, Abdipaşa and Kumluca townships. There are 6 forest management chiefs (Ulusçayı, Abdipaşa, Sökü, Kumluca, Ardıç and Drahna) at the Ulus Forest Enterprise (Figure 1).



Figure1. Study area of the Ulus Forest Enterprise

The total area of the Ulus Forest Enterprise is 77,891 hectares. 68% (52,616.3 ha) of this are forest. The total population in the study area is 21,116 (UOİM, 2018a). In the study, the data of the research is composed of the information obtained from the crime records of the Ulus Forest Enterprise and its affiliated forest management chiefs. In addition, the data obtained from the case files and expert reports that were transferred to the Ulus Courthouse were used as data. According to the data obtained in the last 10 years, forest crimes were explained by tables and percentage method in term of various characteristics. Compensation accounts regulated by the forest enterprise and compensation accounts in case files were compared, the reasons of the differences, the scientific accountability of the compensation accounts, the adequacy and the suitability of social justice were discussed. Damage and compensation calculations were made on the current prices of that year. Excel computer program was used to evaluate the data.

# 3. RESULTS AND DISCUSION

# Theoretical Foundations of Loss and Compensation Accounts in Forestry

Damage and compensation calculations in forestry are scientifically and theoretically examined under these headings (Daşdemir, 2018): In case of exploitation of mineral assets in forest land, roads and power lines pass through the forest, fire in forests storm, snow and frost damage, grazing damage, insect and mushroom damage, gas damage, illegal tree cuttings.

Above damage and compensation calculations in forestry are carried out with the help of approaches and methods, which was known as *Appreciation of Forest Assets* and its scientific foundations located in the related literatures (Firat, 1971; Firat and Miraboğlu, 1977; Günel, 1988; Türker, 2013; Daşdemir, 2018). In these methods, compound interest calculation method is used and the formulas which give the sum of the finite and infinite geometric series are used. These methods, which are very large in scope, are not described here as they exceed the scope of the paper. However, the forest crimes in the Ulus Forest Enterprise were examined in terms of various characteristics and the damages and compensation calculations related to some crimes were exemplified.

# Forest Crimes in the Ulus Forest Enterprise Directorate

The forest crimes that occurred in the Ulus Forest Enterprise during 2008-2019 period and transferred to Ulus Courthouse were determined as unauthorized tree cutting, transporting, holding, consuming, opening-settling, occupying-benefiting, fire and stubble incineration (Table 1 and 2).

| Chieffainay Cutting Transporting | Halding                      | Conguming | Opening-  | Occupying-        | Fire | Stubble Incineration | Total              |        |     |      |
|----------------------------------|------------------------------|-----------|-----------|-------------------|------|----------------------|--------------------|--------|-----|------|
| Cillentanicy                     | traincy Cutting Transporting | Holding   | Consuming | Settling Settling |      | гпе                  | Stubble memeration | Number | %   |      |
| Ulusçayı                         | 23                           | 1         | 0         | 2                 | 4    | 32                   | 4                  | 5      | 71  | 24.5 |
| Abdipaşa                         | 28                           | 5         | 6         | 0                 | 1    | 10                   | 10                 | 2      | 62  | 21.4 |
| Sökü                             | 14                           | 4         | 1         | 0                 | 1    | 5                    | 3                  | 1      | 29  | 10.0 |
| Kumluca                          | 32                           | 3         | 8         | 7                 | 17   | 7                    | 6                  | 7      | 87  | 30.0 |
| Ardıç                            | 0                            | 0         | 0         | 0                 | 0    | 0                    | 0                  | 0      | 0   | 0.0  |
| Drahna                           | 26                           | 1         | 1         | 1                 | 4    | 7                    | 1                  | 0      | 41  | 14.1 |
| Total                            | 123                          | 14        | 16        | 10                | 27   | 61                   | 24                 | 15     | 290 | 100  |
| %                                | 42.4                         | 4.8       | 5.5       | 3.4               | 9.3  | 21.0                 | 8.3                | 5.2    | 100 |      |

**Table 1.** Forest crimes by forest management chiefs and types (2009-2018)

Accordingly, the highest number of wood cutting crime (42.4%) was observed in the Ulus Forest Enterprise during the last 10 years. this was followed by 21% occupying-benefiting, 9.3% opening-settling, 8.3% fire, 5.5% holding, 5.2% stubble incineration, 4.8% transporting and 3.4% consuming crime. The most crime (30%) is Kumluca Forest Management Chieftaincy. Ardıç Forest Management Chieftaincy has not any crime. The most important reason for this is that Kumluca has more population than the other chiefs and is not a settlement unit within the borders of Ardıç Chieftaincy.

**Table 2.** Forest crimes as of years and types

| Years | Cutting | Transporting | Holding | Consuming | Opening-<br>Settling | Occupying-<br>Benefiting | Fire | Stubble<br>Incineration | Total | %    |
|-------|---------|--------------|---------|-----------|----------------------|--------------------------|------|-------------------------|-------|------|
| 2009  | 26      | 4            | 6       | 1         | 8                    | 3                        | 4    | 0                       | 52    | 17.9 |
| 2010  | 16      | 3            | 2       | 4         | 2                    | 8                        | 0    | 1                       | 36    | 12.4 |
| 2011  | 17      | 4            | 2       | 0         | 2                    | 18                       | 5    | 0                       | 48    | 16.6 |
| 2012  | 7       | 0            | 3       | 1         | 1                    | 3                        | 7    | 8                       | 30    | 10.3 |
| 2013  | 16      | 1            | 0       | 0         | 0                    | 3                        | 4    | 0                       | 24    | 8.3  |
| 2014  | 6       | 1            | 0       | 0         | 3                    | 13                       | 0    | 1                       | 24    | 8.3  |
| 2015  | 12      | 0            | 3       | 1         | 4                    | 9                        | 0    | 3                       | 32    | 11.0 |
| 2016  | 7       | 0            | 0       | 3         | 4                    | 3                        | 2    | 1                       | 20    | 6.9  |
| 2017  | 11      | 1            | 0       | 0         | 2                    | 0                        | 2    | 1                       | 17    | 5.9  |
| 2018  | 5       | 0            | 0       | 0         | 1                    | 1                        | 0    | 0                       | 7     | 2.4  |
| Total | 123     | 14           | 16      | 10        | 27                   | 61                       | 24   | 15                      | 290   | 100  |
| %     | 42.4    | 4.8          | 5.5     | 3.4       | 9.3                  | 21.0                     | 8.3  | 5.2                     | 100   |      |

According to Table 2, although there is no significant difference in the distribution of crimes by years, the highest number of crimes occurred in 2009, followed by 2011 and 2010, 2015 and 2012 years. In recent years, there has been a significant decrease in the number of crimes and it has been minimized in 2018. The most important reason for this is rural migration.

# Analysis of Litigation-Tracking Information of Ulus Forest Enterprise

In the period 2009-2018, a total of 429 cases were finalized in the Ulus First Instance and Criminal Courts (Table 3). Tree cutting crime is the first (39.6%), opening-settling crime is second (19.6%), fire crime is third (18.6%), occupying and

benefiting is fourth (15.6%). The most cases (73 cases) were examined in 2009. In parallel with the decrease in the number of crimes in recent years, the total number of cases has decreased.

| Criminal Cases | Cutting | Transporting | Opening-Settling | Occupying- Benefiting | Fire | Other | Total |
|----------------|---------|--------------|------------------|-----------------------|------|-------|-------|
| Total          | 170     | 22           | 84               | 67                    | 80   | 6     | 429   |
| %              | 39.6    | 5.1          | 19.6             | 15.6                  | 18.6 | 1.4   | 100   |

| Table 3  | Criminal | case | statistics | hv | Ullus | Courthouse  | <b>TIA</b> | 2019   | ۱ |
|----------|----------|------|------------|----|-------|-------------|------------|--------|---|
| Table J. | CImmai   | case | statistics | Dy | Ulus  | Courtinouse | UA         | , 4UI) | , |

According to the examination of the case files in Ulus Courthouse, 65% of forest crimes were determined by forestry officers, 25% by written orders and 10% by notification. These results indicate that the local people are not sufficiently sensitive to report the forest crimes to the relevant authorities.

# Compensation Accounts for Forest Crimes in the Ulus Forest Enterprise Compensation Accounts on Tree Cutting Crime

In 2015, the forest enterprise compensation report for 11 sterling oak trees without any interruption in 0.02 ha area was calculated as follows according to Articles 113 and 114 of the Forest Law No. 6831 (OK, 2015);

Cost of Cut Oak= Oak Quantity x Unit Price = 11 steres x 75 TL/steres = 825 TL,

Afforestation Cost= Damaged Area x Afforestation Unit Price = 0.02 ha x 12,481 TL/ha 249.62 TL,

Total Compensation Amount (a + b) = 1074.62 TL

In the expert report submitted to the Court in 2016 for the same offense, the total amount of compensation was found as 1074.62 TL using the same logic and method according to Articles 113 and 114 of the Forest Law. In other words, there is no difference between the amount of compensation calculated by the forest enterprise and the amount of compensation calculated by the experts in the courts.

#### **Compensation Accounts on Opening-Settling Crime**

In 2017, 0.0169 ha of forest area was opened and 6 mm diameter concrete bucket and cover were placed. In the compensation report of the forest enterprise, the amount of the damage was calculated only as forestation expense;

*Total Compensation Amount* = Opening Area x Afforestation Unit Price = 0.0169 ha x 17,420 TL = 294.40 TL

In the expert report submitted to the Court in 2018 for the same offense, the amount of compensation was calculated as 294.40 TL by using the same logic and method only. There is no difference between the amount of compensation calculated by the forest enterprise and the amount calculated by the experts in the courts.

# **Compensation Accounts for Occupying and Benefiting**

In 2010, it was determined that there was a total of 0.019616 hectares of occupation and exploitation in the area of catchment and pipe area where spring water emerged. The compensation and compensation amount in the compensation report issued by the forest enterprise as follow;

Total Compensation Amount = Occupied Area x Afforestation Unit Price = 0.019616 ha x 6,695 TL = 131.33 TL

In the expert report submitted to the Court in 2012 regarding the same case, the amount of damage and compensation was similarly calculated as 131.33 TL. Therefore, there is no difference between the amount of compensation calculated by the forest enterprise and the amount of compensation calculated by the experts in the courts.

# **Compensation Accounts in Fire Crime**

In the compensation report prepared by the forest enterprise in relation to the fire in the 3.6 ha larch area, the loss and compensation amount is calculated as follows, based on the articles 112, 113 and 114 of the Forest Law.

Afforestation Cost= Burned Area x Afforestation Unit Price = 3.6 ha x 5,172 TL/ha 18,691.20 TL

Extinguishing Expense= Worker Cost (Number of Extinguishing Workers x Worker Premium Price) + Vehicle Cost = 2.35 (1 x 2.35 TL) + 7.30 TL = 9.65 TL
Sapling Management Damage= Number of Dried Plants in Burned Area x Unit Price x 2 = 33 x 10 x 2,660 TL

Total Fire Compensation (a + b + c) = 19,360.85 TL.

In 2013, in the expert report submitted to the Court on the same case, the damage and compensation calculations were made as follows:

Afforestation Cost= 359.77 TL

Extinguishing Expense=9.65 TL

Sapling Management Damage= 898.5 TL

Total Fire Compensation (a + b + c) = 1,267.92 TL.

While the forest enterprise has calculated 19,360.85 TL for the fire compensation, the expert has calculated 1,267.92 TL. The compensation calculated by the expert is 15.3 times less than that of the forest enterprise. In both calculations, the extinguishing expense is the same. The expert accounted for the damage of the seedlings, and also added to the calculations the damages of soil, suceyrat, early cut and yield. For afforestation cost; While the forestry operation amounted to 3.6 ha as the amount of burning area, the expert has calculated both the amount of burning area (0.075 ha) and the unit price low, and therefore, the afforestation cost was very low.

### 4. CONCLUSIONS AND RECOMMENDATIONS

At the end of the study, some assessments and recommendations were made. According to this, in the last 10 years, 290 forest crimes occurred in the Ulus Forest Enterprise. Illegal tree cutting, opening-settling, occupying-benefiting and fire crimes mostly occurred. In recent years, the number of crimes decreased and decreased to the minimum in 2018.

In accordance with Articles 112, 113 and 114 of the Forest Law, damage and compensation calculations are based on unit prices determined by OGM in the current year and accepted by courts. There is no difference between the compensation of the forest enterprise and the compensation calculated by the experts in tree cutting, opening-settling, occupying and benefiting crimes. However, in the case of fire crime, the expert's compensation is 15.3 times lower than that of the forest enterprise. This difference allows the expert to keep both the amount of burning area and the plantation unit price low.

In the compensation calculations, there is a calculation approach which is mainly focused on the cost of wood raw material and sapling and the cost of afforestation of the field. There is no approach taking into account all the damages (nature conservation, climate and water regime regulation, water supply, erosion prevention, biodiversity conservation, carbon storage, hunting and wildlife service, etc.) that may occur within the forest ecosystem integrity.

It is understood that there is no use of the approaches and methods that are known as *Appreciation of Forest Assets* in forestry and their scientific foundations are in the related literature. For this reason, legal arrangements should be based on the methods of *Appreciation of Forest Assets* in the scientific literature.

#### Acknowlegment

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#### REFERENCES

- Acun, E. 1976. Orman Yangınlarında Zarar ve Kayıpların Hesaplanması İle İstenecek Tazminatın Saptanması Kuralları. İÜ, Orman Fakültesi Dergisi, Seri B, Sayı 2, s.86-103, İstanbul.
- Ayanoğlu, S. 1987. 3116 Sayılı Orman Kanunu ve Sonrası, Cumhuriyet Dönemi Ormancılığımızda 3116 Sayılı Orman Yasası ve Sonrası Sempozyumu 7 Şubat 1987, Ankara, Türkiye Ormancılar Derneği Yayın no:10, 21-33.

- Ayaz, H. and İnanç, S. 2018. Orman Suçlarında Tazminat ve Ağaçlandırma Bedeli Hesabı Artvin Çoruh Üniversitesi Orman Fakültesi Dergisi ISSN:2146-1880, e-ISSN: 2146-698X Yıl: 2018, Cilt: 19, Sayı:2, Sayfa:175-185.
- Ayanoğlu, S. and Güneş, Y. 2003. Orman Suçları Ders Kitabı. İstanbul Üniversitesi Orman Fakültesi Yayınları, Rektörlük Yayın No:4420, Orman Fakültesi Yayın No:471, İstanbul.
- Bozaran, M. A. and Görücü, Ö. 2017. Adıyaman Orman İşletme Müdürlüğünde Orman Suçlarının Ormancılık Hukuku Açısından İncelenmesi. *Turkish Journal of Forest Science*, 1(2) 2017: 112-133.
- Daşdemir, İ. 2018. Ormancılık İşletme Ekonomisi (4. Baskı). Bartın Üniversitesi Yayın No: 10, Orman Fakültesi Yayın No: 6, ISBN 978-605-60882-8-5, 407 s., Bartın.
- Erbakıcı, F. 2007. Orman Suçlarıyla Mücadelede Kolluğun Etkinliğinin Arttırılması. Kara Harp Okulu Savunma Bilimleri Enstitüsü, Güvenlik Bilimleri Anabilim Dalı Yüksek Lisans Tezi, 186 s., Ankara.
- Fırat, F. 1971. Ormancılık İşletme İktisadı. İÜ, Orman Fakültesi Yayın No:156, 336 s., İstanbul.
- Fırat, F. and Miraboğlu, M. 1977. Orman Kıymetlerinin Takdirinde Kullanılan Formüller ve Uygulanmasına Ait Örnekler. İÜ, Orman Fakültesi Yayın No:226, 123 s., İstanbul.
- Geray, A. U. 1987. Yatağan Termik Santralinin Çevredeki Ormanlarda Yaptığı Zararların Hesaplanması. Çevre ve Ormancılık Dergisi, Yayın No: 3, 20 s., Ankara.
- Günel, A. 1988. Ormancılık İşletme Ekonomisi. KTÜ, Orman Fakültesi Ders Teksirleri Serisi No: 26, 168 s., Trabzon.
- Güneş, Y. 2004. Orman Suçlarının Ceza Hukuku Açısından İncelenmesi İÜ, Orman Fakültesi Dergisi, Seri A, Sayı 1, s.144-163, İstanbul.
- Gümüş, C. 1992. Amasya Orman Bölge Müdürlüğünde Orman Suçlarının Nedenleri. KTÜ, Fen Bilimleri Enstitüsü, Doktora Tezi, Trabzon.
- OGM, 2016. Orman Genel Müdürlüğü Stratejik Plan 2017-2021. 70 s., Ankara.
- OK, 2015. 6831 Sayılı Orman Kanunu (2015 yılında değişik). Yayımlandığı Resmi Gazete Tarih: 8.9.1956, Sayı: 9402, Yayımlandığı Düstur Tertip: 3, Cilt: 37, Sayfa: 2457.
- Kapucu, F. 1987. Ormancılık Bilgisi (Orman ve Ormancılıkta Temel Kavramlar). KTÜ, Orman Fakültesi Ders Notu, Trabzon.
- Miraboğlu, M. 1979. Ormanlık Alanlarda Açık Maden İşletmelerinde Tazminat Hesabı. OGM Yayın No: 624/16, Ankara.
- Şen, G. and Ünal, S. (2003). Orman Suçlarının Nedenleri Üzerine Bir Araştırma (Karadere Orman İşletme Müdürlüğü Örneği. Kafkas Üniversitesi, Artvin Orman Fakültesi Dergisi, (2003) : 1-2 (41-51).
- Sekendiz, O. A. 1980. Orman Korunması Ders Notları, KTÜ, Orman Fakültesi Ders Notları Serisi, Yayın No:33, Trabzon.
- Türker, M. F. 2013. Ormancılık İşletme Ekonomisi (Güncellenmiş ve Genişletilmiş 2. Baskı). Ormancılık ve Tabiatı Koruma Vakfı Yayın Nu:5, ISBN 978-605-85748-0-9, 287 s., Trabzon.
- UA, 2019. Ulus Adliyesi UYAP Ekranı Sistemi ve Arşivleri (2009-2018). Ulus.
- UOİM, 2018a. Ulus Orman İşletme Müdürlüğü Brifing Dosyası Notları (2009-2018). Ulus.
- UOİM, 2018b. Ulus Orman İşletme Müdürlüğü Arşivleri 2009-2018 Zabıt Tutanakları. Ulus.
- URL-1, 2019. Orman Genel Müdürlüğü Web Sitesi. https://www.ogm.gov.tr/SitePages/OGM/OGMBiliyormuydunuz, (15.02.2019).
- Ünal, S. 1990. Orman Yangınlarından Doğan Zarar Düzeyinin Hesaplanması Üzerine Araştırmalar. İÜ, Fen Bilimleri Enstitüsü Yüksek Lisans Tezi, 131 s., İstanbul.



# Tree Flora of Mount Ledang, Johor National Park, Malaysia

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**Abstract:** A field survey was conducted in the forest trails of Gunung Ledang, Johor National Park to study the forest communities and provide a check-list of tree flora occur in the area. The areas surveyed encompassed a diverse range of forest types from riparian vegetation, lowland, hill and upper hill dipterocarp forests up to lower and montane forests. From the survey, a total of 62 tree families were encountered which consist of 62 families, 143 genera and 222 taxa. Overall, the areas are rich with the tree species from the families of Myrtaceae, Euphorbiaceae, Moraceae, Dipterocarpaceae and Rubiaceae. Common montane taxa encountered include trees from Podocarpaceae family (i.e., *Podocarpus neriifolius, Dacrydium elatum* and *Dacrydium beccarii*. An interesting finding was the discovery of *Maesa fraseriana* (Maesaceae) in the upper montane forest of Gunung Ledang which can potentially be a new record for the area. Therefore, the work presented here may serve as a first step in assessing the conservation importance and long term strategies for biological diversity.

Keywords: Gunung Ledang, Florasitic composition, Maesa fraseriana

# **1. INTRODUCTION**

Gunung Ledang or also known as Mount Ophir is a mountain situated at Gunung Ledang, Johor National Park. The mountain is standing at an altitude of 1,276 m above sea level in the area of 8,675.2 ha, and is located between the states of Johor and Malacca. Gunung Ledang was gazzetted as a National Park in 2003. The park holds four distinct vegetation types which include lowland dipterocarp forest, hill dipterocarp forest, lower montane and montane ericaceous. In Peninsular Malaysia, montane rainforest communities are scattered and few. With the exception of Cameron Highlands, they are mainly minimally-disturbed, undisturbed or totally protected such as Gunung Benom in Krau Wildlife Reserve (A. Latiff and Mohd Shaffea, 2011) and Gunung Tahan in Taman Negara Pahang.

At least two formations are developed in major mountains, i.e., lowland forest and montane forest. Montane forest and lowland forest are distinct floristically. They can be distinguished by a number of structural characters which include the size of canopy height, canopy layer, leaves, the presence of vascular and non-vascular epiphytes and climbers. The montane forest also differs from lowland in having fewer and smaller emergent trees, flattish canopy surfaces, gnarled limbs and denser subcrowns (Whitmore, 1984).

Due to the variation in biogeography, habitat and diturbance, tree species in tropical forest varies greatly from place to place in terms of composition and diversity (Whitmore, 1998). In response to an obvious lack of current information on tropical montane of Gunung Ledang Johor National Park, this paper identifies the major forest types and tree communities in Gunung Ledang, and presents the list of tree species collected in the expedition areas ranging from riparian forest trails, dense and species-rich lowland dipterocarp forests and extending to hill, upper hill dipterocarps and montane trails. Gunung Ledang was chosen due to its unique plant communities as well as its virgin reserve jungle (VJR) status. Such basic information is of importance to the understanding of the species distribution, conservation requirements and economic potential of tree resources which may contribute towards developing and managing the available resources on a sustainable basis. In addition, results from this study may provide baseline data for biological conservation and management.

### 2. MATERIAL AND METHODS

Botanical specimens were collected at two trails of Gunung Ledang Johor National Park which include along the riparian zones near the park rangers' office and the trail before Damn Base Camp to the upper montane forest at the Plateau Base Camp and continued the highest point where the telecommunication tower was erected (Figure 1). During the survey, attempts were made to make a collection of fresh leaves along with flowers or fruits with the assistance from a tree climber. As collecting complete specimens from canopy tree is often difficult, fallen leaves, fruits and flowers were collected from the ground. Specimens collected, where possible, were identified during the collection. A GPS receiver was used to determine the topographic elevation of the specific points in the trails. Trail elevation profile was then produced in relation to common species occurrence in relation to the altitude.

All specimens collected were kept in the herbarium laboratory at the Centre of Biodiversity and Sustainable Development, Universiti Teknologi MARA, Puncak Alam campus. Whenever possible, the conservation status of the species was crosschecked with the information from the IUCN Red List (IUCN, 2013). The IUCN red list provides taxonomic, conservation status, and distribution information on taxa that are facing a risk of extinction.



**Figure 1**. Topographic map of Gunung Ledang, Johor National Park shows the topography, expedition trail (blue dotted lines) and base camp location is shown in red triangles.

#### 3. RESULTS AND DISCUSSION

## Forest Communities and Tree Flora of Gunung Ledang, Johor National Park Family of Trees

The tree flora collected and identified from Gunung Ledang, Johor National Park were alphabetically listed in Appendix 1. The Gunung Ledang Johor National Park area holds a high diversity of trees with a total number tree families enumerated along the study trails amounted to 62. Figure 2 shows the distribution of tree families occur in the study trails in relation to the number of taxa. Observations of specimens indicated that the tree families are consisting of 143 genera and 222 taxa. Generally, the areas surveyed are rich with the species from the family of Myrtaceae. Overall, a total of 24 taxa which belong to Myrtaceae family were encountered in the study trails. The second was the family of Euphorbiaceae with 21 taxa, followed by Moraceae (18 taxa) and Dipterocarpaceae (17 taxa) and Rubiaceae (15 taxa). The next five important tree families occur in the study trails include Theaceae, Guttiferae, Apocynaceae, Leguminosae and

Hypericaceae, with a number of taxa ranged from 10 - 13. Twelve families recorded the number of taxa between 5 - 9, and the remaining 40 families with the range of 1 - 4 taxa (Figure 2).



Figure 2. Family-wise distribution of tree species collected from Gunung Ledang, Johor National Park.

### **Riparian Forests**

Riparian forests are narrow, ribbon-like corridors that occur adjacent to many streams (Baker *et al.*, 2002). The ecological structure and function of riparian forests and the associated streams are profoundly intertwined. According to Damasceno-Junior *et al.* (2005), variations in topography, landform and soils in riparian forests have strong effects on species composition, distribution and structure. Near the park rangers' office of Gunung Ledang Johor National Park are riparian forests that harbor some interesting plant life. The river flows down through rocky mountain and many cascades at different heights which created many small pools. Smaller trees and treelets that are especially common in the moist areas include *Ixora* sp., *Rennellia elliptica, Canthium didymum* (Rubiaceae), *Barringtonia macrostachya* (Lecythidaceae), *Baccaurea parviflora* (Phyllanthaceae), *and Microcos latifolia* (Tiliaceae). Beside these, *Saraca multiflora* (Leguminosae) was frequent and easy to identify by its purple young leaves hanging from the ends of the branchlets.

Tree species from Dipterocarpaceae (i.e., *Shorea multiflora*), Moraceae (i.e., *Artocarpus elasticus*), Sapotaceae (i.e., *Palaquium obovatum*) and Leguminosae (i.e., *Sindora coriacea* and *Dialium platysepalum*) are big trees encountered on farther away from the stream. *Dillenia reticulata* (Dilleniaceae) can be identified from its big obovate leaves and stiltroots. Other medium-sized tree species found include *Knema scortechinii* (Myristicaceae), *Diospyros styraformis* (Ebanaceae) *and Ixonanthes reticulata* (Ixonantaceae). *Knema* is distinguished by its red sap produced from the stem from a slight incision made on the bark whereas *Diospyros* by its distinctive black bark, and *Ixonanthes* by its bole which often fluted at the base. In the openings or gaps of the stream banks, it is not usual to find pioneer species from Euphorbiaceae (i.e., *Croton argyratus, Croton laevifolius* and *Macaranga* sp.).

#### Lowland, Hill and Upper Hill Dipterocarp Forests

The expedition trail and its elevation profile for the journey to the peak of Gunung Ledang are presented in Figures 1 and 3. The exploration began from the lowland dipterocarp forest zone at 257 m altitude. As the name implies, the Dipterocarpaceae are mainly lowland rainforest trees. Trees from this family are the most important timber in Malaysia. A number of *Dipterocarpus* and *Shorea* were found in this area. These include *Dipterocarpus crinitus*, *Dipterocarpus cornutus*, *Dipterocarpus kerrii* and *Dipterocarpus kunstleri*. Five species of *Shorea* encountered along the trails were *Shorea leprosula*, *Shorea macroptera*, *Shorea parvifolia*, *Shorea pauciflora* and *Shorea ovalis*. Other dipterocarps found within this elevation include *Anisoptera curtisii*. Trees from Dipterocarp group found in this area include *Koompassia malaccensis* (Leguminosae), *Ochanostachys amentacea* (Olacaceae) and *Dyera costulata* (Apocynaceae).

Small and medium-sized trees that are common in this area include *Cinnamomum iners* (Lauraceae), *Gardenia tubifera* (Rubiaceae), *Gnetum gnemon* (Gnetaceae), *Memecylon cantleyi* (Melastomaceae) and *Pimeleodendron griffithianum* (Euphorbiaceae). *Cinnamomum* can be identified by its aromatic smell from the inner bark and its trinerved leaf characters. Other than timber trees, many other groups of plants contribute to the complexity of the lowland dipterocarp forests. For example, a variety of bamboo, rattans and palms are especially common as well as climbers and epiphytes.



Figure 3. Trail elevation profile and some common genus encountered.

Different tree communities occurred in the higher elevation along the trail to the peak of Gunung Ledang. In the hill dipterocarp forests, *Shorea platyclados* (Dipterocarpaceae) is common. Tree species such as *Cratoxylum formosum* (Hypericaceae), *Syzygium filifrome* (Myrtaceae), *Calophyllum* sp. (Guttiferae) and *Gluta wallichii* (Anacardiaceae) grow occasionally in this zone. *Cratoxylum* is distinctive from its iodine-colored sap from the inner bark and its prickly stem. *Syzygium* can be indentified from its simple opposite leaves arrangement and jambu smell characters, whereas *Calophyllum* also from its simple and opposite leaf arrangement but with parallel secondary veins. Meanwhile, *Gluta* can be distinguished from other trees by its black sap from the inner bark.

In the open sites of hill dipterocarp forest, some pioneer species such as *Macaranga gigantea*, *Macaranga triloba*, *Sapium baccatum*, *Endospermum diadenum* (all from Euphorbiaceae) and *Cratoxylum cochinchinense* (Hypericaceae) are common. Together with these secondary species, a number of medium-sized non-dipterocarp trees such as *Lithorcarpus* sp. (Fagaceae), *Scaphium macropodum* (Sterculiaceae) and *Artocarpus scortechinii* (Moraceae) were encountered along the hill dipterocarp forests.

Within the upper hill dipterocarp forest, tree species of *Shorea* (Dipterocarpaceae) are very common big trees along the trails. These include *Shorea curtisii* and *Shorea excelliptica*. *Shorea curtisii* is a typical member of upper hill forest habitat, specifically in valleys of the hill. It is a large emergent tree with a straight and fissured bole. Together with these emergent dipterocarps are a number of non-dipterocarp trees such as *Gynotroches axillaris* (Rhizophoraceae), *Elaeocarpus floribundus* (Elaeocarpaceae) and *Tristaniopsis razakiana* (Myrtaceae). *Tristaniopsis* is distinctive in terms of its peeling barks with mixed reddish brown to grey-white hues. Smaller trees occasionally encountered along the upper hill trails include Randia scortechinii, Timonius wallichianus (Rubiaceae), *Scutinanthe brunnea* (Burseraceae) and *Gynotroches axillaris* (Rhizophoraceae).

# Lower Montane Forest

During the expedition, lower montane forest was observed before the peak of trails with the presence of lower montane vegetation (e.g., *Ploiarium alternifolium* – Bonnetiaceae) at 1,018 m altitude. At this altitude, dipterocarps and other common lowland families such as Leguminosae, Euphorbiaceae and Myristiceae begin to diminish and are replaced by a

diversity of species from tree families such as Myrtaceae and Theaceae At this elevation, *Baeckea frutescens*, *Leptospermum flavescens* (Myrtaceae) and *Eurya nitida* (Theaceae) form the main tree association. At 1,030 m altitude, the forest is mossy and is characterized with low-statured vegetation and devoid of emergent trees.

## **Upper Montane Forest**

The upper montane zone occurs on area of telecommunication tower at the end of the trail (1,190 m altitude) which is among highest peaks in Gunung Ledang. Within the zone, trees from the family of Podocarpaceae are very common montane taxa such as *Podocarpus neriifolius*, *Dacrydium elatum*, *Dacrydium beccarii*. They are also much encountered along the trails before the summit. Together with these communities are *Magnolia montana* (Magnoliaceae), *Ardisia retinervia* (Myrcinaceae) and *Mastixia retinervia* (Cornaceae) which distinctly common around the summit zone. This elevation zone also supports a large variety of pitcher plants. Other montane taxa such as *Clerodendrum* sp. (Lamiaceae), *Schima wallichii* (Theaceae), *Leptospermum flavescens* (Myrtaceae) as well as *Ficus* cf. *sinuata* (Moraceae) also occur.

## Maesa fraseriana - A Potential New Record for Gunung Ledang, Johor National Park

*Maesa fraseriana*, belongs to Maesaceae, is a small shrub (up to 2 m tall) or woody climbers (up to 7 m tall) that is endemics to montane forest of Fraser's Hill. Utterridge (2012) reported that this species is known from five localities based on ten collections. All collection was from Fraser's Hill except for a single outlier from Ulu Klang. However, according to Utterridge (2012), it is possible that *Maesa fraseriana* is found within the central range of Peninsular Malaysia with an extent of occurrence of the collection within 1,000 km<sup>2</sup>. Due to the evidence of habitat decline in Fraser's Hill, this species was assigned a rating of Endangered B1 ab (iii).

It is interesting to note that, during this expedition, this taxon was found at the moist forest edge (1,048 m altitude) near the road side of Plateau Base Camp, together with other montane taxa such as *Dacrydium elatum*, *Eurya nitida* and *Podocarpus neriifolius*. Identification of this species was made possible from a consultation with FRIM's herbarium personnel and a thorough literature review. According to Kiew (1992), as compared to lowland forests, endemic species in montane forest may only be confined to a single mountain peak or group of peaks in which in this expedition, this phenomenon is illustrated by the discovery of *Maesa fraseriana*. In contrast, the geological distribution for the species in the lowland forests is wider. Therefore, the discovery of a new record from the montane forest may have profound implications of conservation as disturbance in the small habitat could affect the survival of the species.

## **Other Species of Interest**

Gunung Ledang National Park is an interesting area in terms of geographical variation, species composition and conservation potential. It shelters species that are endemics and endangered not to the area but also from other parts of Malaysia. The IUCN Red List data was used to provide the information on the conservation status of the listed tree species collected from this survey. In terms of conservation status, among all trees documented, *Anisoptera curtisii* and *Dipterocarpus cornutus* are assigned as Critically Endangered (IUCN, 2013). The other four species from the family of Dipterocarpaceae, *viz*; *Shorea leprosula, Shorea platyclados, Shorea pauciflora* and *Dipterocarpus kerrii* are reported to be endangered (Appendix 1) which may require a combination of sound research and some conservation attention.

## 4. CONCLUSIONS

From this expedition, it can be concluded that Gunung Ledang Johor National Park area covers a diverse range of geographical elements and natural vegetation communities from riparian vegetation, lowland, hill and upper hill dipterocarp forests up to lower upper montane forests. This National Park is likely to harbour a significant number of endemic species, however the data collected from this expedition were inadequate to document comprehensive information on species richness, endemism and checklists of rare and endangered species for the area. While information from this survey may provide reference for ecologically useful species as well as species of special concern, a follow-up plant inventories are necessary to assess the conservation importance of a particular species.

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#### REFERENCES

- A. Latiff, M and Mohd Shaffea, L. 2011. Introduction. *In*: Gunung Benom: Krau Wildlife Reserve Pahang: Geology, Biodiversity and Socio-Economic Environment, Academy of Science Malaysia and Department of Wildlife and National Parks, ix-xii.
- Baker, J.R., Ringold, P.L. and Bollman, M. 2002. Patterns of Tree Dominance in Coniferous Riparian Forests, Forest Ecology and Management, 166: 311-329.
- Damasceno-Junior, G.A., Semir, J., Santos, F.A.M.D, and H.F. Leitâo-Filho, H.F. 2005. Structure, distribution of species and inundation in a riparian forest of Rio Paraguai, Pantanal, Brazil, Flora, 200, pp. 119-135.
- IUCN, 2013. The IUCN Red List of Threatened Species. Web page, available at http://www.iucnredlist.org/amazing-species, retrieved date 27 August 2013.
- Kew, R. 1992. The Montane Flora of Peninsular Malaysia: Threats and Conservation. Background Paper, Malaysian National Conservation Strategy. Economic Planning Unit, Kuala Lumpur

Utteridge, T. M. A. 2012. Four New Species of Maesa Forssk. (Primulaceae) from Malesia. Kew Bulletin. 67 (3): 367-378

Whitmore, T.C. 1984. Tropical Rain Forests of the Far East. (2nd edition). Oxford University Press, Oxford. 352 pp.

Whitmore, T.C. 1998. An Introduction to Tropical Rain Forests, 2<sup>nd</sup> Edition, Oxford University Press, New York.



# Quality of Urban Life in Development Areas: An Assessment of Citizens in Kastamonu, Turkey

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Abstract: People requires some certain conditions in urban areas in every period of life. In addition to basic needs that you must have to survive, there are requirements in which the level of satisfaction desired to be reached depends on subjective values. Urban quality of life, which has different targets according to culture and value system, is used as a tool for realizing sustainable practices based on scientific data-based policies especially in order to determine urban planning decisions. Since the concept of quality of life is affected by all factors of life, the indicators used for measurement are context-dependent. However, there are standard scales and widely used indicators developed by some organizations. In the study, the natural environment, open green areas, historical texture, transportation, architectural texture, security, services, neighborhood and economy indicators, which are frequently used in many studies, are examined by comparing importance and satisfaction levels. Priority order was used for importance analysis and Likert type questions were used for satisfaction analysis. Kuzeykent Neighborhood, which is one of the neighborhoods, Kastamonu university campus located in and thus the development process has been accelerated, has been selected as a case for this study. The urban environment is reshaped after all the rapid physical developments and changes in socio-status in urban development depending on the population density of the area. In this context, the main aim of this study is to determine the level of quality of life components in the city depending on spatial change. As a result of the research, historical texture, architectural texture and neighborhood were the most important issues, while transportation security and economy are the least important indicators. However, the satisfaction level in the neighborhood indicator was the lowest and no quality of life was found to be high.

Keywords: Quality of Urban Life, Indicators, Urban Development Areas, Kastamonu, Turkey

# **1. INTRODUCTION**

The main objective to be reached when planning cities is meet the vital requirements of individuals living there. The basic requirements have changed with the development of modern life and modernization of societies. The concept of quality of life, which expresses the well-being of individuals and their level of satisfaction with the conditions, has come to the fore as the subject of many disciplines.

The concept of quality of life as a current research subject of urban planning is frequently addressed. Moreover, there is no consensus about the definition of 'quality of life' because of being the subject of independent disciplines such as health, social sciences and having relationship with the individual, social and / or economic indicators. According to the definition of the World Health Organization, it is the way of perception and evaluation of individuals in their life situations in the context of the system of cultural structures and values they belong to (Joshi et al. 2018). In other words, it expresses social, urban infrastructure, communication, transportation, housing and similar economic and spatial elements of the level of presentation above the predetermined extent of the state. It contains social, cultural and political elements and processes. It also includes equal and balanced distribution of possibilities and opportunities offered by the city (Geray, 1998).

Aydıner Boylu and Paçacıoğlu (2016) show the indicators of urban quality of life; sex, age, marital status, education, income, health, housing characteristics, work life and leisure activities. These indicators focus on the qualifications of the individual and include the qualities offered by the space. If

the quality of life and the standards of living are raised in the places where people are located, economic growth will be sustainable and prosperity will be possible. Therefore, it has become a necessity to measure the quality of life of cities (Volkan et al. 2018).

## 2. MATERIAL AND METHODS

The research was carried out in the District of Kuzeykent in the central district of Kastamonu. Kastamonu province, Turkey to the north is located in the Western Black Sea Region (Figure 1). The primary attractive elements of the historical and architectural heritage in the city are the traditional residential architecture. There are 19 districts in the district of Kastamonu.



Figure 1. Location of the case area

Kuzeykent District, which is chosen as the study area, has a total population of 26,157 (TSI, 2018). Due to the presence of Kastamonu University, it has a young population in the 20-24 age group. Kuzeykent District, due to the developments in recent years, Kastamonu is a neighborhood in the city is described as a new development zone.

In the study, the questionnaire method was used depending on the subjective variables shaped according to the individual's own perception. The aim of the questionnaire applications is to measure the quality of life in the traditional texture and new development area of the town of Kuzeykent, Kastamonu. The questionnaire consists of 3 parts. In the first part, the demographic characteristics of the participants, the satisfaction levels in terms of the components of quality of life in the second part, and the importance given to the components of quality of life in the last part were evaluated.

# 3. RESULTS AND DISCUSSION

In this study, the data obtained from the surveys were obtained from the people living in the Kuzeykent neighborhood of Kastamonu province. Demographic characteristics of the participants are given in Table 1. According to this table, 51% of the participants were male and 49% were female. Most of the participants are young and middle aged individuals. If education levels are examined, individuals who are undergraduate have a ratio of 63.3 %. 51% of the individuals living in the area are tenants and 45% are landlords.

## Table 1. Demographic Characteristics

| Qualifications        | Person (n) | Percent (%) |  |
|-----------------------|------------|-------------|--|
| Gender                |            |             |  |
| Men                   | 88         | 51          |  |
| Women                 | 83         | 49          |  |
| Age                   |            |             |  |
| 18-25                 | 92         | 62          |  |
| 26-35                 | 0          | 0           |  |
| 36-45                 | 27         | 18          |  |
| 46-55                 | 20         | 13          |  |
| 55 and above          | 8          | 7           |  |
| Education             |            |             |  |
| Elementary            | 6          | 3.5         |  |
| Secondary             | 7          | 4,1         |  |
| High School           | 40         | 23.5        |  |
| Undergraduate         | 111        | 65.3        |  |
| Master PhD            | 6          | 3.5         |  |
| Property Status       |            |             |  |
| Owner                 | 75         | 45          |  |
| Tenant                | 85         | 51          |  |
| Public housing        | 2          | 1           |  |
| Does not pay any rent | 4          | 3           |  |

The answers obtained from the questionnaires were evaluated using the Likert scale and the answers given in terms of the life satisfaction in the neighborhood are shown in Table 2. Quality of life components were found to be most satisfied with Natural environment (14%) and Historic texture (13%), satisfied with Neighborhood Relationships (42%) and transportation (39%). In addition, it is noteworthy that there is a group of unstable participants in terms of services (37%) and security (35%). In the emergence of this result, it is of great importance that the concept of quality of life is the result of a subjective evaluation of the person.

|      | Qualifications              | Not at al | l satisfied | Dissa | tisfied | Unde | cided | Sati | sfied | Very S | atisfied |  |
|------|-----------------------------|-----------|-------------|-------|---------|------|-------|------|-------|--------|----------|--|
|      |                             |           |             |       |         |      |       |      |       |        |          |  |
|      |                             | f         | %           | f     | %       | f    | %     | f    | %     | f      | %        |  |
|      | Natural environment         | 8         | 5           | 37    | 22      | 39   | 23    | 57   | 33    | 24     | 14       |  |
|      | Light green fields          | 9         | 5           | 41    | 24      | 45   | 26    | 56   | 33    | 14     | 8        |  |
| ife  | Historic texture            | 34        | 20          | 36    | 21      | 27   | 16    | 45   | 26    | 23     | 13       |  |
| ofl  | Easy access                 | 14        | 8           | 25    | 15      | 41   | 24    | 67   | 39    | 17     | 10       |  |
| ţ,   | Architecture                | 17        | 10          | 38    | 22      | 51   | 30    | 48   | 28    | 11     | 6        |  |
| illi | Security                    | 18        | 11          | 28    | 16      | 60   | 35    | 48   | 28    | 11     | 6        |  |
| õ    | Services                    | 21        | 12          | 32    | 19      | 64   | 37    | 38   | 22    | 10     | 6        |  |
|      | Neighbourhood Relationships | 12        | 7           | 21    | 12      | 38   | 22    | 72   | 42    | 22     | 13       |  |
|      | Economic Causes             | 18        | 11          | 32    | 19      | 56   | 33    | 47   | 27    | 12     | 7        |  |

In this section, the importance and satisfaction values of the residents were calculated. These calculations are given in Table 3. It has the highest value from its values. The neighborhood has the highest value in importance values. In parallel with this, the satisfaction value of the neighborhood has also the highest value. While the value with the lowest value is the economy and security, it ranks higher in terms of satisfaction. But satisfaction of architectural texture, historical texture and transportation issues are lower although they give more importance.

|      | Qualifications        | Significance Score | Satisfaction Value | Meaning      |
|------|-----------------------|--------------------|--------------------|--------------|
|      | Neighborliness        | 4,44               | 3,4                | Undecided    |
|      | Architectural texture | 3,64               | 3,0                | Undecided    |
| ife  | Historic texture      | 3,33               | 2,9                | Undecided    |
| ofl  | Natural environment   | 2,96               | 3,3                | Undecided    |
| ţ    | Services              | 2,86               | 2,9                | Undecided    |
| iali | Open- green fields    | 2,76               | 3,2                | Undecided    |
| ð    | Transportation        | 2,58               | 2,3                | Dissatisfied |
|      | Security              | 1,82               | 3,0                | Undecided    |
|      | Economy               | 1,82               | 3,0                | Undecided    |

#### 4. CONCLUSION

As a result of changing the requirements in the process, studies involving the user gain importance (Ozturk vd., 2013). In this study, the importance and satisfaction levels of the inhabitants of Kuzeykent neighborhood of Kastamonu province are determined. In order to ensure a high quality of life, the urban areas should be planned and renewed within the framework of the sustainability principle. In this context, in urban areas, the economy should be developed, the environment should be clean and well-kept, there should be a convenient transportation system, social services such as health, education, open green areas should be comprehensive and adequate level. The preservation of historical and cultural values and the strength of social relations is another important issue.

According to the average responses of people living in Kuzeykent neighborhood, the satisfaction level in terms of value is not satisfied. It is observed that there is no satisfaction about transportation and other average values are unstable. This situation can be interpreted that people are not satisfied with the qualities specified for the quality of life. Today, one of the most important goals that societies aim to reach is a high quality of life. Extensive studies at the national level should be made by increasing awareness of the measurement of quality of life.

## REFERENCES

- Aydıner Boylu, A., and Paçacıoğlu, B. (2016). Quality of Life and its Indicators, Journal of Academic Research and Studies, 8 (15), 137-150.
- Geray, C. (1998). Urban Quality of Life and Municipalities, Turkish Administrative Journal (421), 327.
- Joshi M.R., Chalise H.N. and Khatiwada P.P. (2018). Quality of Life of Nepalese Elderly Living in Rural Nepal, Journal of Gerontology & Geriatric Research (7),484, DOI: 10.4172/2167-7182.1000484.
- Shubbar F., Raffaello F. (2018). Sustainable Neighborhoods in the State of Qatar: Msheireb Downtown Doha. Saudi Journal of Engineering and Technology, DOI: 10.21276/sjeat.2018.3.7.3.
- Ozturk S., Bozdoğan E., Gülgün B., Enez K., 2013. Evaluation of Traditional Housing Areas For Life Quality Indicators: An Investigation on The Case of Kastamonu City, Conference Paper, 5th Congress of Landscape Architecture, Adana.
- Volkan İ.S. and Ahmet K. (2018). Analysis of Urban Quality of Life Indicators in Turkey, SAI Journal (108).

WHO, 2019. https://www.who.int/healthinfo/survey/whoqol-qualityoflife/en/



# Spatial Accessibility to Primary Health Care Centers at Neighborhood Scale in Kastamonu, Turkey

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Abstract: The concept of accessibility expresses physical and economic availability of a service for every citizen fairly. Accessibility of health facilities is one of the standards that must be provided as a part of the right to health. Primary health centers, which is a kind of health cares offered in different scales and scopes, are the most important and basic units in order to protect the population health. In the study conducted within the scope of health services, accessibility of health centers for neighborhoods in Kastamonu Central District was tasted generating two scenarios as driving and walking to reach health centers. For analysis, Euclidean distance to family health centers was calculated by using ArcMap package program. Thus, it is aimed to identify the neighborhoods where the service is inadequate. According to the results of the research, only 7 neighborhoods have a health center although there are a total of 18 neighborhoods in the urban area. These facilities are usually found in developing neighborhoods of the city. Traditional texture was found to be insufficient in terms of the presence of the health center. However, according to distance analysis period from the most remote residential area to the facility in the neighborhood was found 1 minute according to the scenario 1, 3,5 minutes according to the scenario 2 in the neighborhood of Hepkebirler located in the city center. However, it was calculated as 2.7 minutes according to scenario 1 and 22 minutes according to scenario 2 in Kuzeykent neighborhood, which is one of development areas. While the principle of existence is provided in the development areas, accessibility of these neighborhoods is determined to be lower than the traditional texture. As a result, it is thought that the consideration of the study in the reconstruction studies to be done in the traditional texture will increase the level of accessibility of health services.

Keywords: Spatial accessibility, health centers, geographic information systems, Kastamonu

# **1. INTRODUCTION**

Livable cities are habitats where all individuals and social groups come together easily, where services can be accessed and integrated. Access to urban services is an important criterion in increasing living standards of cities. These services can be classified as health, education, socio-cultural facilities and active and passive green areas.

Access to health care plays an important role in improving health equality and quality of life. Health-related facilities should therefore be universally accessible, acceptable, appropriate and of good quality (Campbell, 2013; Luis and Cabral, 2016). In particular, primary health care services are perceived as the backbone of a national health care system (Starfield, 1994; Starfield et al., 2005; Wang et al., 2018).

Patients tend to use more if they are using health facilities more often than they are in their vicinity (Mizen et al., 2015; Luis and Cabral, 2016). Therefore, the relationship between housing units and health units is gaining importance. The spatial location of health units is vital. Health areas have always been very important in city planning. Gravity models are frequently used in urban service site selection decisions. It has been repeatedly mentioned that these models are more adapted to the planning and geography studies than other mathematical models (Lee, 1973) Cubukcu and Taha (2016) discussed Network Distance relationship with Euclidean Distance.

At the end of the research, they conclude that this is a boundary between the Euclidean and the city. However, the traditional and most common techniques used to calculate accessibility in public health research are still Euclidean and network distance (Mizen et al., 2015; Luis and Cabral, 2016). Access to health care depends on the characteristics of the distribution system as well as on the characteristics of the population in need of such services (Aday and Andersen, 1974).

The study area is among the first 10 provinces with the highest immigration (Koç and Saraç, 2018) and the population of the elderly has been increasing since 2016 (TSI, 2018). This increases the importance of primary health care centers.

#### 2. MATERIAL AND METHODS

In the study, the spatial accessibility of health facilities was investigated in the case of Kastamonu city. The material of the study is primary health care centers located in the central district. Health facilities in all neighborhoods have been identified spatially.

The research focuses primarily on the presence of primary health care centers in neighborhoods. Then, Euclidean Distance was calculated considering the maximum distance to the residential areas of the neighborhoods within the neighborhoods. Euclidean distance (ED) between the two points with the coordinates  $(x_1, y_1)$  and  $(x_2, y_2)$  is calculated as (Cubukcu and Taha, 2016):

ED = 
$$\sqrt{(x_1 - x_2)^2 + (y_1 - y_2)^2}$$

This technique can now be applied by geographic information systems. In the study, the distance from all facilities was determined by ArcGIS. 1. Driving according to the scenario, 2. Walking times are calculated according to the scenario. The average driving speed is 50 km / h and the average walking speed is 90 m / min. Considering the distances obtained, the time spent by individuals on accessing primary health care centers was determined.

### 3. RESULTS AND DISCUSSION

The spatial distribution of primary health care centers is given in the study carried out in the neighborhoods of Kastamonu Merkez District. There are a total of 19 neighborhoods in the central district. There are primary health care centers in the neighborhoods such as Esentepe, Aktekke, Candaroğulları, Saraçlar, Mehmet Akif Ersoy, Kuzeykent and İnönü. However, there are no primary health care centers in the Beyçelebi, Isfendiyar, Cebrail, Kırkçeşme, Yavuzselim, Atabeygazi, Hepkebir, Hisarardı, Akmescit, Topçuoğlu, Honsalar and İsmailbey neighborhoods.



Figure 1. Spatial Distrubiton of Primary Health Care Centers

The results obtained as a result of the calculations are given in Table 1. According to Scenerio-1, the driving time is between 3.4 and 0.1 minutes, according to Scenerio-2 between 2.3 and 31.1. When these periods are taken into account, the quality of primary health care centers which are not sufficient for accessibility should be increased and improved.

Table 1. Accessibility times by scenarios

| Neighbourhood     | Scenerio-1    | Scenerio-2       |
|-------------------|---------------|------------------|
| Esentepe          | Driving (min) | Walking<br>(min) |
| Aktekke           | 0,1           | 2,3              |
| Candaroğulları    | 0,8           | 7,8              |
| Saraçlar          | 0,9           | 8                |
| Mehmet Akif Ersoy | 1,7           | 15               |
| Kuzeykent         | 2             | 17               |
| İnönü             | 2,7           | 22               |

In this study, as a result of calculating access times due to transportation of vehicles and pedestrians to primary health care centers areas in neighborhoods, it has been reached that Mehmet Akif Ersoy, Kuzeykent and İnönü neighborhoods are in the last places. Especially in the primary health care centers in İnönü neighborhood, it is one of the most important problems to be solved in 31.1 minutes.

## 4. CONCLUSION

Health problems have not lost their importance since human life. In the provision of health services in Turkey is the fact that there are problems. The right decisions cannot be produced by moving away from holistic approaches in the selection of health facilities. The aim of the study was to determine travel times due to driving and walking. One of the new development areas, Mehmet Akif Ersoy, Kuzeykent and İnönü neighborhoods were found to be the last in terms of time. While the lowest values are obtained in terms of time in Esentepe neighborhood due to the size of the neighborhood, its location can be discussed according to the residential areas. However, the accessibility of a health center location in İnönü neighborhood with an average walking speed of 31.1 minutes should be taken into consideration in relation to the elderly, disabled, child or sick people.

Nowadays, as in all subjects, a planning approach with human-oriented design criteria should be adopted in health areas. The planning of new health areas and existing health areas should be done by taking into consideration the accessibility phenomenon.

Due to the physical or mental disturbance of health areas, some movements, senses or functions are restricted and serve to people who act in order to solve this problem; therefore, mandatory minimum standards are required to ensure accessibility and eliminate negative barriers in these areas. In the planning of the built environment, it is necessary to increase the service quality by adopting a "patient-oriented" approach in the organization and planning of spaces.

There are limitations due to reasons such as inadequacy of personnel and economic costs in increasing the service quality. However, when determining the distance to be produced for primary health care centers, the walking distance, which is taken as a basis in the zoning plans, is limited to 500 meters in the Spatial Plans Building Regulation. In terms of access within 500 meters of the legal direction, there is a problem outside Esentepe District. It is important to minimize spatial barriers and to review planning studies in the accessibility of primary health care centers designed to meet the primary health care demand of the local population.

#### REFERENCES

Aday L. A. Andersen R., (1974). A framework for the study of Access to medical care, Health Serv. Res. 9:208.

- Campbell J, Buchan J, Cometto G, David B, Dussault G, Fogstad H, Fronteira I, Lozano R, Nyonator F, Pablos-Méndez A, Quain EE, Starrs A, Tangcharoensathien V., (2013). Human resources for health and universal health coverage: fostering equity and effective coverage. Bull World Health Organ. 2013;91(11):853–863. doi: 10.2471/BLT.13.118729.
- Cubukcu K. M., Taha H., (2016). Are Euclidean Distance and Network Distance Related?, 7 th Asia-Pacific International Conference on Environment-Behaviour Studies, St Leonard Hall, Edinburgh University, United Kingdom, 27-30 July 2016.

- Koc I. and Sarac M., (2018). The change of the size and direction of Elderly Migration in Turkey, Journal of Aging Studies, 2, pp. 19-37.
- Luis A. A., and Cabral P., (2016). Geographic accessibility to primary healthcare centers in Mozambique, Int J Equity Health, 15: 173, doi: 10.1186/s129 39-016-0455-0.
- Mizen A., Fry R., Grinnell D, Rodgers S.E. (2015). Quantifying the Error Associated with Alternative GIS-based Techniques to Measure Access to Health Care Services. AIMS Public Heal, 2(4):746–761. doi: 10.3934/publichealth.2015.4.746.
- Nischith K.R., Bhargava M., Akshaya K.M. (2018). Physical accessibility audit of primary health centers for people with disabilities: An on-site assessment from Dakshina Kannada district in Southern India. J Family Med Prim Care;7:1300-3.

Starfield B., (1994). Is primary care essential?, The Lanced, vol. 344, October 1994, pp. 1129-1133.

- Starfield B., Shi L., Macinko J., (2005). Contribution of Primary Care to Health Systems and Health, The Milbank Quarterly, https://doi.org/10.1111/j.1468-0009.2005.00409.x.
- Wang X., Yang H., Duan Z., Pan J., (2018). Spatial accessibility of primary health care in China: A case study in Sichuan Province,

TSI (2018). Population Projections: 2018-2080. Newsletter, Issue: 30567, Ankara from http://www.tuik.gov.tr/PreHaberBultenleri.



# Awareness of the Intended Uses of Plants in Istanbul

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**Abstract:** There are many different uses of plants in landscape. However, a lot of of these intended uses are not known by many. With the help of this study, it is aimed to reveal the knowledge levels of individuals with different educational level about the intended uses of plants in the landscape and the relationship between education level and plant utilization aim awareness is determined. In order to determine the level of knowledge of people at different levels of education, a survey of over 500 people is carried out at the meeting on 100 primary school students, 100 secondary school students, 100 high school students, 100 university students and 100 postgraduates in İstanbul. In the survey, 12 different uses of plants are given to the participants and they are asked to mark what they know from these usage areas. In order to determine the effect of the level of education on the results, chi-square analysis is conducted. Results revealed that the level of education was effective on awareness of nine of the intended uses of plants in the landscape.

Key words: the purpose of the use of plants, awareness, landscape and plant

## **1. INTRODUCTION**

Open-green areas are very important areas in terms of physical, aesthetic, psychological, social, recreational, technical, economic and urban health. Urban open green areas, plenty of sun and fresh air allow to move freely, thus creating a healthier, more balanced, regenerative and beneficial environment for people. It also plays a calming, relaxing role on people (Smardon, 1990). Open-green areas prevent visual pollution in the city, and allow people to live, to relax in a beautiful landscape and to have sincere love for all living and non-living beings (Hillman, 1994). The open-green areas offer a landscape and nature source to the urban people. These areas affect people's spiritual selves, increase their allegiance to space, and help renew their lost energy.

Today, planting in urban and rural areas is very important in terms of aesthetics and functional aspects. The right choice and right use of plant material in urban outdoor arrangements causes significant effects for bringing people closer to nature. These contributions can be categorized under four headings by using Jernberg (2001), Relf (1992) and Kaplan (1992). Plants (i) away from a stressful urban environment, (ii) stimulate the mental and physical senses of people for research and discovery of new things, and revive history (ii) increase interest and concentration by addressing senses, (iv) helping to meet individual needs

Plants are a part of our life and remind us that we live. Plants constitute the living structure of the garden (Tyson, 1998). Plants also play an important role in self-respect (Mattson, 1992; Lewis, 1994; Hewson, 1994; Rice and Remy, 1994). Plants, attract people and animals with their flowers, leaves, stems, fruits and fragrances. Plants undertake the tasks such as closeness, enclosing, creating personal area, softening, living, filtering the sun, cleaning the air, providing an environment for birds and bringing naturalness to human life (Tyson, 1998). Value is added to the space with the help of planting design in open green areas, monotony is prevented, to socialize. It can be allowed recreational activities and socialization of people. Urban open-green areas play a calming, relaxing role on the nerves of people (Smardon, 1990). Landscape architects should realize the planting design by knowing the properties of plants and their contributions to the field, and should make decisions about the location of plants and plant species from the beginning of the design while performing space arrangements (Sakıcı, 2014).

Plants used in urban environment contribute to the urban ecosystem in a functional sence, such as energy saving, moisture formation (Beckett et al., 2000; Akbari et al., 2001), noise reduction (Çepel, 1988; Walker, 1991), wind, dust and greenhouse effects reduction (Novak et al., 2000; Akbari et al., 2001; Novak and Crane, 2002), preventing light reflections (Heisler, 1986; Walker, 1991; Heisler and Grant, 2000). In addition, due to seasonal conditions such as early flowering,

late coloration and long-term flowering, the plants remind people that time has passed by providing sensory stimulation (Sakıcı, 2009). In addition to some plant's charecteristic features in plant selection such as the fragrance, softness / hardness, edible fruit, seeds, color changes, shadowing, visual boundary, wind screen, wildlife habitat values should be taken into account (Sakıcı, 2014).

Although the plants have such important functions, it is aimed to determine the level of knowledge of the people in different education level about the purposes of using the plants in the landscape and to determine the level of relationship between the level of education and plant awareness.

## 2. MATERIALS AND METHODS

In order to reveal the awareness of the uses of plants, a questionnaire was conducted in Istanbul in 500 people with different education levels, primary school (100 people), secondary school (100 people), high school (100 people), university students (100 people) and university graduates (100 people). In the survey study, the subjects were given 12 different intended use of plants and asked to mark what they know. The most known and least known plant use objectives were determined as percentage and Chi-square analysis was performed to determine the effect of education on the results. The 12 different purpose of plant use in survey is as follows,

- Clean the air,
- To enrich the soil,
- Create a beautiful fragrance,
- Providing benefit with fruit and leaves,
- Providing aesthetic beauty and visual richness with characteristic features,
- Providing slope and erosion control,
- Prevent global warming,
- Providing climatic control,
- Space identification,
- Providing wind control,
- Functional objectives such as routing, separating, focusing,
- Providing noise control

## 3. RESULTS AND DISCUSSION

## **Demographic Characteristics of Survey Participants**

The questionnaires were conducted in Istanbul and 100 participants from each educational level were preferred. A total of 500 people were surveyed in primary, secondary, high school, university and university graduates. In total, 299 people were female and 201 people were male. Gender distribution by educational level is shown in Table 1.

Table 1. Gender distribution according to the educational level of the participants

| Education | n level | Primary<br>School | Secondary<br>School | High | University | Graduate<br>University | All |
|-----------|---------|-------------------|---------------------|------|------------|------------------------|-----|
|           | Woman   | 46                | 44                  | 88   | 59         | 62                     | 299 |
| Gender    | Man     | 54                | 56                  | 12   | 41         | 38                     | 201 |
|           | Total   | 100               | 100                 | 100  | 100        | 100                    | 500 |

## Knowledge Levels of Participants About Purposes of Plant Use

In order to determine the level of knowledge of the participants about the intended use of the plants, 12 different purpose of plant use were given in survey and it was asked to mark what they knew from these usage. The results are shown in Table 2. In order to determine the effect of education level on the results, Chi-square analysis was performed and according to the results;

There were significant differences between the groups in terms of the level of knowledge about the ability of plants releted to 'providing aesthetic beauty and visual richness with characteristic features' (p < 0.05) and while 66-70% of the elementary school, secondary school, high school and university students know this feature, 86% of the University graduates know that.

A significant difference was not found between the groups in terms of the level of knowledge about the ability of plants releted to 'providing benefit with fruit and leaves' (p> 0.05) and this feature of the plants is known by 75-85% of the all groups.

A significant difference was found between the groups in terms of the level of knowledge about the ability of plants releted to 'providing clean the air' (p < 0.05) and, while 80% of university students and 88-90% of primary and high school students knew this feature, 95% of secondary students and 97% for University graduates know this feature of the plants.

A significant difference was not found between the groups in terms of the level of knowledge about the ability of plants releted to 'providing enrich the soil' (p > 0,05). 80-89% of all different groups know this feature of the plants.

Significant differences were found between the groups in terms of the level of knowledge about the ability of plants releted to 'prevent global warming' (p < 0.05), while 46-51% of primary, secondary and high school students know this feature, 64% of university students and 73% of university graduates know.

A significant difference was found between the groups in terms of the level of knowledge about the ability of plants releted to 'providing slope and erosion control' (p < 0.05). While this feature is known by 43-53% of primary, secondary and high school students, 61% of university students and 75% of university graduates know this feature.

A significant difference was found between the groups in terms of the level of knowledge about the ability of plants raleted to 'Space identification'. (P <0,05), While 43-52% of elementary, middle school, high school and university students know this feature, 71% of university graduates were aware of this feature.

There was significant difference between the groups in terms of the level of knowledge about the ability of plants raleted to 'functional objectives such as routing, separating, focusing' (p < 0,05). While only 24-36% of primary school, middle school, high school and university students know this feature of plants, 65% of university graduates know this feature.

A significant difference was not found between the groups in terms of the level of knowledge about the ability of plants raleted to 'create a beautiful fragrance' (p > 0.05), and 75-88% of all different groups know this feature of the plants.

A significant difference was found between the groups in terms of the level of knowledge about the ability of plants raleted to 'providing wind control' (p < 0.05) and, While 39-50% of primary school, secondary school, high school and university students knew this feature of plants, 69% of university graduates appeared to be known.

There was a significant difference between the groups in terms of the level of knowledge about the ability of plants raleted to 'providing noise control' (p < 0.05). While primary school, middle school and high school students knew this feature by 13-19%, this rate has reached 31% for university students and 50% for university graduates.

There was significant difference between the groups in terms of the level of knowledge about the ability of plants releted to 'providing climatic control' (p < 0.05), While only 33% of elementary school students and 48-49% of secondary and high school students were aware of this feature, %61 of University students and 74% for University graduates were aware of this feature

|                               | Information |         |           | GROUI  | 2          |            |       |     |
|-------------------------------|-------------|---------|-----------|--------|------------|------------|-------|-----|
| Purpose of plant use          | Status      | Primary | Secondary | High   |            | University | p     |     |
|                               | Status      | School  | School 1  | School | University | Graduates  |       | Σ   |
| Clean the air                 | Unfamiliar  | 10      | 5         | 12     | 20         | 3          | 0.001 | 50  |
|                               | Familiar    | 90      | 95        | 88     | 80         | 97         | 0,001 | 450 |
| To enrich the soil            | Unfamiliar  | 11      | 15        | 18     | 18         | 20         | 0.465 | 82  |
|                               | Familiar    | 89      | 85        | 82     | 82         | 80         | 0,403 | 418 |
| Create beautiful              | Unfamiliar  | 25      | 13        | 21     | 21         | 12         | 0.076 | 92  |
| fragrance                     | Familiar    | 75      | 87        | 79     | 79         | 88         | 0,070 | 408 |
| Providing benefit             | Unfamiliar  | 25      | 23        | 19     | 25         | 15         | 0.246 | 107 |
| with fruit and leaves         | Familiar    | 75      | 77        | 81     | 75         | 85         | 0,540 | 393 |
| Aesthetic beauty and          | Unfamiliar  | 33      | 34        | 34     | 30         | 14         | 0.007 | 145 |
| visual richness               | Familiar    | 67      | 66        | 66     | 70         | 86         | 0,007 | 355 |
| Prevent global                | Unfamiliar  | 49      | 54        | 53     | 36         | 27         | 0,000 | 219 |
| warming                       | Familiar    | 51      | 46        | 47     | 64         | 73         |       | 281 |
| Slope and erosion             | Unfamiliar  | 47      | 52        | 57     | 39         | 25         | 0.000 | 220 |
| control                       | Familiar    | 53      | 48        | 43     | 61         | 75         | 0,000 | 280 |
| Climatic control              | Unfamiliar  | 67      | 52        | 51     | 39         | 26         | 0.000 | 235 |
|                               | Familiar    | 33      | 48        | 49     | 61         | 74         | 0,000 | 265 |
| Space identification          | Unfamiliar  | 56      | 54        | 57     | 48         | 29         | 0.000 | 244 |
|                               | Familiar    | 44      | 46        | 43     | 52         | 71         | 0,000 | 256 |
| Wind control                  | Unfamiliar  | 50      | 56        | 61     | 58         | 31         | 0.000 | 256 |
|                               | Familiar    | 50      | 44        | 39     | 42         | 69         | 0,000 | 244 |
| Functional objectives         | Unfamiliar  | 71      | 71        | 76     | 64         | 35         | 0.000 | 317 |
|                               | Familiar    | 29      | 29        | 24     | 36         | 65         | 0,000 | 183 |
| Noise control                 | Unfamiliar  | 86      | 81        | 87     | 69         | 50         | 0.000 | 373 |
|                               | Familiar    | 14      | 19        | 13     | 31         | 50         | 0,000 | 127 |
| The average of 12             | Unfamiliar  | 44,17   | 42,5      | 45,5   | 38,92      | 23,92      |       |     |
| different features $(\Sigma)$ | Familiar    | 55,83   | 57,5      | 54,5   | 61,08      | 76,08      |       |     |

Table 2. Differences in knowledge levels of participants about the purpose of plant use

## 4. CONCLUSION

The plants are used in landscaping for many purposes. It appeared that some of these purposes such as cleaning the air, enriching the soil, creating a beautiful fragrance, providing benefits with fruits and leaves, providing aesthetic beauty and visual richness with characteristic features and providing slope and erosion are known by many people, however, other purposes such as providing climatic control, space identification, providing wind control, functional objectives such as routing, separating, focusing, and providing noise control are less well known with this study.

In addition, This study was determined that there is a significant difference between the levels of education and the level of knowledge of the participants about all intended use of the plants except for, three features such as enriching the soil, creating a beautiful fragrance and providing benefit with fruit and leaves. Generally, It was revealed that the level of knowledge of these purposes also increased as the level of education increased. But high school students have the lowest level of knowledge about knowledge level of all purposes of plant use. The knowledge level of the groups from the lowest to the highest, respectively; High school students (%54,5), Primary school students (%55,83), Secondary school students (%57,5), University students (%61,08) and University graduates (%76,08).

#### REFERENCES

- Akbari H, Pomerantz M & Taha H (2001). Cool surfaces and shape trees to reduce energy use and improve air quality in urban areas. Solar Energy,70(3): 295-310.
- Beckett K P, Freer-Smith P H & Taylor G (2000). Particulate pollution capture by urban trees: effects of species and windspeed. Global Change Biology, 6: 995-1003.

Çepel N (1988). Peyzaj ekolojisi. İstanbul Üniversitesi Orman Fakültesi, Yayın No: 3510, İstanbul.

Heisler G M (1986). Effects of individual trees on the solar radiation climate of small buildings. Urban Ecology 9: 337-359.

Heisler G M & Grant R H (2000). Ultraviolet radiation in urban ecosystems with consideration of effects on human healthy, Urban Ecosystems, 4(3): 193-229.

Hewson M (1994). Horticulture as Therapy, A Practical Guide to Using Horticulture as A Therapeutic Tool, M.L. Hewson, 153 s.

- Hillman J (1994). The role of urban parks, the future of our urban parks, Finding of The Symposium Church House Conference Centre, London, 6-7.
- Jernberg A (2001). Nature and garden within care and caring, The Swedish University of Agricultural Science.
- Kaplan S (1992). The restorative environment: Nature and human experience, The role of horticulture in human well-being and social development, 134–142.
- Lewis C A (1994). The evolutionary importance of people-plant relationships. Journal of home & consumer horticulture, 1(2-3): 239-254.
- Mattson R H (1992). Prescribing health benefits through horticultural activities. In The Role of Horticulture in Human Well-Being and Social Development: ANational Symposium, Timber Press, Porland, Oregon, 161-168.
- Novak D J, Civerolo K L, Rao S T, Sistla G, Luley C J & Crane D E (2000). A modelingstudy of the impact of urban trees on ozone. Atmospheric Environment, 34(10): 1601-1613.
- Novak D J & Crane D E (2002). Carbon storage and sequestration by urban trees in the USA. Environmental Pollution, 116(3): 381-389.
- Relf D (1992). Human issues in horticulture. Horticulture Technology, 2(2):159-171.
- Rice J S & Remy L L (1994). Evaluating horticultural therapy: The ecological context of urban jail inmates. Journal oj home & consumer horticulture, 1(2-3): 203-224.
- Sakıcı Ç (2009). Ruh ve sinir hastalıkları hastanelerinde açık alan terapi ünitelerinin peyzaj tasarımı: Ataköy (Trabzon) Ruh Sağlığı ve Hastalıkları Hastanesi örneği. Doktora Tezi, K.T.Ü. Fen Bilimleri Enstitüsü, Trabzon.
- Sakıcı Ç (2014). Bitkilerin Psikolojik Katkıları. Gıda Tarım ve Hayvancılık Bakanlığı Dergisi, Türktarım, 216 (Mart-Nisan): 38-43. (ISSN: 1303-2364).
- Smardon R C (1990). Perception and aesthetics of the urban environment: review of the role of vegetation. Landscape and Urban Planning, 15 (1-2): 85-106.

Tyson M M (1998). The healing landscape: Therapeutic outdoor environments. McGrawHill, New York, 224.

Walker T D (1991). Planting design. John Wiley & Sons, New York.



# Plant-Space Relationship: An Example of Mosque Courtyard

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Abstract: The most important of the spatial elements a and components that are thought to be parallel to the meaning of the place and contribute to the understanding of the place are plants. Although it is difficult to reveal the perception of spatial - plant interactions in humans, it can be achieved through experimental studies. In this study, visual impressions of the users in evaluating the perception of plants with spaces were determined by experimental study and the survey technique was used. The study was conducted in Istanbul over 500 people, including 100 primary school students, 100 secondary school students, 100 high school students, 100 university students and 100 university graduates. In this study, 28 plants, which are frequently seen and falmiliar with outdoor areas, were used. In this study, it was aimed to reveal the opinions about which of these plants were identified with the mosque courtyard and which characteristic of the plants were emphasized. Gender and educational level differences were investigated and results revealed that gender and educational level were effective on preferences. According to the results, flower bushes were preferred primarily for the mosque courtyard and rose, pine, tulip, violet and buxus plants were preferred in the first row respectively.

Keywords: Plant-space, Mosque courtyard, Plant's meaning



# A New Biotechnological Application in Aquaculture: Germ Cell Transplantation

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Abstract: Aquatic product is an important source of protein as well as a food source. With the global population expected to exceed nine billion in 2050, demand is likely to increase. Many fish are already on the brink of extinction due to overfishing; consequently, aquafarming has become important to the challenge of securing food resources. Numerous studies aim to develop new aquaculture technology, such as surrogate broodstock technology to remedy these situations. Primordial germ cell (PGC) isolate from the fish species that are difficult to obtain gametes and after PGC inject with microinjection technique to other fish, namely surrogate, which aquaculture conditions are well known can be defined as germ cell transplantation. Although the use of germ cell transplantation has been relatively well established in mammals, the technique has only been adapted for use in fish after entering the 2000s. During the last decade, several different approaches have been developed for germ cell transplantation in fish using recipients of various ages and life stages, such as blastula-stage embryos, newly hatched larvae and sexually mature specimens. As germ cells can develop into live organisms through maturation and fertilization processes, germ cell transplantation in fish has opened up new avenues of research in reproductive biotechnology and aquaculture.

Keywords: Biotechnology, fish, germ cell, aquaculture, surrogate

## **1. INTRODUCTION**

Freshwater or marine aquatic ecosystems are diverse systems that are inhabited by more than 33.000 fish species. However, some fish species have become endangered or extinct (http://www.iucnredlist.org) recently because of unknown or known factors such as rapid global climate change, anthropogenic pressure on water bodies, habitat loss and aquatic pollution. In order to preserve endangered species, there are two main approaches to be adopted. The first is in situ preservation, in which the species needs to be preserved within the ecosystem [1]. Moreover, the second is ex situ preservation of the germlines and allowing reproduction of the individuals of the species artificially in a suitable environment [2, 3]. Although, both method seems convenient in order to conserve mentioned fish species, habitat conservation should be prioritized because it will ensure to protect whole ecosystems are affected by natural events and anthropogenic activities constantly. After all, both methods should be considered to function together in, order to manage fishery resources [4].

Surrogate broodstock development of endangered or commercially important fish species through germ cell transplantation seems one of the most promising approach of ex situ conservation [5-7]. Feasibility of the mentioned technique successfully demonstrated in mice by microinjection of spermatogonia derived from donor males to sterilized male recipients, resulting in formation of donor-derived spermatogenesis [8]. Later on, this method was used in treatment of infertility, reproductive medicine and basic researches extensively. In fish, application of the method was achieved in rainbow trout, where primordial germ cells (PGC) that carry green fluorescent protein (GFP) were transplanted into peritoneal cavity of hatchlings, leading to production of sperm, which has the genetic characteristics of the donor [9]. Xenogeneic transplantation between masu salmon (*Onchorhynchus masou*) and rainbow trout (*Onchorhynchus mykiss*) was performed successfully using the same application method as well [10]. In addition, it was demonstrated that cryopreservation of PGCs for extensive duration before transplantation is possible without any alteration regarding spermatogenesis in the testis of the recipient [11].

Germ cell transplantation in fish may be performed in 3 different development stages: adult, hatchling and embryonic stage. In this review, we aimed to inform the reader about current germ cell transplantation methods and purpose of those applications in fisheries.

## 2. GERM CELL TRANSPLANTATION AT VARIOUS DEVELOPMENTAL STAGES

Embryos (blastula stage): PGCs harvested from the donor hatchling are transplanted into recipient embryo by micromanipulator (blastula stage) that have had endogenous PGC development blocked by the injection of a dead end antisense morpholino oligonucleotide. The method was successfully applied into zebrafish [12] and pearl danio [13] since this method was proposed. The major advantage of this method is that immunorejection of transplanted PGCs was not observed because the transplantation was performed before the sex determination stage in recipient fish [13]. This technique is considered one of the viable approach as it allows generating millions of gametes of fish species, with desired traits such as growth, disease resistance, fecundity etc. by using only one donor PGC [14].

Hatchling stage: In this cage (In this stage), the donor germ cells are transplanted into the peritoneal cavities of newly hatched larvae using a micro injector [15]. Ever since they lack a functional immune system indicated by the absence of differentiation in both their thymus and T cells, the newly hatched larvae are selected as recipients [16]. Immunorejection of exogenous (donor-derived) germ cells do not occur due to lack of a functional immune system. The PGC derived from transgenic rainbow trout were transplanted into the body cavity of masu salmon hatchlings [14]. These hatchlings upon reaching adulthood, produce the xenogenic donor derived offspring [5]. Later, the approach was also successfully applied to trout model where spermatogonial stem cell from vasa-Gfp rainbow trout were transplanted into sterile triploid masu salmon hatchling that resulted in establishment of gametogenesis in male and female recipients [10]. However, in this approach the only disadvantage is that the donor-derived gametes could be generated approximately 2-3 years after the transplantation, depending on the attainment of gonadal maturity of the animal.

Adult stage: This method of germ cell transplantation in teleost fish was proposed by Lacerda and her colleagues [7], using Nile tilapia fish (Oreochromis niloticus) model, in which spermatogonia were transplanted through the urogenital papilla of adult fish [17]. The most important issue in PGC transplantation that needs consideration is the absence of genetic material of the surrogate individual. Therefore, in this method, recipient adult fish was made partially sterile by treatments of cytotoxic drug, busulfan (1,4-butanediol dimethanesulfonate), in association with elevated water temperature. An enriched type A spermatogonial cell suspension harvested from donor testes and labelled with the fluorescent lipophilic dye such as PKH-26 was injected into the adult testes of the recipient through the common spermatic duct. This method confirmed the possibility of using sexually mature adult fish as recipients in germ cell transplantation [7]. Making refinement to this approach, Majhi and his colleagues developed an intra-gonadal surgical method for xenogeneic germ cell transplantation in sexually competent adult fish. They used germ cells from juvenile pejerrey (Odontesthes bonariensis) as donor, while sexually competent adult Patagonian pejerrey (Odontesthes hatchery) as recipients for germ cell transplantation [6]. The endogenous spermatogenesis of recipient fish was depleted using busulfan and high temperature treatments [18]. Donor germ cells isolated by discontinued percoll gradient were labelled with CFDA-SE (green fluorescent dye) and introduced into the recipient gonad. Injection of the germ cell suspension was made possible in this procedure by a long midline cut made in the abdomen of the recipient fish followed by the exposition of the gonads from the coelomic cavity. The transplanted spermatogonial cells were able to colonize the gonads of recipient Patagonian pejerrey and generate donor-derived sperm within 6 months post transplantation [6]. Later, the same group have simplified the transplantation approach by developing a non-surgical method and produced the functional surrogate eggs and sperm capable of fertilization in very short time [18]. The major advantage of using adult as the recipient for germ cell transplantation is that, it considerably reduces the time needed to obtain viable gametes and offspring of donor genotype.

# 3. ADVANTAGES OF SURROGATE PRODUCTION AND APPLICATION AREAS

Some fish species are critically endangered due to overfishing for caviar or high economic value and interference in their natural habitats [19]. For example; some sturgeon species have life spans of over 100 years and sexual maturity is attained between 20 to 25 years [20]. Sterlet (*Acipenser ruthenus*) has fastest reproductive cycle; thus, this species can be used for surrogate production in sturgeons [24]. Other usage area of surrogate production is reduction of the space for keeping fish in aquaculture. Intra-species transplantation is expected to be a potentially effective strategy for these. This latter technology is expected to be able to produce bluefin tuna from a small mackerel species such as the chub mackerel. Adult

bluefin tuna are quite large and require 3–5 years to reach sexual maturity. In contrast, adult chub mackerel, which also belongs to the Scombridae, weigh 300 g and reach maturity in 1 year [21]. Some bluefin species were used as donors in some researches [22, 23]. The conservation of endangered fish is an urgent issue. Although cryo-banking of fish gametes might ultimately help conserve endangered fish, cryopreservation of fish eggs or embryos is still not possible due to their large size and high yolk content [26]. Technology for preservation of genetic resources in liquid nitrogen with sperm is well developed, however, maternal genes and mitochondria cannot be stored. PGC, spermatogonia and oogonia might be materials cryopreservation. Thus, this application might represent a magic solution for preserving the valuable genetic resources of endangered fish species [25]. Moreover, surrogate production is used for prevention against fish diseases by taking advantage of the resistant characteristics of the recipient, prevention of genetic diversity and controlling total egg and sperm production between species that have small and large number of gametes [27].

#### REFERENCES

1. Margules CR, Pressey RL. Systematic conservation planning. Nature. 2000; 405: 243-253.

2. Holt WV, Pickard AR. Role of reproductive technologies and genetic resource banks in animal conservation. Rev Reprod. 1999; 4: 143-150.

3. Wildt DE, Wemmer C. Sex and wildlife: the role of reproductive science in conservation. Biodiverse Conserv. 1999; 8: 965-976.

4. Pukazhenthi BS, Comizzoli P, Travis A, Wildt DE. Applications of emerging technologies to the study and conservation of threatened and endangered species. Reprod Fertil Dev. 2006; 18: 77–90.

5. Takeuchi Y, Yoshizaki G, Takeuchi T. Surrogate broodstock produces salmonids. Nature. 2004; 430: 629-630.

6. Majhi SK, Hattori RS, Yokota M, Watanabe S, Strüssmann CA. Germ cell transplantation using sexually competent fish: an approach for rapid propagation of endangered and valuable germlines. PLOS ONE. 2009; 4: e6132.

7. Lacerda SMSN, Batlouni SR, Costa GMJ, Segatelli TM, Quirino BR, Queiroz BM, et al. A new and fast technique to generate offspring after germ cells transplantation in adult fish: The Nile tilapia (Oreochromis niloticus) model. PLOS ONE. 2010; 5: e10740.

8. Brinster RL, Averbock MR. Germline transmission of donor haplotype following spermatogonial transplantation. Proc Natl Acad Sci USA. 1994; 91: 11303-11307.

9. Takeuchi Y, Yoshizaki G, Takeuchi T. Generation of live fry from intraperitoneally transplanted primordial germ cells in rainbow trout. Biol Reprod. 2003; 69: 1142–1149.

10. Okutsu T, Shikina S, Kanno M, Takeuchi Y, Yoshizaki G. Production of trout offspring from triploid salmon parents. Science. 2007; 317: 1517.

11. Kobayashi T, Takeuchi Y, Takeuchi T, Yoshizaki G. Generation of viable fish from cryopreserved primordial germ cells. Mol Reprod Dev. 2007; 74: 207–213.

12. Giraldez AJ, Cinalli RM, Glasner ME, Enright AJ, Thomson JM, Scott Baskerville S, et al. MicroRNAs regulate brain morphogenesis in zebra fish. Science. 2005; 308: 833-838.

13. Saito T, Goto-Kazeto R, Arai K, Yamaha E. Xenogenesis in teleost fish through generation of germ-line chimeras by single primordial germcell transplantation. Biol Reprod. 2008; 78: 159–166.

14. Yoshizaki G, Okutsu T, Ichikawa M, Hayashi M, Takeuchi Y. Sexual plasticity of rainbow trout germ cells. Anim Reprod. 2010; 7: 187–196.

15. Yoshizaki G, Takeuchi Y, Sakatani S, Takeuchi T. Germ cell-specific expression of green fluorescent protein in transgenic rainbow trout under control of the rainbow trout vasa-like gene promoter. Int J Dev Biol. 2000; 44: 323–326.

16. Yoshizaki G, Fujinuma K, Iwasaki Y, Okutsu T, Shikina S, Yazawa R, et al. Spermatogonial transplantation in fish: a novel method for the preservation of genetic resources. Comp Biochem Physiol Part D Genom Proteomics. 2011; 6: 55–61.

17. Lacerda SMSN, Batlouni SR, Silva SBG, Homem CSP, Franca LR. Germ cells transplantation in fish: The Nile tilapia model. Animal Reproduction. 2006: 2: 146-159.

18. Majhi SK, Hattori RS, Rahman SM, Strüssmann CA. Surrogate production of eggs and sperm by intrapapillary transplantation of germ cells in cytoablated adult fish. PLOS ONE. 2014; 9: e95294.

19. Bemis, W.E.; Kynard, B. Sturgeon rivers: An introduction to acipenseriform biogeography and life history. *Environ. Biol. Fish.* **1997**, 167–183.

20. Dettlaff, T.A.; Ginsburg, A.S.; Schmalhausen, O.I. Sturgeon Fishes: Developmental Biology and Aquaculture; Springer-Verlag: New York, NY, USA, 1993.

21. Yoshizaki, G. & Yazawa, R. Application of surrogate broodstock technology in aquaculture. Fish Sci (2019) 85: 429. https://doi.org/10.1007/s12562-019-01299-y

22. Yazawa R, Takeuchi Y, Morita T, Ishida M, Yoshizaki G (2013) The Pacific bluefin tuna (*Thunnus orientalis*) dead end gene is suitable as a specific molecular marker of type A spermatogonia. Mol Reprod Dev 80:871–880

23. Bar I, Smith A, Bubner E, Yoshizaki G, Takeuchi Y, Yazawa R, Chen BN, Cummins S, Elizur A (2016) Assessment of yellowtail kingfish (*Seriola lalandi*) as a surrogate host for the production of southern bluefin tuna (*Thunnus maccoyii*) seed via spermatogonial germ cell transplantation. Reprod Fertil Dev 28:2051–2064

24. Pšenička M, Saito T, Linhartová Z, Gazo I (2015) Isolation and transplantation of sturgeon early-stage germ cells. Theriogenology 83:1085–1092

25. Yoshizaki G, Lee S (2018) Production of live fish derived from frozen germ cells via germ cell transplantation. Stem Cell Res 29:103–110

26. Mazur P, Leibo SP, Seidel GE Jr (2008) Cryopreservation of the germplasm of animals used in biological and medical research: importance, impact, status, and future directions. Biol Reprod 78:2–12

27. Saito, Taiju & Goto, Rie & Fujimoto, Takafumi & Kawakami, Yutaka & Arai, Katsutoshi & Yamaha, Etsuro. (2010). Inter-species transplantation and migration of primordial germ cells in cyprinid fish. The International journal of developmental biology. 54. 1481-86. 10.1387/ijdb.103111ts.



# Extraction and Characterization of Acid-soluble Collagen from Skin of Rainbow Trout (Oncorhynchus mykiss)

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Abstract: Rainbow trout skin is a by-product from rainbow trout fillet production. The aim of this study was to characterize collagen extracted from the rainbow trout (*Oncorhynchus mykiss*) skin as an alternative to mammalianderived collagen. Acid soluble collagen (ASC) was extracted from the skins of rainbow trout using 0.5 M acetic acid. Then, collagen precipitates with NaCl and purification to 0.1M acetic acid solution and distilled water. SDS-PAGE showed that the collagens contained two alpha components ( $\alpha$ 1,  $\alpha$ 2). Based on the data from the SDS-PAGE and the composition of amino acid, it was probable that the obtained collagen classified as type I collagen. Also, FTIR analysis revealed the presence of helical arrangements of collagen. The functional group of amide A, I, II and III bands have been detected. Moreover, UV spectrophotometry exhibited this collagen have an absorbance at 220 nm. Differential Scanning Calorimetry (DSC) analysis indicated that the collagen exhibits good thermal stability and denatures at a high temperature in a similar manner to mammalian collagen. This report indicates that rainbow trout skin have potential in supplementing the skin of land vertebrates as a source of collagen.

Keywords: Rainbow trout, skin, collagen, isolation, characterization



# ORAL PRESENTATION

# Trend Analysis for Annual Streamflow of Ilgaz Stream (Turkey)

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**Abstract:** The main aim of this study is to detect the trends of mean annual, seasonally and monthly streamflow of Ilgaz Stream in Turkey. Data for the streamflow were acquired from the streamflow gauging station between the period of 2001 and 2016. In order to determine the change-point of the time series for the streamflow data, change-point analysis was applied. Then, trend analysis was performed to determine the tendency of the streamflow by annual, seasonally and monthly. As a result, change-point analysis put forward that the change year was 2005 for annual streamflow of Ilgaz Stream. The results of trend analyses indicated that annual streamflow presented a decreasing trend during the monitoring period. In addition, decreasing trends were also found for the seasonal streamflow of spring and summer though the streamflow of autumn and winter presented increasing trends. Nonetheless, these trends in the streamflow were found statistically insignificant for all periods covering annual, seasonally and monthly time series. In summary, decreasing trends for the streamflow of Ilgaz Stream were anticipated for annual and seasonally (spring and summer) in the study period. The streamflow of the river could be affected by the increase in temperature and evaporation, the reduction in rainfall, and additional reasons occurred by the climate change. Henceforward, the trends in the streamflow should be determined and monitored to forecast the further availability and amount of rivers and other water resources.

Keywords: Trend analysis, climate change, streamflow, Ilgaz Stream, Turkey

## **1. INTRODUCTION**

River is one of the significant sources of freshwater resources. Rivers provide 70% of total easily accessible water (Hisar et al., 2015). Therefore, the past, present and future of available water resources must be continuously monitored. Developing water resource management strategies and policies is mandatory for ensuring the supply of water while it required. To sustain the sufficient amount of water resources in a river basin or to investigate the streamflow patterns in rivers, trend analysis has crucial importance.

Numerous scientists globally and regionally investigated the trends in rivers (Kahya and Kalaycı, 2004; Özel et al., 2004; Cigizoglu et al., 2005; Topaloğlu, 2006; Saplıoğlu et al., 2014; Sütgibi, 2015; Yenigün and Ülgen, 2016; Ercan and Yüce, 2017; Kişi et al., 2018). Several approaches were experienced for understanding the trends in water resources (Sen, 1968; Helsel and Hirsch, 2002; Şen, 2012) and they were applied to predict the streamflow trends in rivers (Tekkanat and Sarış, 2015; Ejder et al., 2016a, b; Kale et al., 2016a, b; Tekkanat, 2017; Tosunoglu and Kisi, 2017; Kale and Sönmez, 2018a, b; Kale et al., 2018; Kişi et al., 2018; Sönmez and Kale, 2018). Unfortunately, there is no study for the trends of streamflow of Ilgaz Stream. Therefore, this paper targeted to carry out a trend analysis for the streamflow of Ilgaz Stream. This paper is the first study on the investigation of trends in the streamflow of Ilgaz Stream.

# 2. MATERIALS AND METHODS

# Study Area

Ilgaz Stream has 21.92 km length (Anonymous, 2013a) and flows through İhsangazi and Araç counties of Kastamonu province (Anonymous, 2013b). Obruk, Sarıpınar, Kızıleller, and Arazya Creeks form branches of Ilgaz Stream (Tot et al., 2017). There is one hydroelectric power plant with 1 MW installed capacity on Ilgaz Stream. Climatic conditions of the region show continental climate characteristics with frost and cold autumn and spring seasons, snowy winters, and warm summers (Kale and Sönmez, 2018b).

Data for the streamflow were collected from the station (D13A061) of The General Directorate of State Hydraulic Works (DSI) (Figure 1). Statistical analyses were applied to mean monthly data and trends were detected for monthly, seasonally, and annual period.



Figure 1. The location of Ilgaz Stream and streamflow gauging station

#### **Change-Point Analysis**

Pettitt (1979) suggested a technique to identify the change point of a time series data. Numerous scientists used this method to detect the change time of the climatic/hydrologic time series. Change point analysis was applied to the streamflow data by using R statistical software (R Core Team, 2019). Pettitt's change point test is given in Equation 1:

$$U_{t,T} = \sum_{i=1}^{t} \sum_{j=t+1}^{T} sgn(x_i - x_j) \text{ for } t = 2, \dots, T, K_T = max |U_{t,T}|,$$
(1)

### **Trend Analysis**

Trend analysis is a largely implemented method to define the tendency in a time series. Box and Jenkins (1976) methodology was used to define the trends in hydrologic time series. The calculation is expressed in Equation 2.

$$X_t = c + \Phi_l X_{t-1} + \dots + \Phi_p X_{t-p} + \theta_l e_{t-1} + \theta_q e_{t-q} + e_t$$

$$\tag{2}$$

#### Mann-Kendall and Spearman's Rho Tests

Non-parametric Mann-Kendall and Spearman's rho tests were used to understand the correlation between the parameters. Mann-Kendall test (Mann, 1945; Kendall, 1955) is extensively used to determine the trends in time series. The formula is as follow:

$$S = \sum_{i=1}^{n-1} \sum_{k=i+1}^{n} sgn(x_k - x_i)$$
(3)

Spearman's rho test is extensively used to measure the magnitude of monotonic relationship between two variables (Lehmann, 1975; Sneyers, 1990). The formula is identified in Equation 4:

$$\rho = 1 - \frac{6(\sum d^2)}{n(n^2 - 1)} \tag{4}$$

## 3. RESULTS AND DISCUSSION

#### Results

The descriptive statistics for streamflow data containing standard deviation (SD), maximum, minimum, and mean values, range, coefficient of kurtosis (CK), coefficient of variation (CV), and coefficient of skewness (CS) were tabulated in Table 1.

| Streamflow | Mean  | Standard Deviation | <b>Coefficient of Variation</b> | Coefficient of Skewness | Max   | Min  |
|------------|-------|--------------------|---------------------------------|-------------------------|-------|------|
| Annual     | 13.14 | 5.90               | 0.449                           | 0.016                   | 21.83 | 4.26 |
| Spring     | 29.63 | 13.00              | 0.439                           | 0.345                   | 55.57 | 9.44 |
| Summer     | 9.44  | 8.66               | 0.918                           | 0.298                   | 35.37 | 2.52 |
| Autumn     | 4.27  | 3.48               | 0.815                           | 0.759                   | 13.59 | 1.22 |
| Winter     | 10.85 | 8.49               | 0.783                           | 0.962                   | 26.77 | 1.85 |

Table 1. The descriptive statistics for mean annual and seasonally streamflow data

Change point analysis results were presented in Table 2. Change point was found to be 2005 for mean annual streamflow for this period. Furthermore, trend analysis results noticed a decreasing trend in the mean annual streamflow of Ilgaz Stream during the monitoring period (Figure 2).

Table 2. Change-point analysis results of mean annual, seasonally and monthly streamflow

| Streamflow |           | Change Point |
|------------|-----------|--------------|
| Annual     |           | 2005         |
|            | Spring    | 2005         |
| Seggenally | Summer    | 2010         |
| seasonally | Autumn    | 2014         |
|            | Winter    | 2006         |
|            | January   | 2011         |
|            | February  | 2005         |
|            | March     | 2013         |
|            | April     | 2005         |
|            | May       | 2009         |
| Monthly    | June      | 2005         |
| Moniniy    | July      | 2006         |
|            | August    | 2008         |
|            | September | 2012         |
|            | October   | 2009         |
|            | November  | 2011         |
|            | December  | 2007         |



Figure 2. Trend analysis result for mean annual streamflow of Ilgaz Stream

Trend analysis results demonstrated that decreasing trends were determined for mean seasonally streamflow of Ilgaz Stream for spring and summer while upward trends were found for autumn and winter seasons (Figure 3). Furthermore, decreasing trends were also found for mean monthly streamflow of Ilgaz Stream for March, April, May, July and August whereas the streamflow presented upward trend for other months (Figure 4).



Figure 3. Trend analysis results for mean seasonally streamflow of Ilgaz Stream

Knowledge on the availability and amount of water resources is of great importance for ensuring the sustainability of water resources. Understanding the past, present and future trends of water resources contribute to the knowledge on the water resources. Therefore, investigations and analyses of trends in the streamflow of rivers play significant role for understanding the trends and make significant contributions to the knowledge.



Figure 4. Trend analysis results for mean monthly streamflow of Ilgaz Stream

#### Discussion

Many scientists have paid great attention to the investigate the trends in water resources (Douglas et al., 2000; Zhang et al., 2001; Birsan et al., 2005; Kundzewicz et al., 2005; Zhang et al., 2006; Salarijazi et al., 2012; Herawati et al., 2015; Yeh et al., 2015; Zhou et al., 2015; Pumo et al., 2016; Kişi et al., 2018). Likewise, numerous scientists investigated the trends in streamflow of rivers in Turkey. Özel et al. (2004) indicated that monthly streamflow of Sakarya River basin generally presented downward trend. Kahya and Kalaycı (2004) found decreasing trends for river basins in the west part of Turkey. Similarly, Topaloğlu (2006) reported significant decreasing trends for the streamflow in western Turkey. On the other hand, Topaloğlu (2006) informed that streamflow of few rivers tended to increase which located in the Black Sea. Ozkul (2009) reported that there were downward trends in streamflow of Büyük Menderes and Gediz rivers. Durdu (2010) determined a decreasing trend for streamflow of Büyük Menderes River. Bahadır (2011) documented a decreasing trend in the streamflow of Kızılırmak River. Koçman and Sütgibi (2012) indicated that streamflow of Gediz River basin had a decreasing trend. Saplioğlu et al. (2014) determined significant decreasing trends for monthly and annual streamflow of streams in the western Mediterranean Basin of Turkey. Tekkanat and Sarış (2015) noted that there was significant decreasing trend in Porsuk Creek Basin. Ejder et al. (2016a) informed about a decreasing trend for the streamflow of Sarıçay Stream. Ejder et al. (2016b) stated that a decreasing trend found in the streamflow of Kocabaş Stream. Kale et al. (2016a) noticed a downward trend for the streamflow of Karamenderes River. Kale et al. (2016b) found a decreasing trend in the streamflow of Bakırçay River. Tekkanat (2017) stated that there were noticeable decreasing trends for all months except May and June in the streamflow of Porsuk Creek. On the other hand, increasing trends were detected for May and June. Kale et al. (2018) reported that there were downward trends in the streamflow of Büyük Menderes, Gediz, and Tuzla Rivers. Furthermore, Sönmez and Kale (2018) indicated a downward trend for the streamflow of Filyos River. Kale and Sönmez (2018b) pointed out a decreasing trend in the streamflow of Daday Stream. Kale and Sönmez (2018a) stated that streamflow of Akkaya Stream tended to decrease. Özel et al. (2004) determined decreasing trends for mean monthly streamflow in Sakarya River Basin. Yerdelen (2013) found a downward trend for streamflow of Susurluk Basin. Decreasing and increasing trends observed in the streamflow of Ilgaz Stream for the monitoring period were found statistically insignificant. Although determine trends in the streamflow were statistically insignificant, these fluctuations could be related to the climate change, global warming, snow melting. Kale (2017a) put forward that temperature had tendency to increase and Kale (2017b) pointed out that evaporation has also tended to increase. Rising temperature and evaporation could decrease the streamflow while increase in the precipitation could increase the streamflow. On the other hand, decline in the precipitation and snow melting might lead to decrease in the

streamflow. Therefore, changes in climatic conditions have important impacts on the streamflow trends. Tekkanat (2017) indicated that hydrological drought in river basin could result in declined streamflow and climatic factors (such as temperature, relative humidity, rainfall) had impacts on the streamflow. Sönmez and Kale (2018) pointed out that streamflow meaningfully interrelated with the precipitation. Authors informed about that decreasing precipitation and increasing temperature affected the streamflow and led to decrease in the streamflow of Filyos River. Kale and Sönmez (2018b) stated that decreasing trends in annual, seasonally and monthly streamflow of Daday Stream were associated with the climatic changes mainly rainfall and temperature. Similarly, Kale and Sönmez (2018a) indicated that decreasing trends in annual, seasonally and monthly streamflow of Daday Stream were associated with the climatic changes mainly rainfall and temperature. Similarly, Kale and Sönmez (2018a) indicated that decreasing trend in the streamflow of Akkaya Stream could be connected with the decline in snow melting and precipitation in addition to rising temperature attributable to the climate change. For that reason, assessing potential climate change impacts on the river streamflow by continuously monitoring has a vital importance. Trend analysis implementation might help for the monitoring of trends in water resources. Therefore, the findings of this paper will contribute to the knowledge of the past, present and future of water resources. Water resources managers and decision-makers should take into account the results of the paper to ensure the sustainability of water resources and make applicable management strategies.

### 4. CONCLUSION

The results of trend analyses indicated that annual streamflow presented a decreasing trend during the monitoring period. In addition, decreasing trends were also found for the seasonal streamflow of spring and summer though the streamflow of autumn and winter presented increasing trends. Nonetheless, these trends in the streamflow were found statistically insignificant for all periods covering annual, seasonally and monthly time series. The streamflow of the river could be affected by the increase in temperature and evaporation, the reduction in rainfall, and additional reasons occurred by the climate change. Henceforward, the trends in the streamflow should be determined and monitored to forecast the further availability and amount of rivers and other water resources.

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#### REFERENCES

Anonim (2013a) Taşkın Direktifinin Uygulanması İçin Kapasitenin Geliştirilmesi. Avrupa Birliği Eşleştirme Projesi Su Yönetimi Genel Müdürlüğü.

Anonim (2013b) İhsangazi İlçe Analizi. Kuzey Anadolu Kalkınma Ajansı.

- Bahadir M (2011) A Statistical Analysis of the Flow Changes of Kızılırmak River. Turkish Studies International Periodical for the Languages, Literature and History of Turkish or Turkic, 6: 1339-1356.
- Birsan M V, Molnar P, Burlando P & Pfaundler M (2005) Streamflow Trends in Switzerland. Journal of Hydrology, 314: 312-329.
- Box G E P & Jenkins G (1976) Time Series Analysis: Forecasting and Control. Holden Day, San Francisco.
- Cigizoglu H K, Bayazit M & Onoz B (2005) Trends in the Maximum, Mean and Low Flows of Turkish Rivers. Journal of Hydrometeorology, 6 (3): 280-290.
- Douglas E M, Vogel R M & Kroll C N (2000) Trends in Floods and Low Flows in the United States: Impact of Spatial Correlation. Journal of Hydrology, 240 (1–2): 90-105.
- Durdu Ö F (2010) Effects of Climate Change on Water Resources of the Büyük Menderes River Basin, Western Turkey. Turkish Journal of Agriculture and Forestry, 34: 319-332.
- Ejder T, Kale S, Acar S, Hisar O & Mutlu F (2016a) Restricted Effects of Climate Change on Annual Streamflow of Sarıçay Stream (Çanakkale, Turkey). Marine Science and Technology Bulletin, 5 (1): 7-11.
- Ejder T, Kale S, Acar S, Hisar O & Mutlu F (2016b) Effects of Climate Change on Annual Streamflow of Kocabaş Stream (Çanakkale, Turkey). Journal of Scientific Research and Reports, 11 (4): 1-11.
- Ercan B & Yüce M İ (2017) Trend Analysis of Hydro-Meteorological Variables of Kızılırmak Basin. Nevşehir Bilim ve Teknoloji Dergisi, 6 (ICOCEE 2017 Özel Sayı): 333-340.

- Helsel D R & Hirsch R M (2002) Statistical Methods in Water Resources Techniques of Water Resources Investigations, Book 4, Chapter A3. U.S. Geological Survey. 522 p.
- Herawati H, Suripin & Suharyanto (2015) Impact of Climate Change on Streamflow in the Tropical Lowland of Kapuas River, West Borneo, Indonesia. Procedia Engineering, 125: 185-192.
- Hisar O, Kale S & Özen Ö (2015) Sustainability of Effective Use of Water Sources in Turkey. In: Leal Filho, W., Sümer, V. (Eds.), Sustainable Water Use and Management: Examples of New Approaches and Perspectives. Springer International Publishing, Switzerland, pp. 205-227.
- Kahya E & Kalaycı S (2004) Trend Analysis of Streamflow in Turkey. Journal of Hydrology, 289: 128-144.
- Kale S (2017a) Climatic Trends in the Temperature of Çanakkale City, Turkey. Natural and Engineering Sciences, 2 (3): 14-27.
- Kale S (2017b) Analysis of Climatic Trends in Evaporation for Çanakkale (Turkey). Middle East Journal of Science, 3 (2): 69-82.
- Kale S & Sönmez A Y (2018a) Trend Analysis of Streamflow of Akkaya Stream (Turkey). In: Proceedings of the 1st International Conference on Food, Agriculture and Animal Sciences, Antalya, Turkey. pp. 33-45.
- Kale S & Sönmez A Y (2018b) Trend Analysis of Mean Monthly, Seasonally and Annual Streamflow of Daday Stream in Kastamonu, Turkey. Marine Science and Technology Bulletin, 7 (2): 60-67.
- Kale S, Ejder T, Hisar O & Mutlu F (2016a) Climate Change Impacts On Streamflow of Karamenderes River (Çanakkale, Turkey). Marine Science and Technology Bulletin, 5 (2): 1-6.
- Kale S, Ejder T, Hisar O & Mutlu F (2016b) Effect of Climate Change on Annual Streamflow of Bakırçay River. Adıyaman University Journal of Science, 6 (2): 156-176.
- Kale S, Hisar O, Sönmez A Y, Mutlu F & Filho W L (2018) An Assessment of the Effects of Climate Change on Annual Streamflow in Rivers in Western Turkey. International Journal of Global Warming, 15 (2): 190-211.
- Kendall M G (1955) Rank Correlation Methods. 2nd ed. Hafner Publishing Co., New York. 196 p.
- Kişi Ö, Guimarães Santos C A, Marques da Silva R & Zounemat-Kermani M (2018) Trend Analysis of Monthly Streamflows using Şen's Innovative Trend Method. Geofizika, 35 (1): 53-68.
- Koçman A & Sütgibi S (2012) Hydrograpic/Hydrologic Characteristics of Gediz River Basin in the Context of the Environmental Components Problems and Suggestions. Eastern Geographical Review, 28: 155-174.
- Kundzewicz Z W, Graczyk D, Maurer T, Pińskwar I, Radziejewski M, Svensson C & Szwed M (2005) Trend Detection in River Flow Series: 1. Annual Maximum Flow. Hydrological Sciences–Journal–des Sciences Hydrologiques, 50 (5): 797-810.
- Lehmann EL (1975) Nonparametrics: Statistical Methods Based on Ranks. Holden-Day, San Francisco, CA, USA. 457 p.
- Mann H B (1945) Nonparametric Tests against Trend. Econometrica, 13: 245-259.
- Ozkul S (2009) Assessment of Climate Change Effects in Aegean River Basins: The Case of Gediz and Buyuk Menderes Basins. Climatic Change, 97: 253-283.
- Özel N, Kalaycı S, Sevimli F M & Büyükyıldız M (2004) Trend Analysis of Monthly Streamflow Data by Using Nonparametric Methods in Sakarya River Basin. Journal of The Engineering and Architecture Faculty of Selcuk University, 19 (2): 11-22.
- Pettitt A N (1979) A Non-Parametric Approach to the Change-Point Problem. Journal of the Royal Statistical Society. Series C (Applied Statistics), 28: 126-135.
- Pumo D, Caracciolo D, Viola F & Noto L V (2016) Climate Change Effects on the Hydrological Regime of Small Non-Perennial River Basins. Science of The Total Environment, 542 (Part A): 76-92.
- R Core Team (2019) R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria [online] https://www.R-project.org/ version (01/2019).
- Saplıoğlu K, Kilit M & Yavuz B K (2014) Trend Analysis of Streams in the Western Mediterranean Basin of Turkey. Fresenius Environmental Bulletin, 23 (1): 1-12.
- Sen P K (1968) Estimates of the Regression Coefficient Based on Kendall's Tau. Journal of the American Statistical Association, 63 (324): 1379–1389.

- Sneyers R (1990) On the Statistical Analysis of Series of Observations. World Meteorological Organization, Technical Note no. 143, WMO no. 415. 192 p.
- Sütgibi S (2015) Büyük Menderes Havzasının Sıcaklık, Yağış ve Akım Değerlerindeki Değişimler ve Eğilimler. Marmara Coğrafya Dergisi, 31: 398-414
- Salarijazi M, Akhond-Ali A-M, Adib A & Daneshkhah A (2012) Trend and Change-Point Detection for the Annual Stream-Flow Series of the Karun River at the Ahvaz Hydrometric Station. African Journal of Agricultural Research, 7 (32): 4540-4552.
- Sönmez A Y & Kale S (2018) Climate Change Effects on Annual Streamflow of Filyos River (Turkey). Journal of Water and Climate Change (*In press*). DOI: 10.2166/wcc.2018.060.
- Şen Z (2012) Innovative Trend Analysis Methodology. Journal of Hydrologic Engineering, 17 (9): 1042-1046.
- Tekkanat İ S (2017) Porsuk Çayı Üst Kesiminde Ortalama Akımların Şen Trend Metodu Kullanılarak Araştırılması. Coğrafi Bilimler Dergisi, 15 (1): 49-57.
- Tekkanat İ S & Sarış F (2015) Porsuk Çayı Havzasında Akarsu Akımlarında Gözlenen Uzun Dönemli Eğilimler. Türk Coğrafya Dergisi, 64: 69-83.
- Topaloğlu F (2006) Trend Detection of Streamflow Variables in Turkey. Fresenius Environmental Bulletin, 15 (7): 644-653.
- Tosunoglu F & Kisi O (2017) Trend Analysis of Maximum Hydrologic Drought Variables Using Mann–Kendall And Şen's Innovative Trend Method. River Research and Applications, 33: 597-610.
- Tot A, Kadak A E & Aras S (2017) Research on Physico-Chemical Properties of Ilgaz River Basin (Kastamonu). Menba Journal of Fisheries Faculty, 2 (1-2): 8-16.
- Yeh C F, Wang J, Yeh H F & Lee C H (2015) Spatial and Temporal Streamflow Trends in Northern Taiwan. Water, 7 (2): 634-651.
- Yenigün K & Ülgen M U (2016) Trend Analysis of Maximum Flows under Climate Change Evaluation and Its Impact on Spillway. Disaster Science and Engineering, 2 (1): 25-28.
- Yerdelen C (2013) Investigation of Trend Analysis and Change Point Detection for Annual Mean Streamflows of Susurluk Basin. DEÜ Mühendislik Fakültesi Mühendislik Bilimleri Dergisi, 15 (2): 77-87.
- Zhang Q, Liu C, Xu C, Xu Y & Jiang T (2006). Observed Trends of Annual Maximum Water Level and Streamflow During Past 130 Years in the Yangtze River Basin, China. Journal of Hydrology, 324 (1-4): 255–265.
- Zhang X, Harvey K D, Hoggy W D, Yuzyk T R (2001) Trends in Canadian Streamflow. Water Resources Research, 37 (4): 987-998.
- Zhou Y, Shi C, Fan X & Shao W (2015) The Influence of Climate Change and Anthropogenic Activities on Annual Runoff of Huangfuchuan Basin in Northwest China. Theoretical and Applied Climatology, 120: 137-146.



# Cytokine Responses of *Prunus domestica* Aqueous Methanolic Extract on Head-kidney Leucckocyte Cell-line of Rainbow Trout (*Oncorhynchus mykiss*)

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**Abstract:** In this in vitro study, effect of *Prunus domestica* (PD) aqueous methanolic extract on immune related gene expression in rainbow trout was investigated. In the study, PD extract was performed into rainbow trout leucocyte cell lines at the dose of 0 (Control), 5 10 and 15  $\mu$ g ml<sup>-1</sup> during 48 hours and 1, 4, 12, 24 and 48 hour of the study, cells were harvested and cytokine responses such as TGF- $\beta$ , TNF- $\alpha$ , IFN-1 and IL-8 gene expression were determined. TGF- $\beta$  showed an increase at 1 H on 5  $\mu$ g group compared to control. Increased TGF-B results were also observed oat 24 hour on 5 and 15  $\mu$ g groups (P<0.05). at the end of the study all groups TGF-B expression was significantly increased up to 80 fold chance compared to control. TNF- $\alpha$  was significantly increased ln all experimental group except 48 h of the study. IFN-1 gene expression was significantly increased in all experimental compared to control. This increase was also observed at 48 hour of the study as well. These results suggest that *Prunus domestica* (PD) aqueous methanolic extract showed a regulatory effects on rainbow trout leukocyte cytokines.

Keywords: Cytokine, Prunus domestica, rainbow trout, gene expression


## The Efficiency of Different Disinfectants on Fish Eviscerating Knives

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Abstract: Disinfection of equipment and environmental surfaces in fresh seafood processing plants are very important for producing high-quality value products. Disinfectant substances exhibit diversity. Technological advances nowadays enable the production of new advanced nano-particulate disinfectants. Their use and their efficacy on conventional disinfectants are investigated. In this study, the efficacy of colloidal silver added hydrogen peroxide and hydrogen peroxide on the microbiological load of the knives used in fish internal organ extraction (eviscerating) was compared. Fresh-farmed sea bass and sea bream were used as fish material. The total number of aerobic mesophilic bacteria (TAMB) was analyzed for microbiological analysis. For this purpose, it was performed using aerobic mesophilic bacteria petrifilm. Commercially used, plastic handled (plastic part) fillet knives with steel tip (stainless still part) were preferred. Microbiological analysis was made separately from the tip and handle of the knives. Groups for two fish species and two disinfectant liquids were formed. Tap water was used for the control group solution. Groups were set as GL, colloidal silver added hydrogen peroxide group for seabass; GC, colloidal silver added hydrogen peroxide group for seabream; HL hydrogen peroxide group for seabass; HC, hydrogen peroxide group for seabream; KL represents the control group for seabass and KC control group knives for seabream. The analyses were performed three times as initial, after evisceration and 3 minutes after the immersion in disinfectant solutions (with 5% concentrations). The initial total bacterial load of knives' tips was examined as <10 log CFU/cm<sup>2</sup> for all groups except GC (1.49 log CFU/cm<sup>2</sup>). The initial TAMB of the handle of knives were determined as 1.45, 0.6, 1.27, 1.72, 0.7, and 1.38 log CFU/ cm<sup>2</sup> for HC, GC, HL, GL, KC, and KL, respectively. After eviscerating TAMB were reached higher values almost for all groups both tip and handles of knives due to internal organs microbiological loads. The handle of knives TAMB was reached to 2.09, 1.47, 2.52, 2.36, 1.84, and 2.50 log CFU/cm<sup>2</sup> for HC, GC, HL, GL, KC, and KL, respectively. Tip of knives TAMB was counted as 1.6, 1.62, 3.17, 3.19, 2, and 2.87 log CFU/cm<sup>2</sup> for HC, GC, HL, GL, KC, and KL, respectively. All knives were dipped into the prepared solutions for 3 minutes and the microbiological load decreased. According to the results, it was determined as <10 log CFU/cm<sup>2</sup> for HC, GC, HL, and GL groups (both tip and handle of knives). However, the handle of KC and KL groups were determined as 0.77 and 2.17 log CFU/cm<sup>2</sup>, respectively. While the tip of knives was 1.39 and 2.33 log CFU/cm<sup>2</sup>, respectively. These results show that 5% solutions of colloidal silver added hydrogen peroxide and hydrogen peroxide are effective to reduce the microbiological load of the fish-eviscerating knives. For future studies, our suggestion is to conduct researches to determine the effectiveness of these solutions on specific microorganisms.

Keywords: Sanitizing, hydrogen peroxide, aerobic mesophilic bacteria, eviscerating, fish

## **1. INTRODUCTION**

Food production processes have been changing due to more food demand (FAO, 2009). The production volume is getting larger and processed food product variety is getting more complex. These changes made sanitation and disinfection of all the equipment used during the food processing process even more important (Betta, Barbanti, and Massini 2011). This is not just an issue of concern to the food business but also controlled by the regulating authorities at the highest level.

Disinfection and sanitation are quite comprehensive. Different applications are applied in many stages. Surface disinfection is one of the most important application. A method for surface sanitation is selected depending on the characteristics of the equipment used, the food product and the requirement of the plant. These surface disinfectants are usually selected from mild chemical solutions to reduce trace elements. In addition, it is expected to have advantages such as being cheap and easy to find. Hydrogen peroxide is a surface disinfectant with these properties (Atar, Akbaş, and Ayvaz 2018). Also in recent years, different concentration of this chemical or nanoparticles added kinds are also produced.

There are limited scientific studies on the efficacy of this chemical, which is widely used as a surface disinfectant in seafood industry.

Eviscerating is one of the first processes before filleting in fish processing plants. This step is performed with a steel fillet knife. The use of fillet equipment is also an option. However, the use of labor is also common. When the fillet knives are used, the microorganisms in the internal organs of the fish are transmitted to these knives. It is therefore important to disinfect these knives to reusing.

The aim of this study was to evaluate efficiencies of colloidal silver added hydrogen peroxide and hydrogen peroxide on the microbiological load of the knives used in fish internal organ extraction (eviscerating).

## 2. MATERIAL AND METHOD

Fresh farmed fish samples, seabream, and seabass were purchased from a local fish market. Fish samples were transported to the laboratory in ice. Whole 30 seabreams and 30 seabasses were used to eviscerating with knives.

The chemicals used in the study were hydrogen peroxide (30%, aqueous) (Tekkim Inc., İstanbul, Turkey) and colloidal silver added hydrogen peroxide (50%, aqueous) (SilverTech Kimya Inc., Ankara, Turkey). Five percent solutions of these chemicals were prepared as an immersion solution. Tap water was used for control groups. The knives were immersed in these solutions for 3 minutes after eviscerating.



Figure 1. The knives used in the study

The stainless still fillet knives (FNK-798, Rooc, King Fish Knife, China) with 279 mm size (Figure 1) were purchased from a local market. The knife groups used in the study were determined as follows:

- GL: Colloidal silver added hydrogen peroxide group knives for seabass,
- GC: Colloidal silver added hydrogen peroxide group knives for seabream,
- HL: Hydrogen peroxide group knives for seabass,
- HC: Hydrogen peroxide group knives for seabream,
- KL: Control group knives for seabass,
- KC: Control group knives for seabream.

The microbiological sample collection procedure was conducted before eviscerating (initial), after eviscerating and after immersion in triplicate. The microbiological analyses were done as follows. First, an area was selected about  $100 \text{ cm}^2$  area on the knife handle and stainless still tip parts (both side) separately. A sterile swab used to the collection. Swabbing was done from the determined surface. The swab sponges were stomached for 2 min. The samples were added to the total aerobic plate count samples for dilution. Serial dilutions of the total aerobic plate count samples were prepared and plated

on 3M Petrifilm aerobic count plates (3M Corporation, Maplewood, Minnesota, U.S.A) following AOAC official method 990.12 (AOAC, 2002). Petrifilm plates were incubated for 48 h at  $35\pm 1^{\circ}$ C, and the colonies were counted.

## **3. RESULTS AND DISCUSSION**

## Results

The initial TAMB of the knives was determined as 1.45, 0.6, 1.27, 1.72, 0.7, and 1.38 log CFU/ cm<sup>2</sup> for HC, GC, HL, GL, KC, and KL, respectively (Figure 2). After eviscerating the TAMB of the samples dramatically increased. It was reached to 2.09, 1.47, 2.52, 2.36, 1.84, and 2.50 log CFU/cm<sup>2</sup> for HC, GC, HL, GL, KC, and KL, respectively. After eviscerating, the samples were immersed in the prepared solutions. According to the results, TAMB of the knives handles was determined as  $<10 \log CFU/cm<sup>2</sup>$  for HC, GC, HL, and GL groups. However, the handle of KC and KL groups were determined as 0.77 and 2.17 log CFU/cm<sup>2</sup>, respectively.



Figure 2. TAMB of the knives handle of the groups during process

The initial total bacterial load of knives' tips was examined as  $<10 \log \text{CFU/cm}^2$  for all groups except GC (1.49 log CFU/cm<sup>2</sup>) (Figure 3). After eviscerating, TAMB value of the samples was increased due to internal organ microbiological load of the fish. TAMB of knives tips TAMB was counted after evisceration as 1.6, 1.62, 3.17, 3.19, 2, and 2.87 log CFU/cm<sup>2</sup> for HC, GC, HL, GL, KC, and KL, respectively. The knives were immersed in the solutions. According to the results, it was determined as  $<10 \log \text{CFU/cm}^2$  for HC, GC, HL, and GL groups after immersion. While the tip of knives was 1.39 and 2.33 log CFU/cm<sup>2</sup>, respectively.



Figure 3. TAMB of the knives tip of the groups during process

#### Discussion

These results show that 5% solutions of colloidal silver added hydrogen peroxide and hydrogen peroxide are effective to reduce the microbiological load of the fish-eviscerating knives surface. This probably due to the hydrogen peroxide surface disinfection effect (Lee, Hong, and Kim 2014). However, the results of the study did not reveal which solution had superior properties.

## 4. CONCLUSION

For future studies, our suggestion is to conduct researches to determine the effectiveness of colloidal silver added hydrogen peroxide and hydrogen peroxide solutions on specific microorganisms.

## REFERENCES

- AOAC International. 2002. Method 990.12. In Official methods of analysis of AOAC International. AOAC International, Gaithersburg, Md.
- Atar, Hasan Hüseyin, Saliha Akbaş, and Zayde Ayvaz. 2018. "Use Of Hydrogen Peroxide In Aquaculture And Aquatic Products." Ziraat Mühendisliği 366: 14–20.
- Betta, Giampaolo, Davide Barbanti, and Roberto Massini. 2011. "Food Hygiene in Aseptic Processing and Packaging System: A Survey in the Italian Food Industry." *Trends in Food Science & Technology* 22(6): 327–34. https://www.sciencedirect.com/science/article/pii/S0924224411000331 (November 19, 2018).
- Lee, Hyun-Hee, Seok-In Hong, and Dongman Kim. 2014. "Microbial Reduction Efficacy of Various Disinfection Treatments on Fresh-Cut Cabbage." *Food Science & Nutrition* 2(5): 585–90. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4237489/pdf/fsn30002-0585.pdf (October 23, 2018).
- FAO, 2009 . High Level Expert Forum How to Feed the World in 2050-Report. Office of the Director, Agricultural Development Economics Division Economic and Social Development Department Viale delle Terme di CaracallaRome Italy.



## Heavy Metal Concentrations in Cyprinus carpio From Sırakaraağaçlar Stream in Sinop

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**Abstract:** The purpose of this study was the investigation of Zn, Cu, Pb and Cd levels in entire gills, liver and muscle tissues in the common carp (*Cyprinus carpio*) collected from Sırakaraağaçlar Stream in Sinop Province. It is very important to determine the levels of heavy metals which can be a risk to people health. The highest concentration of Pb (0.66 mg kg<sup>-1</sup> wet wt.) was in gills, while Zn (23 mg kg<sup>-1</sup> wet wt.), Cu (6.2 mg kg<sup>-1</sup> wet wt.) and Cd (0.081 mg kg<sup>-1</sup> wet wt.) were deposited in liver. The lower concentrations of all metals were found in muscle tissue. Zn, Cu, Pb and Cd in edible tissues of the common carp were 6, 0.6, 0.025 and 0.015 mg kg<sup>-1</sup> wet wt., respectively. Concentrations of all metals in edible tissues of *C. carpio* were below the maximum of acceptable concentrations and the fish is acceptable for the human consumption. With this study for the first time heavy metal amounts in common carp in the region was carried out.

Keywords: Sırakaraağaçlar Stream, heavy metal, Cyprinus carpio, estimated daily intakes

## **1. INTRODUCTION**

One of the important problems is presence environmental contaminants especially heavy metals in aquatic ecosystems. Essential heavy metals such as Zn and Cu are important for many biochemical processes in living organisms and they are essential elements for aquatic biota. However, in the case of higher levels of those metals, they become toxic. Many fish species and their feed habits have a great influence on accumulation of contaminants. Moreover, in fish species heavy metals can be accumulated in different levels in their tissues. Liver and gills accumulate heavy metals in higher amounts, while muscles or edible tissues contain lower amounts (Bat, 2014). Consumption of fish as food due to protein and omega fatty acids directly affects human health. It is therefore important to detect heavy metal amounts in fish, since toxic effect of heavy metal presence in their body can result in hazardous effects on human health (Bat, 2017a; Bat and Arici, 2018).

Sırakaraağaçlar Stream flows into Akliman location of Sinop coasts in the Black Sea. It is located at 42° 2' 24" N, 35° I' 5" E and 42° 2' 40" N 35° 2' 10"E. The average depth of Sırakaraağaçlar Stream is 1.5 m and maximum depth is 2.5 m. Its bottom is muddy and the mouth of the streams sandy. *C. carpio* inhabits in Sırakaraağaçlar Stream naturally. There are areas of settlement and agriculture around the stream to which waste water is discharged (Bat et al., 2000). Thus it is necessary to determine amounts of contaminants, as heavy metals in order to reduce pollution and to provide background data.

The purpose of this study is to determine the amounts of heavy metals in the entire gills, liver and muscle tissues of *C. carpio* from Sırakaraağaçlar Stream of Sinop Province as an indicator of environmental pollution and health risk assessment.

## 2. MATERIAL AND METHOD

Fish samples were taken from the Sırakaraağaçlar Stream of Sinop Provincial (Figure 1) during autumn of 2016. This stream passes through the agricultural area and near small locality.



Figure 1. Study area

The fish samples were dissected in order to obtain muscle, livers and entire gills that were mixed homogeneously and kept at -21 °C till analysis. For determination of heavy metal contents in fish samples, digested with Suprapur® HNO<sub>3</sub> using Milestone Systems, Start D 260 microwave for metal analysis. All reagents used in the analysis were of reagent grade. The element standard solutions from Merck that were used for the calibrations were made by diluting stock solutions of 1000 mg l<sup>-1</sup>. After digestion, the heavy metal levels were analysed by ICP-MS (Agilent Technologies, 7700X). Metal concentrations in fish tissues were determined on wet weight basis as milligram of metal per kilogram. Standard reference material TORT-3 lobster hepatopancreas for metals was used to determine the reliability of the analysis. All digested samples were analysed at least three times for each metal.

The estimated daily intake (EDI) depends on both the metal concentration level and the amount of consumption of fish. The EDI of metals was determined using the following equation:

$$EDI = \frac{c_{metal} \, x \, W}{bwt} \tag{1}$$

Where:  $C_{metal}$  is the concentration of metals in fish; W represents the daily average consumption of fish; bwt is the body weight. EWI values were calculated from EDI values. Intake estimates were expressed as per unit body weight (mg kg<sup>-1</sup> body wt./weekly). Fish consumption is approximately to 27.4 g day<sup>-1</sup> in Sinop (Bat, 2017b). This value is equal to about 191.8 grams.

## 3. RESULTS AND DISCUSSION

#### Results

Recovered values of all metals range between 97% and 104% of the certified value. The highest concentration of Pb (0.66 mg kg-1 wet wt.) was in gills, while Zn (23 mg kg<sup>-1</sup> wet wt.), Cu (6.2 mg kg<sup>-1</sup> wet wt.) and Cd (0.081 mg kg<sup>-1</sup> wet wt.) were deposited in liver (Figures 2-5). The lower concentrations of all metals were found in muscle tissue. Zn, Cu, Pb and Cd in edible tissues of the common carp were 6, 0.6, 0.025 and 0.015 mg / kg wet wt., respectively.



**Figure 2.** Mean concentration and standard deviation of Pb (mg kg<sup>-1</sup> wet wt.) in muscle, gills and livers of *Cyprinus carpio* from Sırakaraağaçlar Stream in 2016.



**Figure 3.** Mean concentration and standard deviation of Cu (mg kg<sup>-1</sup> wet wt.) in muscle, gills and livers of *Cyprinus carpio* from Sırakaraağaçlar Stream in 2016.



**Figure 4.** Mean concentration and standard deviation of Pb (mg kg<sup>-1</sup> wet wt.) in muscle, gills and livers of *Cyprinus carpio* from Sırakaraağaçlar Stream in 2016.



**Figure 5.** Mean concentration and standard deviation of Cd (mg kg<sup>-1</sup> wet wt.) in muscle, gills and livers of *Cyprinus carpio* from Sırakaraağaçlar Stream in 2016.

#### Discussion

The significantly highest concentration (p<0.05) of Pb was found in entire gills of *C. carpio* and distribution of Pb decreases in a following order: gills > liver > muscle. The significant higher concentrations of Cd, Cu and Zn were present in liver of *C. carpio*. The most deposited metal in gills was Pb which is 26.4 times higher than in edible tissues. However the concentration of Cd, Cu and Zn in liver was 5.4, 10.3 and 3.8 times higher than in edible tissues, respectively.

The metal levels in muscle samples of *C. carpio* are lower than the other tissues and this result is in agreement with previous researches from Altınkaya Dam Lake (Öztürk et al., 1995; Bat and Öztekin, 2018), Balık Lake (Bat et al., 2015) of Samsun Province and Sarikum Lake of Sinop Province (Bat et al., 2019).

The rise amounts of heavy metals in liver and gills is the result of higher levels of these heavy metals in surrounding ecosystem and poses an ecological damage to biota in aquatic environment. Fish samples were caught in autumn after agricultural treatment and these results can be explained by this influence. One of the reasons for these results is the fact that several branches flow into the sampling point in Sırakaraağaçlar Stream, and small localities can be domestic contaminants of the environment. *C. carpio* is widespread in lakes and rivers and feeds mainly on the bottom on water plants, worms, crustaceans and it can be one of the reasons for their higher metal bioaccumulation in liver. However, the liver and gills of the fish are not consumed. In this study, thus, the following research of health risk assessment, amounts of Cd, Pb, Cu and Zn were calculated in the edible tissues of *C. carpio* (Table 1).

**Table 1.** Estimated Weekly Intakes (EWI) of heavy metals in edible muscles of *C. carpio* from Sırakaraağaçlar Stream in Sinop Province.

| Metals  | PTWI <sup>a</sup> | <b>PTWI</b> <sup>b</sup> | EWI <sup>c</sup> |  |  |  |
|---|-------------------|--------------------------|------------------|--|--|--|
| Pb  | 0.025             | 1.75                     | 0.0048           |  |  |  |
| Cd  | 0.007             | 0.49                     | 0.0029           |  |  |  |
| Cu  | 3.5               | 245                      | 0.1151           |  |  |  |
| Zn  | 7                 | 490                      | 1.1508           |  |  |  |
| <sup>a</sup> PTWI (Provisional Tolerable Weekly Intake) in mg /week/kg body wt. |                   |                          |                  |  |  |  |
| <sup>b</sup> PTWI for 70 kg adult person (mg /week/70 kg body wt.)              |                   |                          |                  |  |  |  |
| <sup>c</sup> EWI (Estimated Weekly Intake) (mg /week/ 70 kg body wt.)           |                   |                          |                  |  |  |  |

The daily intakes (EDI) of the metals were estimated taking into account the mean of heavy metal levels in the edible tissues and the mean consumption of fish per day for adults. PTWI is the maximum amount of a contaminant to which a person can be exposed per week over a lifetime without an unacceptable threat of health effects (FAO/WHO, 2010 and 2011). These results are significantly lower than the recommended values of the Food and Agriculture Organization/World Health Organization (FAO/WHO) Joint Expert Committee on Food Additives (JECFA).

Cd and Pb are not essential metals for living organisms and their presence in edible tissues of fish are limited. The Turkish Food Codex (TFC, 2008) and the Commission of the European Communities (EU, 2006) established the maximum amounts of non-essential metals in fish muscles and Cd and Pb in muscles were determined (0.025 and 0.015 mg kg<sup>-1</sup> wet weight, respectively) in concentrations far from the proposed concentrations for Cd and Pb in edible tissues of fish (0.05 and 0.3 mg kg<sup>-1</sup> wet weight, respectively).

## 4. CONCLUSION

Present study provides new information on the concentration of heavy metals in *C. carpio* from Sırakaraağaçlar Stream in Sinop Province. Based on the sample analysis, metal concentrations found in the edible part of fish were below the limit values for fish proposed by TFC and EC, hence fish were suitable for people consumption in this region. Even though, there are no high amounts of heavy metals in the common carp a possible threat may occur in the future depending on the agricultural and domestic effluent in this area. It is essential to monitor the existence of heavy metals as environmental contaminants in the future, consequently making a good ecological status of aquatic environment, as well as supplying the healthy and safe fresh fish.

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## REFERENCES

- Bat L, Akbulut M, Çulha M, Sezgin M (2000). The macrobenthic fauna of Sırakaraağaçlar Stream flowing into the Black Sea at Akliman, Sinop. Turkish J. Mar. Sci, 6 (1): 71-86.
- Bat L (2014). Heavy metal pollution in the Black Sea. In: Düzgüneş E, Öztürk B, Zengin M, eds. Turkish Fisheries in the Black Sea. Published by Turkish Marine Research Foundation (TUDAV), Publication number: 40, ISBN: 987-975-8825-32-5 Istanbul, Turkey, pp. 71-107.
- Bat L, Arıcı E, Sezgin M, Şahin F (2015). Heavy metal levels in the liver and muscle tissues of the four commercial fishes from Lake Balik, Kızılırmak Delta (Samsun, Turkey). Journal of Coastal Life Medicine, 3(12): 950-955.
- Bat L (2017a). The contamination status of heavy metals in fish from the Black Sea, Turkey and potential risks to human health. In: Sezgin M, Bat L, Ürkmez D, Arici E, Öztürk B, eds. Black Sea Marine Environment: The Turkish Shelf. Turkish Marine Research Foundation (TUDAV), Publication No: 46, ISBN- 978-975-8825-38-7, Istanbul, TURKEY, pp. 322-418.

- Bat L (2017b). Kirlilik ve Balıkçılık (Pollution and fishing). In: Karadeniz ve Balıkçılık Çalıştayı Kitabı (Black Sea and Fishing Workshop Book) 13-14 Ekim 2016, Sinop (Editörler: Sezgin M, Şahin F, Özsandıkçı U). Sinop Üniversitesi 13. Bilimsel Yayın. Şimal Ajans Sertifika No: 21439; ISBN: 978-605-88024-3-8, 17-54.
- Bat L, Arici E (2018). Chapter 5. Heavy metal levels in fish, molluscs, and crustacea from Turkish seas and potential risk of human health. In: Holban AM, Grumezescu AM, eds. Handbook of Food Bioengineering, Volume 13, Food Quality: Balancing Health and Disease. Elsevier, Academic Press, ISBN: 978-0-12-811442-1, pp. 159-196.
- Bat L, Öztekin A (2018). Heavy metal levels in muscle, livers and digestive tracts of Cyprinus carpio L., 1758 from Altınkaya Dam Lake of Samsun Province, Turkey. First International Marine & Freshwater Sciences Symposium (MarFresh2018), 18-21 October 2018 Kemer, Antalya, Turkey. pp. 185-189.
- Bat L, Yardım Ö, Öztekin A, Sahin F (2019). Bioaccumulation of metals in fish from Sarikum Lake. Aquatic Science and Technology, 7 (1): 1-7.
- Commission Regulation (EC) (2006). Official Journal of the European Union. Setting maximum levels for certain contaminants in food stuffs. Commission Regulation (EC) No 1881/2006, 364: 5-24.
- FAO/WHO (2010). Summary report of the seventy-third meeting of JECFA. Joint FAO/WHO Expert Committee on Food Additives. Geneva.
- FAO/WHO (2011). Joint FAO/WHO food standards programme codex committee on contaminants in foods, Fifth Session, working document for information and use in discussions related to contaminants and toxins in the GSCTFF, The Hague, The Netherlands, 90 pages.
- Öztürk M, Bat L, Öztürk M (1995). The heavy metal levels in different organs and tissues of carp Cyprinus carpio L. 1758, living in Altınkaya Dam Lake, Samsun. II. Ulusal Ekoloji ve Çevre Kongresi, 11-13 Eylül, Ankara, (inTurkish), pp. 650-667.

TFC (2008). Notification on the maximum limits of contaminants in foodstuffs (in Turkish). (Notification No: 2008/26), Issue: 26879.



## Fish Diversity of Homa Lagoon (Izmir Bay, Aegean Sea, Turkey)

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**Abstract:** Until 1980s, there were five lagoons (Homa, Çalıbaşı, Ragıppaşa, Kırdeniz and Çakalburnu) in Izmir Bay. At the present time, four of them lost their functions and vanished. Homa Lagoon remains the unique active lagoon for fishing and it located within Izmir Bay, North-eastern Aegean Sea. Fishery in Homa Lagoon is carried out with trammel nets, fyke nets and traditional barrier traps that were made from the pikes and powered by wooden stakes. Main target fishes are grey mullets (*Mugil cephalus, Chelon labrosus, C. auratus, C. saliens, C. ramada*), following gilthead seabream (*Sparus aurata*), sea bass (*Dicentrarchus labrax*) and some eel (*Anguilla anguilla*). However, many of fish species and their juvenile forms inhabit in the lagoon owing to good food availability. Homa lagoon system is a sensitive wetland where located in southern Gediz Delta (Special Protected Area, SPA no. 16). It has a total of 1873 ha surface area, and it has 7.4 km length, 3 km width and <1 m depth. This sensitive area within Gediz estuary has been under protected since 1980s. Both many birds and fishermen catch in the same area. But the diversity of fishes in the lagoon is unknown exactly. This study contributes the determining of fish diversity in the lagoon, and so far, a total of 31 fish species were determined from the Homa Lagoon, Izmir Bay. It is obviously that fish diversity of Homa lagoon might rise a little more in the future due to global warming. Namely, some alien fish observed nowadays in the Bay might also be seen in the lagoon.

Keywords: Fish diversity, lagoon, seine net, Homa, İzmir Bay, Aegean Sea

## **1. INTRODUCTION**

Until 1980s, there were five lagoons (Homa, Çalıbaşı, Ragıppaşa, Kırdeniz and Çakalburnu) in Izmir Bay. At the present time, four of them lost their functions and vanished. Homa Lagoon remains the unique active lagoon for fishing and it located within Izmir Bay, North-eastern Aegean Sea.

Homa Lagoon system is a sensitive wetland where located in southern Gediz Delta (Special Protected Area, SPA no. 16). It has a total of 1873 ha surface area, and it has 7.4 km length, 3 km width and <1 m depth. In terms of biodiversity, a total of 288 birds with about 50 000 immigrants per year in the area, 314 plants, 295 plankton, 67 benthic organisms, 74 fishes, 3 frogs, 25 reptiles and 10 mammals are determined in the SPA (Tosunoğlu et al 2017).

The lagoon has been used an experimental fishing area by Ege University Fisheries Faculty since 1986. As mentioned above, this area connected with a SPA, *i.e.* Gediz Delta is a "bird paradise" and it has been protected as a wildlife protected area since 1984 and by RAMSAR agreement since 1998 (Akyol 2005).

Fishery in Homa Lagoon is carried out with trammel nets, fyke nets and traditional barrier traps that were made from the pikes and powered by wooden stakes. Main target fishes are grey mullets (*Mugil cephalus, Chelon labrosus, C. auratus, C. saliens, C. ramada*), following gilthead seabream (*Sparus aurata*), sea bass (*Dicentrarchus labrax*) and some eel (*Anguilla anguilla*). However, many of fish species and their juvenile forms inhabit in the lagoon owing to good food availability. Barrier traps were constructed on the unique entrance of the lagoon between June and December to catch especially grey mullets. However, other adult fishes with grey mullets are targeted by using gillnets. Additionally, fyke nets are used to catch eels in the lagoon during the fishing season. For the next season, the barrier trap is abolished between January and June in order to allow to entrance of young fish individuals into the lagoon.

This study presents the fish diversity in Homa Lagoon, Izmir Bay via collected works and actual collection of fish species.

## 2. MATERIAL AND METHOD

This study carried out by using beach seine net in bimonthly basis in the Homa lagoon (Figure 1) between January 2015 and December 2015. Beach seine net had very small mesh and wings (Figure 2) to collect small and young fish. Fish species were fixed in 6% formalin solution to examine in the laboratory. Additionally, previous studies covers fish species in Homa lagoon were collected. For the taxonomy and systematics, (WoRMS 2019) was used.



Figure 1. Study Area (Homa Lagoon, Izmir Bay)



Figure 2. Beach Seine Net used the Fish Sampling

## 3. RESULTS AND DISCUSSION

(Akyol 2005) reported a total of 24 fish species in 1999, whereas (Acarlı et al 2014) determined 29 fish in the lagoon during 2009-2010. Thus, this study presents the reported fish up to now and new actual results, obtained by using beach seine in 2015 on fish diversity of the lagoon (Table 1).

| Species / Year                                     | <b>1999</b> <sup>[1]</sup> | <b>2009-10</b> <sup>[2]</sup> | 2015<br>this study |
|--|----------------------------|-------------------------------|--------------------|
| Anguilla anguilla (Linnaeus 1758)                  | +                          | +                             | -                  |
| Aphanius fasciatus (Valenciennes 1821)             | +                          | +                             | +                  |
| Atherina boyeri (Risso 1810)                       | +                          | +                             | +                  |
| Atherina hepsetus (Linnaeus 1758)                  | -                          | +                             | -                  |
| Belone belone (Linnaeus 1760)                      | +                          | +                             | -                  |
| Blennius ocellaris (Linnaeus 1758)                 | +                          | +                             | -                  |
| Chelon auratus (Risso 1810)                        | +                          | +                             | +                  |
| Chelon labrosus (Risso 1827)                       | +                          | +                             | +                  |
| Chelon ramada (Risso 1827)                         | +                          | +                             | +                  |
| Chelon saliens (Risso 1810)                        | +                          | +                             | +                  |
| Dicentrarchus labrax (Linnaeus 1758)               | +                          | +                             | +                  |
| Diplodus annularis (Linnaeus 1758)                 | +                          | +                             | +                  |
| Diplodus vulgaris (Geoffroy Saint-Hilaire 1817)    | +                          | +                             | -                  |
| Engraulis encrasicolus (Linnaeus 1758)             | -                          | +                             | -                  |
| Gobius niger (Linnaeus 1758)                       | -                          | +                             | +                  |
| Lithognathus mormyrus (Linnaeus 1758)              | -                          | +                             | +                  |
| Mugil cephalus (Linnaeus 1758)                     | +                          | +                             | +                  |
| Mullus barbatus barbatus (Linnaeus 1758)           | +                          | +                             | -                  |
| Mullus surmuletus (Linnaeus 1758)                  | +                          | +*                            | -                  |
| Parablennius sanguinolentus (Pallas 1814)          | +                          | -                             | -                  |
| Planiliza haematocheila (Temminck & Schlegel 1845) | +                          | -                             | -                  |
| Platichthys flesus (Linnaeus 1758)                 | +                          | +*                            | -                  |
| Pomatoschistus marmoratus (Risso 1810)             | +                          | +                             | +                  |
| Pomatoschistus minutus (Pallas 1770)               | -                          | +                             | -                  |
| Sardina pilchardus (Walbaum 1792)                  | +*                         | +                             | +*                 |
| Sarpa salpa (Linnaeus 1758)                        | -                          | +*                            | -                  |
| Solea solea (Linnaeus 1758)                        | +                          | +                             | -                  |
| Sparus aurata (Linnaeus 1758)                      | +*                         | +*                            | +*                 |
| Syngnathus acus (Linnaeus 1758)                    | +                          | +                             | -                  |
| Syngnathus typhle (Linnaeus 1758)                  | +                          | +                             | -                  |
| Zosterisessor ophiocephalus (Pallas 1814)          | -                          | +                             | -                  |

 Table 1. Fish Diversity in Homa Lagoon, Aegean Sea ([1] (Akyol 2005); [2] (Acarlı et al. 2014); \*juveniles)

## 4. CONCLUSION

A total of 31 fish species was identified in this study. It is obviously that fish diversity of Homa lagoon might rise a little more in the future due to global warming. Namely, some alien fish observed nowadays in the Bay might also be seen in the lagoon. Alien species recorded recently in Izmir Bay, such as *Siganus luridus, S. rivulatus, Echeneis naucrates, Etrumeus golani, Stephanolepis diaspros, Lutjanus argentimaculatus*, can settle into the lagoon in the next time.

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## REFERENCES

Acarlı D, Kara A & Bayhan B (2014). Length-weight Relations for 29 Fish Species from Homa Lagoon, Aegean Sea, Turkey. Acta Ichthyol. Piscat., 44(3):249-257.

Akyol O (2005). The Last Lagoon of Izmir Bay: Homa. Ekoloji Magazin, pp.12-16.

- Tosunoğlu Z, Ünal V & Kaykaç M H (2017). Ege Dalyanları. Su Ürünleri Kooperatifleri Merkez Birliği Yayınları No.3, Ankara, 322 s.
- WoRMS Editorial Board (2019). World Register of Marine Species. Retrieved in February, 14, 2019 from http://www.marinespecies.org at VLIZ.



## Length-Weight Relationship of the Bogue, *Boops boops* (Osteichthyes: Sparidae), Captured around the Sea Cage Fish Farms in the South-eastern Aegean Sea

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**Abstract:** Bogue, *Boops boops* (Linnaeus, 1758) is inshore schooling species found in mid-water above sandy, muddy and rocky substrate to depth of 100 m. The bogue is a medium sized species, commonly between 100 and 200 mm TL. However, bogue can be reached the largest size via plenty of taking nourishment beneath the sea-cages. In this research, bogue specimens were collected from six operations in gillnet fishery around the sea-cage fish farms in Güllük Bay between January and April 2018. A total of 425 bogues were obtained and the study presents the length-weight relationship of *B. boops*, captured around the sea-cage fish farms in south-eastern Aegean Sea for the first time. The length and weight distribution of all bogue samples ranged from 17 to 32 cm (average:  $25.30\pm0.13$  cm) and from 102 to 587 g (average:  $296.4 \pm 4.29$  g), respectively. The 26 cm mid length range had the highest ratio that is approximately 28.5%. The LWR parameters (*a*, *b*, *r*<sup>2</sup>) were computed as  $0.1271 \pm 0.78$ ,  $2.39 \pm 0.56$  and  $0.813\pm0.57$ , respectively. The *b* value indicates negative allometric growth (p<0.05). Negative allometric growth of bogue around sea-cage farms point out better nourishment. It is known that if *b* < 3, large specimens have changed their body shape to become more elongated or small specimens were in better nutritional condition at the time of sampling and as known, the sea-cage farms provide better nutritional conditions. The results of the study indicated that *B. boops* around the sea-cage fish farms is reaching the bigger size. The bogue as a wild fish around the sea-cages inhabits in a limited area, and it finds plenty of food easily. Thus, fish gets fattening. This phenomenon reflected to length-weight relationship's parameters of the fish, as well.

Keywords: Bogue, length-weight, size, sea-cage farms, Aegean Sea

## **1. INTRODUCTION**

Bogue, *Boops boops* (Linnaeus, 1758) is inshore schooling species found in mid-water above sandy, muddy and rocky substrate to depth of 100 m. Young specimens feed on plankton while adults consume both vegetation and invertebrates. Spawning season is from February to April. It is hermaphroditic species and reaches sexual maturity after one year where most of them are males; in the second year they become females (Golani et al 2006). The species mainly distributed in the eastern Atlantic, from Norway to Angola, common from the Bay of Biscay to Gibraltar and the Mediterranean Sea, including the Black Sea (Froese & Pauly 2019). It also occurs in the western Atlantic in the Gulf of Mexico and the Caribbean Sea (Bauchot & Hureau 1986).

The bogue is a medium sized species, commonly between 100 and 200 mm TL (Bauchot & Hureau 1986) but with a reported maximum size of 400 mm from the Portuguese coast (Gordo 1996). Maximum published weight is 455 g (Froese & Pauly 2019). However, bogue can be reached the largest size via plenty of taking nourishment beneath the sea-cages. The recent study shown that the huge *B. boops* was caught near a sea-cage fish farm by using gillnets, and it was 402 mm and 986 g (Ceyhan et al. 2018). It seems that fishes around the sea-cage fish farms can reach getting the huge size. However, there is gap of population parameters of the wild fish around the sea-cage fish farms.

This study presents the length-weight relationship (LWR) of *Boops boops*, captured around the sea-cage fish farms in south-eastern Aegean Sea for the first time.

## 2. MATERIAL AND METHOD

Bogue specimens were collected from six operations in gillnet fishery around the sea-cage fish farms in Güllük Bay between January and April 2018. A total of 425 bogues were obtained and specimens were measured to the centimetre

(±0.1 cm) fork length (FL) and wet weight (W) was also recorded in grams (±0.1 g). The LWR was estimated based on power regression as  $W = aL^b$ . The logarithmic transformation was performed as  $\log W = \log a + b \log L$  where W is weight (g), L is length (cm), a is the intercept and b is the slope of the linear regressions.

To test for normality and homoscedasticity, data was evaluated using tests of Kolmogorov–Smirnov (Zar 1999) and statistical method of Skewness and Kurtosis (Tabachnick & Fidell 2013). If the datasets passed the normality test, parametric procedures were employed; otherwise, data were transformed using an appropriate transformation process (e.g., log) to meet the underlying assumptions of normality (Zar 1999). The obtained coefficients of linear regressions of length-weight data were analysed with ANOVA (Zar 1999). The degree of relationship between the variables was evaluated by the determination coefficient,  $r^2$ . The null hypothesis of isometric growth (H<sub>0</sub>: b = 3) was tested by t-test, using the statistic: ts=  $(b-3)/S_b$ , where S<sub>b</sub> is the standard error of the slope for  $\alpha = 0.05$  (Sokal & Rohlf 1987).

## **3. RESULTS AND DISCUSSION**

## Results

The length and weight distribution of all bogue samples ranged from 17 to 32 cm (average:  $25.30\pm0.13$  cm) and from 102 to 587 g (average: 296.4 ±4.29 g), respectively (Table 1). The length distribution of all fish was indicated in (Figure 1). The 26 cm mid length range had the highest rate that is approximately 28.5%.

**Table 1.** Range, mean with standard error (s.e.), median and mode of length (FL, cm), and weight (W, g) for *Boops boops*, captured around the sea-cage fish farms in the Aegean Sea

| n=425      | Length (cm)      | Weight (g)        |
|------------|------------------|-------------------|
| Range      | 17-32            | 102-587           |
| Mean ±s.e. | $25.30 \pm 0.13$ | $296.42 \pm 4.29$ |
| Median     | 26               | 286               |
| Mode       | 26               | 260               |



Figure 1. Length-frequency distribution for Boops boops, captured around the sea-cage fish farms in the Aegean Sea

The LWR was shown in (Figure 2). The LWR parameters (*a*, *b*,  $r^2$ ) were computed as  $0.1271 \pm 0.78$ ,  $2.39 \pm 0.56$  and  $0.813\pm0.57$ , respectively. The b value indicates a negative allometric growth (p<0.05).



Figure 2. Linear length-weight relationship for bogue, captured around the sea-cage fish farms in the Aegean Sea

Negative allometric growth of bogue around sea-cage farms point out better nourishment. It is known that if b < 3, small specimens were in better nutritional condition at the time of sampling (Froese 2006), and as known, the sea-cage farms almost provide better nutritional conditions.

## Discussion

Whereas, many of recent LWR studies shown positive allometric growth for *B. boops* along the Aegean Sea (Table 2).

| n    | L  | $\mathbf{L}_{\min}$ | L <sub>max</sub> | $\mathbf{W}_{\min}$ | W <sub>max</sub> | a       | b     | $r^2$ | Area                       |
|------|----|---------------------|------------------|---------------------|------------------|---------|-------|-------|----------------------------|
| 256  | FL | 9.6                 | 24.3             | -                   | -                | 0.00001 | 3.093 | 0.940 | Euboikos Bay, Greece       |
| 1190 | TL | 9.2                 | 27.6             | 7.18                | 281.7            | 0.0035  | 3.419 | 0.948 | Homa Lagoon, Izmir Bay     |
| 68   | TL | 9.4                 | 24.4             | -                   | -                | 0.0197  | 2.827 | 0.906 | SE Aegean Sea              |
| 189  | TL | 10.5                | 22.0             | 11.0                | 91.0             | 0.0045  | 3.241 | 0.958 | Saroz Bay, NE Aegean Sea   |
| 518  | TL | 10.2                | 32.1             | -                   | -                | 0.0048  | 3.258 | 0.949 | Gökçeada, NE Aegean Sea    |
| 32   | TL | 16.5                | 27.0             | -                   | -                | 0.0085  | 3.092 | 0.945 | Gökova Bay, SE Aegean Sea  |
| 39   | TL | 11.3                | 16.7             | -                   | -                | 0.0057  | 3.347 | 0.959 | Uzunada, Izmir Bay         |
| 378  | TL | 11.2                | 23.8             | -                   | -                | 0.005   | 3.240 | 0.968 | E Aegean Sea               |
| 1197 | FL | 10.7                | 23.5             | 14.3                | 252.6            | 0.0127  | 3.033 | 0.920 | Izmir Bay                  |
| 122  | TL | 14.5                | 28.1             | -                   | -                | 0.01467 | 2.877 | 0.910 | Aegean Sea, Greece         |
| 1231 | TL | 9.4                 | 22.1             | 9.28                | 111.6            | 0.00001 | 2.925 | 0.870 | Edremit Bay, NE Aegean Sea |
| 425  | FL | 17.0                | 32.0             | 102                 | 587              | 0.1271  | 2.390 | 0.813 | Güllük Bay, SE Aegean Sea  |

**Table 2.** Length-weight relationships parameters of *Boops boops* in the Aegean Sea (from Akyol et al. 2017; grey row indicates the results of this study)

## **4.CONCLUSION**

The results of the study indicated that *Boops boops* around the sea-cage fish farms is reaching bigger sizes. The bogue as a wild fish around the sea-cages inhabits in a limited area, and it finds plenty of food easily. Thus, fish gets fattening. This phenomenon reflected to LWR parameters of the fish, as well.

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#### REFERENCES

- Akyol O, Demir-Sağlam Y, Ceyhan T (2017). Ege Denizi Balık Türlerinin Boy-Ağırlık İlişkileri Üzerine Bir Derleme. Ege Journal of Fisheries and Aquatic Sciences, 34(2):235-247.
- Bauchot M L & Hureau J C (1986). Sparidae. In: Whitehead P.J.P., Bauchot M.-L., Hureau J.-C., Nielsen J., Tortonese E. (Eds.). Fishes of the North-eastern Atlantic and the Mediterranean. Vol. II. Paris: UNESCO, pp. 883-907.
- Ceyhan T, Ertosluk O, Akyol O & Özgül A (2018). The Maximum Size of Bogue, *Boops boops* (Perciformes: Sparidae) for the Mediterranean. Acta Aquatica Turcica, 14(4): 399-403.
- Froese R (2006). Cube Law, Condition Factor and Weight-Length Relationships: History, Meta-Analysis and Recommendations. Journal of Applied Ichthyology 22, 241-253.
- Froese R & Pauly D (2019). FishBase. World Wide Web Electronic Publication. Retrieved from http:// www.fishbase.org (accessed 01.04.18).
- Golani D, Öztürk B & Başusta N (2006). The Fishes of the Eastern Mediterranean. Turkish Marine Research Foundation, Publication No. 24, Istanbul, Turkey.
- Gordo L S (1996). On the Age and Growth of Bogue, *Boops boops* (L.) from the Portuguese Coast. Fisheries Management and Ecology, 3, 157-164.
- Sokal R & Rohlf F (1987). Introduction to Biostatistics (2<sup>nd</sup> ed.): Freeman Publication, USA.
- Tabachnick B G & Fidell L S (2013). Using Multivariate Statistics: 6th ed.: Pearson Education Inc., USA.

Zar J H (1999). Biostatistical Analysis, 4th edn: Prentice-Hall, USA.



## Monitoring Color Values of Fillet and Skin of Seabass and Seabream pre-treated with Bleaching Agent Solutions

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Abstract: It is common to apply the pre-treatment to improve the quality of the meat and to extend the shelf life of the products in the meat processing industry. The pretreatments also include color development procedures (redness or lightness). One of the chemicals used in the food industry is hydrogen peroxide. It is commonly using for surface sanitizer or color whitener. There are some researches about hydrogen peroxide as a bleaching agent processed seafood. The whitish-enhanced fish fillet is highly appreciated by the consumer, rather than a fish flesh with red dots and/or yellow pectoral surface. This study was monitored seabream and seabass fillets and skins immersed in colloidal silver added hydrogen peroxide and hydrogen peroxide solutions (800 ppm) during initial, after application and one-day storage at 4°C. The experimental designs of groups were determined as SBS, colloidal silver added hydrogen peroxide group for seabass; SBR, colloidal silver added hydrogen peroxide group for seabream; HBS hydrogen peroxide group for seabass; HBR, hydrogen peroxide group for seabream; CBS represents the control group for seabass and CBR control group of fillets and skin for seabream. Tap water was used for control group solution. Computer-based image analysis technology was used for color analysis. The examined color parameters are  $L^*$ ,  $a^*$ ,  $b^*$ , Chroma, and Whiteness. The samples immersed in the solutions for 24 hours at 4°C. After treatment, it was allowed to drain on the clean bench for 5 min., then image analysis was done. According to results; L\* and Whiteness values of fillets were found to be higher than the initial color values of all groups (p < 0.05). However, no statistical difference was found between one-day of storage and after application in L \* values of HBS, CBS, and CBR (p > 0.05). a \*, b \*, and Chroma values were decreased for fillets of all groups after application (p < 0.05). The changes in these values after one-day storage were found significantly different than after application process (p < 0.05). Skin L\*, b\* and Whiteness values of SBR, HBR, and CBR groups were not shown any statistical difference during applications and storage (p < 0.05). According to this result; there is no effect of solutions on the brightness and yellowness value of seabream skin. However, seabass skin color values changes after applications were found statistically different than initial color values (p < 0.05). Although color changes were followed after the applications, data about the superiority of the applications could not be obtained. In order to explain this, more detailed experiment plans on processed seafood are proposed for future study.

Keywords: Color, skin, fillet, hydrogen peroxide, bleaching, fish



## Seasonal Variations of Water and Sediment Quality in Sarıkum Lake of Sinop

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**Abstract:** In the present study, seasonally sampling was performed at Sarıkum Lake at 4 locations in the year of 2011. The lake is known as eutrophic. The environmental pollution parameters were analysed at the water samples and surface sediment collected from Sarıkum Lake of Sinop Provincial. On the water samples, dissolved oxygen values were found between 3.29 and 6.13 mg  $1^{-1}$ , and surface water temperatures ranged between 6.5 and 27.2°C. Salinity values ranged between 1.0 PSU and 8.9 PSU, pH values from 6.65 to 7.94 and TSS values were found between 211 and 443 mg  $1^{-1}$ . NO<sub>3</sub>-N values were found as between 6.95 and 28.4 mg  $1^{-1}$ . Silicate values ranged from 0.088 to 0.19 (mg  $1^{-1}$ ). On the sediment samples, grain sizes varying between 77-95% were found to be between 0.100 and 0.125 mm. Organic content varied in the ranges of 1.4 to 5.61 %. Oxidation-reduction (Eh) values are generally close to zero or negative. The values of stations 3 and 4 are negatively larger. The reason for this is that the sediment structure sampled at this station is much muddy and contains more organic matter. In addition, the colour is darker the oxygen here is less and can be seen as a sign of the presence of iron sulphides. Eh values are close to zero as stations 1 and 2 and the colour of the sediment is slightly light colour, it may be due to the presence of ferric iron. Results showed that, there was an organic pollution at the sediment layer of some sampling stations and water according to some physico-chemical parameters moderately polluted water, or much polluted water class.

Keywords: Sarıkum Lake, eutrophic, organic content, oxidation-reduction, sediment, water

## **1. INTRODUCTION**

The Sarıkum Lake is 21 km away from Sinop city, which its 10 km long sandy beach. In 1987, Sarikum area has been officially registered as Natural Reserve Area by the abrogated Ministry of Forestry. In 1991, Sarikum Lake and its environment were registered as Nature Protection Area (Anonymous, 1997; Yılmaz, 2005). Sarıkum Lake which has 785 hectares complex ecosystem of sea, sand, freshwater lake, wetland, peatland, marshland and forest and is one of our eutrophic wetlands in terms of biological production and is rich in plankton, fish and bottom organisms (Sıvacı et al., 2008; Yardım et al., 2008; Yardım, 2017). The area of Sarıkum is also a picnic place. Sarıkum is an alluvial set lake. The deepest place is around 2 m.

The purpose of this study is to determine the pollution status by measuring the water and sediment quality of Sarıkum Lake.

## 2. MATERIAL AND METHOD

In the present study, seasonally sampling was performed at Sarıkum Lake at 4 locations in the year of 2011 (Figure 1).



Figure 1. Study area

Water samples were taken from surface of the lake (less than 1 m). Physico-chemical properties of wastewater were determined according to the standards of the American Public Health Association (APHA 2005). Temperature, pH, salinity and dissolved oxygen of surface water were determined at the collection sites using a HORIBA Multi-parameter meter for pH/temperature/ORP/dissolved oxygen. Nitrate and silicate were determined using a UV visible spectrophotometer (Thermo Spectronic, He $\lambda$ ios). Total suspended solids (TSS), unfiltered water samples were filtered with tared filter paper and 0.45 µm Millipore filter system. After filtration, the filter paper was dried in the oven at 105°C for one hour, cool dish in desiccator to balance temperature and weighed. The suspended solids were determined by the weight of the filter papers and the weight differences after filtration and drying.

Total organic contents of sediment samples were dried at 105°C for a day. Five grams of dried sample were then treated with hydrochloric acid vapour overnight in a desiccating jar to convert any calcium carbonates to chlorides. Samples were then placed in a muffle furnace at 1000°C for an hour. The loss ignition was taken as the organic carbon content of the sediment (Buchanan, 1984). Sediment types were determined according to Stove's scale (Kocataş, 2012). Approximately 500 g sediment was weighed and dried in oven at 105°C for 2 days. The dried sediment samples were passed through mesh sieves. Samples of sediment on each sieve are weighed and particle sizes are classified according to weight values % (Wentworth, 1922). Oxidation-reduction (Eh) values were measured with Portable Redox-meter.

## 3. RESULTS AND DISCUSSION

## Results

On the water samples, dissolved oxygen values were found between 3.29 and 6.13 mg  $l^{-1}$  (Figure 2), and surface water temperatures ranged between 6.5 and 27.2°C (Figure 3). Salinity values ranged between 1.0 PSU and 8.9 PSU (Figure 4), pH values from 6.65 to 7.94 (Figure 5) and TSS values were found between 211 and 443 mg  $l^{-1}$  (Figure 6). NO<sub>3</sub>-N values were found as between 6.95 and 28.4 mg  $l^{-1}$  (Figure 7). Silicate values ranged from 0.088 to 0.19 mg  $l^{-1}$  (Figure 8).



Figure 2. Dissolved oxygen values (mg l<sup>-1</sup>) in water from Sarıkum Lake in 2011.



Figure 3. Temperature values (°C) in water from Sarıkum Lake in 2011.



Figure 4. Salinity values (PSU) in water from Sarıkum Lake in 2011.



Figure 5. pH values in water from Sarıkum Lake in 2011.



Figure 6. Total suspended solid values (mg l<sup>-1</sup>) in water from Sarıkum Lake in 2011.



**Figure 7.** NO<sub>3</sub>-N values (mg l<sup>-1</sup>) in water from Sarıkum Lake in 2011.



**Figure 8.** Silicate values (mg l<sup>-1</sup>) in water from Sarıkum Lake in 2011.

On the sediment samples, grain sizes varying between 77-95% were found to be between 0.100 and 0.125 mm (Table 1). Organic content varied in the ranges of 1.4 to 5.61 % (Figure 9). Oxidation-reduction (Eh) values are generally close to zero or negative (Table 2).

| Table 1 | . Mean | sediment | particle | sizes | % |
|---------|--------|----------|----------|-------|---|
|---------|--------|----------|----------|-------|---|

| Sadimant tuna    | Sadimant diamatan (mm) | Particle size (%) of stations |     |     |     |
|------------------|------------------------|-------------------------------|-----|-----|-----|
| Sediment type    | Seument diameter (mm)  | 1.                            | 2.  | 3.  | 4.  |
| Very fine sand   | 1/16-1/10              | 0.5                           | 3.9 | 6.5 | 3.1 |
| Fine sand        | 1/10-1/8               | 95                            | 82  | 77  | 80  |
| Medium sand      | 1/8-1/2                | 1.2                           | 2.3 | 2.8 | 3.4 |
| Coarse sand      | 1/2-1                  | 1.8                           | 8.4 | 9.5 | 9.5 |
| Very coarse sand | 1–2                    | 1.5                           | 3.4 | 4.2 | 3.1 |



Figure 9. Average organic content (%) in sediments

Table 2. Redox values (Eh, Mv) of sediment in Sarıkum Lake.

| Second  |       | Stations |      |      |  |  |  |
|---------|-------|----------|------|------|--|--|--|
| Seasons | 1.    | 2.       | 3.   | 4.   |  |  |  |
| Spring  | +0.09 | +0.13    | -7   | -12  |  |  |  |
| Summer  | +0.11 | +0.08    | -4   | -6   |  |  |  |
| Autumn  | +0.05 | +0.68    | -15  | -37  |  |  |  |
| Winter  | -44   | -157     | -187 | -315 |  |  |  |

## Discussion

Water parameters were significantly different at seasonally. Organic waste and other nutrient inputs from sewage, agricultural and urban runoff can decrease DO levels. In this study, the lowest DO level of station 4 was 3.29 mg l-1. It is concluded that this value shows class 3 at Turkish Environmental Regulation (2004), Quality Criteria of Inland Water Sources according to be Classes. The highest temperature (27.2°C) was measured at station 4 in summer. In general the water temperatures of Sarıkum Lakes were within Water Pollution Control Regulations of 2872 the Official Gazette Environment Law for water. The pH values in all stations were within the standards set by the Turkish Environmental Regulation (2004), General Quality Criteria of Marine Water for pH levels. The observed TSS values were quite above the standards maximum 30 set by the Turkish Environmental Regulation (2004), General Quality Criteria of Marine Water. High TSS values in natural waters effect decreases light transmittance. NO3-N values in the water of Sarıkum Lake exceeded (stations 3 and 4) the standard limit set by Turkish Environmental Regulations (2004). Silicate values were low.

The values of redox values at stations 3 and 4 are negatively larger. The reason for this is that the sediment structure sampled at this station is much muddy and contains more organic matter. In addition, the colour is darker the oxygen here is less and can be seen as a sign of the presence of iron sulphides. Eh values are close to zero as stations 1 and 2 and the colour of the sediment is slightly light colour, it may be due to the presence of ferric iron.

#### 4. CONCLUSION

Results of the present study showed that there was an organic pollution at the sediment layer of some sampling stations in Sarıkum Lake and water according to some physico-chemical parameters moderately polluted water, or much polluted water class. The continuation of monitoring studies at the Sarıkum Lake is important to provide data to decision-makers and to observe the current pollution situation.

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#### REFERENCES

- Anonymous (1997). National Environmental Action Plan. Land Use and Management of Coastal Areas. State Planning Organization, 94 p. (in Turkish).
- APHA (2005). Standard methods for the examination of water and wastewater (Centennial Edition ed.): American Public Health Association.
- Buchanan JB (1984). Sediment analysis. In: N.A. Holme and A.D. McIntyre (Eds.), Methods for the Study of Marine Benthos. Blackwell Sci. Publ., pp. 41-65.
- Kocataş A (2012). Genel oseanoloji. Deniz Bilimlerine Giriş. 10. Baskı. Dora Yayıncılık. 357 pp.
- Sivaci ER, Yardim Ö, Gönülol A, Bat L, Gümüs F (2008). Sarıkum (Sinop-Türkiye) lagününün bentik algleri (Benthic algae of Sarıkum (Sinop-Turkey) lagoon) Journal of FisheriesSciences.com, DOI: 10.3153/jfscom.2008022, 2(4): 592-600.

Turkish Environmental Regulations Water Pollution Control Regulation (2004-2005). http://www.cevreorman.gov.tr/yasa/yonetmelik

Wentworth CK (1922). A Scale of Grade and Class Terms for Clastic Sediments, Journal of Geology, 30 (5): 377-392.

- Yardim Ö, Sendogan E, Bat L, Sezgin M, Çulha M (2008). Sarıkum Gölü (Sinop) makrobentik Mollusca ve Crustacea faunası (Macrobenthic Mollusca and Crustacea fauna of Lake Sarıkum, Sinop, Turkey). E.Ü. Su Ürünleri Dergisi (E.U. Journal of Fisheries & Aquatic Sciences), 25 (4): 301–309.
- Yardim Ö (2017). Study on Fish Species of Sarikum Lake in Sinop Province in the Black Sea Region. Journal of Anatolian Environmental&Animal Sciences, 2 (3): 72-74.

Yilmaz C (2005). Ecosystem of Sarıkum Lake (Sinop). O.M.Ü. Fen Edeb. Fak., Turkey Symposium on Quantum 219-223. (in Turkish).



## Immune Responses of Carp (*Cyprinus carpio*), Fed with Garland Thorn (*Paliurus spina christi*) Aqueous Methanolic Extract

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**Abstract:** In this study, immune responses of *Paliurus spina christi* aqueous methanolic extract as a feed additive on common carp (*Cyprinus carpio*) were investigated. With this purpose, fish were feed diet containing with 4 different doses (0 (Control), 1, 2 and 3 g kg<sup>-1</sup>) of the plant daily ad libitum during 45 days. To determine non specific immune responses Every fifteenth day of the study, respiratory burst activity, lysozyme and myeloperoxidase activity were determined. Respiratory burst activity was increased significantly on experimental groups compared to control (P<0.05). Lysozyme activity was significantly decreased on all experimental groups except 30 days of the study compared to control (P<0.05). Myeloperoxidase activity was significantly increased on all experimental groups especially on 2 g kg<sup>-1</sup> group (P<0.05). There is no differences observed on groups' growth performance compared to control (P>0.05). According to these results, *Paliurus spina christi* could be used as an immunostimulant for fish for 30 days long.

Keywords: Cyprinus carpio, Paliurus spina christi, aqueous methanolic extract, non specific immune response,



# Growth Performance of Carp (*Cyprinus carpio*), Fed with Garland Thorn (*Paliurus spina christi*)

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**Abstract:** In the present study, growth performance of *Paliurus spina christi* aqueous methanolic extract on common carp (*Cyprinus carpio*)  $(1.43\pm0.1)$  were determined. With this aim, fish were feed the diet (4 different doses; 0 (Control), 1 %, 2% and 3%) ad-libitum during 45 days. At the end of the study, the highest final weight was observed on 3% group. However, this increase was not significant. Lowest FCR value was determined on 2% group compared to other groups (P<0.05). No differences observed on groups SGR. These results suggest that despite no differences were observed final weigh of the fish, decreasing on FCR value on 2% group, could be used as a feed additive in common carp.

Keywords: Cyprinus carpio, Paliurus spina christi, aqueous methanolic extract, growth performance.



## Biochemical Composition of *Neochloris aquatic* Cultured in Different Media

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**Abstract:** In this study, biochemical composition of star algae (*Neochloris aquatic*) cultured in different culture media (Modified Bold's Basic Culture Media (B1NV), Bold's Basic Culture Media (BBM), Blue green culture media (BG11) and new culture media (OM)) was determined. With this purpose, at the end of the study, star algae samples were collected from culture media and amino acid, fatty acid compaosition were determined. Proximate analyses were also performed. Glutamic acid and histidine was found highest level in the algae. The most abundant fatty acids was found rate as linoleic acid (19.40%) and linolenic acid (18.96%) respectively. Protein and fat value of the star algae was elevated as 49.27% and 7.62% respectively.

Keywords: Neochloris aquatic, amino acid composition, fatty acid composition, proximate analyses



## Culture of Neochloris aquatic in Different Culture Media

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**Abstract:** In this study, culture possibilities of *Neochloris aquatic* microalgae that uncultured previously, was investigated in some classical culture media. With this aim, *Neochloris aquatic* was inoculated into different culture media such as Modified Bold's Basic Culture Media (B1NV), Bold's Basic Culture Media (BBM), Blue green culture media (BG11) and new culture media (OM) in similar conditions. At the end of the study, it was observed that the cell density was reached to 2.63x10<sup>6</sup> cell /ml in B1NV, 2.51x10<sup>6</sup> cell /ml in BBM and 0.8x10<sup>6</sup> cell/ml in OM groups. In BG11, alternative culture media that used instead of other media, cell density was reached with its highest value up to 2.95x10<sup>6</sup> cell/ml. In BG11 culture media, dry weight of the studied sample was 0.684 gr/l. The best improvement in the culture was observed at the 26<sup>th</sup> day of the study. After 26<sup>th</sup> day of the study, the culture was entered its constant phase and the after this day, the culture entered its dead phase and the cell count started to decrease. All this data showed that *Neochloris aquatica* in BG11 media was reached its aimed cell density that is expected in biotechnological studies.

Keywords: Neochloris aquatica, microalgae, culture media, biotechnology



## A Review of the Lethal Concentrations of Pesticides in Some Freshwater Fish Species

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**Abstract:** Several environmental pollutants especially the pesticides are used extensively in agricultural areas. Furthermore, the pesticides are used to control the insects, aquatic weeds, plants, and fungi and these pollutants contaminates the aquatic environment and aquatic animals. Unfortunately, in spite of its advantages, pesticides have major disadvantages as it threatening the long-term survival of major ecosystems by disruption of ecological relationships between organisms and loss of biodiversity. Numerous studies have conducted and found them to be highly toxic not only to fish but also to the other organisms. This review summarizes the lethal concentration of pesticides on aquatic animals. In this study, peer reviewed scientific articles, theses and dissertations as well as recent books were used as a source. Literature has shown that insecticide and herbicide are among the most frequently reported pesticides. Especially, the acute toxicity of carbaryl, propineb, benomyl, methiocarb, hymexazol, deltamethrin, carbosulfan, captan, endosulfan, malathion and maneb to freshwater fish species such as rainbow trout, common carp, zebra fish was evaluated in 96-h toxicity tests under static conditions in many studies. For example, according to these studies concentrations that killed fifty per cent of the fish in the 96 h toxicity test were ranged from 0.34 to 8.5 mg/l for carbaryl; from 0.3 to 28.0 mg/l for methiocarb; from 0.031 to 12.5 mg/l for carbosulfan; from 0.26 to 0.38mg/L for captan; from 1.6 to 1.75 mg/L for endosulfan. The obtained data could be useful in the environmental risk assessment of the pesticides in freshwater fish species.

Keywords: Pesticides, toxicity, freshwater fish, LC50



## Closed Areas for Fishing in the Iskenderun Bay and Illegal Fishing Activities

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**Abstract:** The aim of the study is to determine closed areas for fishing (CAF) from Iskenderun Bay and to evaluate illegal fishing activities in these areas. For this purpose, in this study, closed areas for fishing in Iskenderun Bay such as port, pier, shelter, and military areas were classified and determined. Then, the total marine area in these areas was measured as km<sup>2</sup> and closed area was determined according to these measures. Since these areas, which have closed to fisheries, are a natural marine protected area (MPA), an evaluation was made about sustainable fisheries management for the Iskenderun Bay fish stocks. Therefore, in this study, general information about possible fish species in these areas was given. In this direction, study presented on the examples of the socio-economic problems of the fishery activity that started for recreational purposes in terms of the fishery management for the region. According to results, closed area fishing in Iskenderun Bay was determined as 64,21 km<sup>2</sup>. With the study, evaluation was made with the aim to raise awareness about the problems of fishing activities which have thought to be simple and recreational purposes in these areas. Hand line, traps and different fishing nets have used in these areas. Although the fishing activities in this area have prohibited, this illegal fishing activity has under controlled by institutions that have control mechanism. Because the absence of entry and exit of non-staff individuals and institutions to these areas often limits the work of fisheries inspectors.

Keywords: Closed area, illegal fishing activities, Iskenderun Bay,

## **1. INTRODUCTION**

Iskenderun Bay is an important trade point for Turkey in the Eastern Mediterranean. Iskenderun Bay, located at the southern border of Turkey, forms the eastern boundary line in marine areas, so there are marine areas used for military purposes. (Ateş, 2014). The share of the total cargo handling of Iskenderun Bay is 9.34 % in Turkey. (Gülmez et al., 2018). In addition, oil transmission lines have a very large capacity in the region. For the power plants established on the shore, it requires marine structures both for the use of cooling water from the sea and for the coal transportation. Likewise, iron and steel industry has makes the majority of the raw materials and fuel transport from sea way in the region. Although it is made for fishing purposes, there are 6 fishing ports in the bay that allow all other sea vehicles.

In the special marine areas listed above, security measures have applied due to the security level, bonded area, maritime safety, commercial and military sensitivity. Access to these areas from both land and sea is limited and under control. According to Ministry of Agriculture and Forestry regulations, any commercial and recreational fishing activities are banned by at a certain distance in these areas. This prohibition is very meaningful because the maritime activities in these areas are not disturbed by small coastal fishing vessels and these fishermen are not harmed by these large maritime activities.

Increasing numbers and capacities in these areas day by day means the reduction of catchment areas for fishery. Smallscale fishery industry has adversely affected by this situation. In fact, these fishermen have punished for illegal fishing activities in these areas. In this research, an evaluation was made with the aim to raise awareness about the problems of fishing activities which are thought to be simple and recreational purposes in these areas. Hand line, traps and different fishing nets have used in these areas. Although the fishing activities in this area are prohibited, this illegal fishing activity has under controlled by institutions that have control mechanism. Because the absence of entry and exit of non-staff individuals and institutions to these areas often limits the work of fisheries inspectors (Melli et al. 2016).

### 2. MATERIAL AND METHOD

In this study, closed areas for fishing in Iskenderun Bay such as port, pier, shelter, and military areas were determined. Then, the total marine area in these areas was measured as km2 and closed area was determined according to these measures. Since these areas, which are closed to fisheries, are a marine protected area, an evaluation was made about sustainable fisheries management for the Iskenderun Bay fish stocks. Therefore, in this study, general information about possible fish species in these areas was given. In this direction, study presented on the examples of the socio-economic problems of the fishery activity that started for recreational purposes in terms of the fishery management for the region (Browman, et al. 2018).

## 3. RESULTS AND DISCUSSION

In this study, due to different maritime activities, the area width of 12 coastal structures which have closed area for fishing was calculated (Figure 1 and Table 1). It is clear that the closure of such an area to fishing activities will create a natural marine protected area (Gill et al 2017). This area, covering a total of 64.21 km<sup>2</sup>, will be a natural refuge for many coastal fish species. It has clear that the fish stocks in this area will have a positive effect on the whole Iskenderun Bay. However, illegal fishing activities in these areas turn the natural positive situation into negative in many respects.



Figure 1. Closed Areas for fishing of coastal structures in Iskenderun Bay

Fishing activities in these areas have a much more negative impact on stocks already under pressure from fisheries. In addition, lighting and safety of these coastal structures and the use of light, fish management is in question. Naturally, this management increases the negative impact of fishing activities on stocks.

| Fable 1. Closed Areas for fishing, use of p | urpose and wide areas of coasta | l structures in Iskenderun Bay |
|---|---------------------------------|--------------------------------|
|---|---------------------------------|--------------------------------|

| No | Region     | Purpose  | Area (km <sup>2</sup> ) |
|----|------------|--|-------------------------|
| 1  | Konacık    | Fishing Port                                       | 0.3                     |
| 2  | Arsuz      | Military Araea and River Outfall                   | 0.83                    |
| 3  | Madenli    | Fishing Port                                       | 0.52                    |
| 4  | İskenderun | Military Araea                                     | 0.32                    |
| 5  | İskenderun | Fishing Port. Military Port. Limak Port. Fuel Pier | 3.43                    |
| 6  | Sarıseki   | Seven trade and one military port                  | 7.38                    |
| 7  | Payas      | ISDEMIR Port and Security area                     | 4.32                    |
| 8  | Payas      | Payas MMK Port                                     | 1.47                    |
| 9  | Dörtyol    | Fishing Port                                       | 0.75                    |
| 10 | Dörtyol    | Two oil and one gas transport ports                | 4.75                    |
| 11 | Gölovası   | Six trade. two oil and one military ports          | 39.8                    |
| 12 | Yumurtalık | Fishing Port                                       | 0.44                    |
| TO | ГAL        |  | 64.21                   |

Commercialization, which is one of the most important problems in recreational fishing, is also present in our region (Demirci, & Arslantaş2018). Due to the security that has closed to fisheries, the fish caught in these areas cannot be sold and the fisheries inspections cannot be performed. Since this sale takes place in a narrow social environment like Iskenderun, commercial fishermen can be informed. Because of their relatively individual size and rare coastal species in commercial fisheries, these caught fish become market-high. This commercialization degenerates the real fishermen and creates administrative social problems.

#### REFERENCES

- Ateş, A. (2014). Türkiye'de liman özelleştirmeleri Iskenderun liman örneği. Mustafa Kemal Üniversitesi Sosyal Bilimler Enstitüsü Dergisi, 11(25), 427-457.
- Browman, H. I., Cooke, S. J., Cowx, I. G., Derbyshire, S. W., Kasumyan, A., Key, B., ... & Watson, C. A. (2018). Welfare of aquatic animals: where things are, where they are going, and what it means for research, aquaculture, recreational angling, and commercial fishing. ICES Journal of Marine Science, 76(1), 82-92.
- Demirci, S., & Arslantaş, E. (2018). Economic Potential and Environmental Effects of Recreational Fishing Activity in Coast of Iskenderun Bay. Fresenius Environmental Bulletin, 27, 9352-9356.
- Gill, D. A., Mascia, M. B., Ahmadia, G. N., Glew, L., Lester, S. E., Barnes, M., ... & Holst, S. (2017). Capacity shortfalls hinder the performance of marine protected areas globally. Nature, 543(7647), 665.
- Gülmez, S, Esmer, S, Ateş, A. (2018). An Analysis of Seaborne Trade Through Iskenderun Bay. Dokuz Eylül Üniversitesi Denizcilik Fakültesi Dergisi, 10 (1), 83-108. DOI: 10.18613/deudfd.428168
- Mellin, C., Aaron MacNeil, M., Cheal, A. J., Emslie, M. J., & Julian Caley, M. (2016). Marine protected areas increase resilience among coral reef communities. Ecology letters, 19(6), 629-637.



## Efficacy of Fenugreek (*Trigonella foenum-graecum*) Seed Aqueous Methanolic Extract Against *Yersinia ruckeri* and *Aeromonas hydrophila*

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**Abstract:** In the present study, antibacterial activity of aqueous methanolic extract of fenugreek seed (*Trigonella foenum-graecum*) was tested against the pathogenic bacteria in fish; *Yersinia ruckeri* and *Aeromonas hydrophila*. With this purpose, 150 µl of fenugreek extract and 3 µl bacterial strains were placed into 96-well plates and 8 different plant concentration (25600 µg/ml, 12800 µg/ml, 6400 µg/ml, 3200 µg/ml, 1600 µg/ml, 800 µg/ml, 400 µg/ml, 200 µg/ml, 100 µg/ml, 50 µg/ml, 25 µg/ml, 24 µg/ml, 12 µg/ml, 6 µg/ml, 3 µg/ml, 1.5 µg/ml, 0.75 µg/ml and 0.375 µg/ml) were mixed with the culture in triplicate. The results of this study showed that aqueous methanolic extracts of fenugreek seed was found non effective against *Aeromanas hydrophila* at any dose. However, 25 µg/ml concentration of the plant extract showed an inhibition against *Yersinia ruckeri*. Considering overall performance, fenugreek showed bactericidal effect against *Yersinia ruckeri*.

Keywords: Aeromanas hydrophila, Yersinia ruckeri, microdilution method, fenugreek, medicinal plant, fish pathogens.

## **1. INTRODUCTION**

Fish farming has been going on for about 4000 years and it is the biggest branch of agriculture (Beveridge and Little, 2002). Global production of farmed fish and shellfish has increased more than 100% in the past 15 years. Global aquaculture production still adds to world fish supplies (Naylor et. al., 2000).

Food requirement is the main problem for man due to rapidly increasing population and subsequent need for food. Therefore, aquaculture has become very important and development of technology led to a significant increase in fish production. Nowadays, fish deaths due to diseases are important in fish production. There are two common methods used to prevent diseases. Vaccination is the most common method for prevention of diseases. On the other hand, the use of antibiotics is also very common. Because of their many disadvantages, it is not possible to control fish diseases only with vaccines and antibiotics sustainably. Vaccination is a method that requires both expensive and excess labor. Furthermore, side effects of antibiotics are major problems especially, the resistance of bacteria to antibiotics (Kim et.al., 2017). The occurrence of mutagenic bacteria together with the damage caused to the environment due to antibiotic waste is a major issue.

There are many immunostimulants used in different ways. The use of medicinal plants in fish diseases has recently become widespread. Fenugreek is a very old spice and has been highly valued in ancient times. Fenugreek seeds have been used as a spice and also for pharmaceutical production.

*Aeromonas hydrophila* is found as a secondary pathogen in water and fish in normal flora. However, it causes bacterial hemorrhagic septicemia in stressed fish (Erer, H., 1983). In diseased fish; skin, fin and muscle bleeding, as well as ulcers (Mancini, 1997), loss of fin, erosions around the mouth, periorbital edema, exophthalmos, ascites and systemic bleeding can be seen (Erer, 2002).

*Yersinia ruckeri* is a gram-negative bacterium. It is the causative agent of yersiniosis or enteric redmouth disease which leads to significant economic losses in salmonid aquaculture. Infection may result in haemorrhages on the body surface and in the internal organs. Despite the importance of the disease, little is known about the pathogenesis that prevents the development of preventive measures to overcome this bacterial agent effectively (Tobback et. al., 2007)

It is considered that the use of some medicinal plant species as a feed additive instead of an antibiotic is a good alternative to control both bacterial fish diseases and to strengthen the immune system of animals and promote growth performance (Hermann et.al., 2003). The use of medicinal plants has become more important in terms of environment, human health and economics.

## 2. MATERIAL AND METHODS

Microdilution method, one of the Minimum Inhibition Concentration tests, was used in the study (CLSI, 2008). Pure cultures of *Aeromonas hydrophila* and *Yersinia ruckeri* bacteria were obtained from the laboratory of Faculty of Fisheries of Kastamonu University. It was aimed to stop the growth of Aeromonas hydrophila and Yersinia ruckeri bacteria by using methanolic extracts of fenugreek seeds at certain concentrations. For this purpose, firstly 50 g. milled fenugreek seeds, 400 ml. methanol and 600 ml. distilled water were mixed. The mixture was kept in a shaded environment for 48 hours. Afterwards, the methanolic extract of fenugreek seeds was obtained by evaporator. Then, bacteria were prepared for microdilution method. 96-well plates and 150 µl bacterial strains per plate were used. Different concentrations of fenugreek (25600 µg/ml, 12800 µg/ml, 6400 µg/ml, 3200 µg/ml, 1600 µg/ml, 800 µg/ml, 400 µg/ml, 200 µg/ml, 100 µg/ml, 50 µg/ml, 25 µg/ml, 24 µg/ml, 12 µg/ml, 6 µg/ml, 3 µg/ml, 1.5 µg/ml, 0.75 µg/ml and 0.375 µg/ml) and 3 µl of bacterium for each well were added to the prepared wells. In addition, only the medium (broth) and 3 µl of the bacteria were added for the viability test of the bacteria in different wells (positive control) and only a mixture of nutrient and methanolic extract was added to see if the medium was contaminating during work (negative control). Each bacterium and concentrations were studied in triplicate. The lid of the plate was closed and wrapped around with parafilm. The plate was then placed in the incubator. It was kept at 25 °C for 48 hours.

#### 3. RESULTS AND DISCUSSION

The antimicrobial properties of *Trigonella foenum-graecum* was evaluated by Bahi et al. (2017), where antimicrobial activities were tested on *Bacillus licheniformis*, *Lactobacillus plantarum* and *Bacillus subtilis* strains in gilthead seabream were determined. The results showed a significant increases in the immune parameters, mainly in fish fed only fenugreek or fenugreek combined with *B. subtilis*. Furthermore, real time pcr results revealed that dietary supplementation significantly enhances the expression of immune-associated genes in the head-kidney, particularly Igm (Immunoglobulin M) gene expression.

The results of present study showed that aqueous methanolic extract of fenugreek seeds did not effect the growth of *Aeromanas hydrophila* at any concentration. However, *Yersinia ruckeri* has been successfully inhibited by using fenugreek aqueous methanolic extract at a concentration of 25 µg/ml.

#### 4. CONCLUSION

According to the results of microdilution method analysis, fenugreek seeds aqueous methanolic extract can be used at a concentration of 25  $\mu$ g/ml in vitro as bactericidal against *Yersinia ruckeri*. This study will be guide for future *in vivo* fish studies.

#### REFERENCES

- Bahi, A., Guardiola, F. A., Messina, C., Mahdhi, A., Cerezuela, R., Santulli, A., ... & Esteban, M. A. (2017). Effects of dietary administration of fenugreek seeds, alone or in combination with probiotics, on growth performance parameters, humoral immune response and gene expression of gilthead seabream (Sparus aurata L.). Fish & shellfish immunology, 60, 50-58.
- Beveridge, M. C., & Little, D. C. (2002). The history of aquaculture in traditional societies. *Ecological aquaculture. The evolution of the Blue Revolution*, 3-29.
- Clinical Laboratory Standards Institute (CLSI) (2008) Antimicrobial Disk and Dilution Susceptibility Tests for Bacteria Isolated from Animals. Approved Standard, 3rd edn. CLSI document M31-A3. Wayne, PA, USA: CLSI.
- Erer, H., 1983. Aynalı sazanlarda (Cyprinus carpio L.) deneysel Aeromonas hydrophila enfeksiyonunda (Bakteriyel hemorajik septisemi) oluşan patolojik bulguların incelenmesi. *A.Ü. Vet. Fak. Derg.*, 30 (4), 674-690.
- Erer, H., 2002. Balık Hastalıkları. 2. baskı, Selçuk Üniv. Basimevi, Konya.
- Hermann, J. R., Honeyman, M. S., Zimmerman, J. J., Thacker, B. J., Holden, P. J., & Chang, C. C. (2003). Effect of dietary Echinacea purpurea on viremia and performance in porcine reproductive and respiratory syndrome virus-infected nursery pigs. *Journal* of Animal Science, 81(9), 2139-2144.

- Kim, H. Y., Lee, I. S., & Oh, J. E. (2017). Human and veterinary pharmaceuticals in the marine environment including fish farms in Korea. *Science of the Total Environment*, 579, 940-949.
- Mancini, M. (1997). Rol de la bacteria Aeromonas hydrophila en ictiopatología. Rev. Medicina Veterinaria, 78(6), 380-387.
- Naylor, R. L., Goldburg, R. J., Primavera, J. H., Kautsky, N., Beveridge, M. C., Clay, J., ... & Troell, M. (2000). Effect of aquaculture on world fish supplies. *Nature*, 405(6790), 1017.
- Tobback, E., Decostere, A., Hermans, K., Haesebrouck, F., & Chiers, K. (2007). Yersinia ruckeri infections in salmonid fish. *Journal* of fish diseases, 30(5), 257-268.



## Distortions in Eastern Mediterranean Deep-Sea Fishery Due to Geopolitical Instability and Fishery Pressures

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**Abstract:** The Eastern Mediterranean has been a major dispute for many years. Moreover, the Arab Spring and the Syrian War increased the uncertainties in the region. The economic limits arising from oil and natural gas reserves in the region have not understood between the coastal states. The lack of management created by these disputes negatively affects fish stocks in the Eastern Mediterranean. The fishing vessels of some Mediterranean countries, especially demersal sources, benefit from this negative situation. Turkey principles of the United Nations Law of the Sea Convention does suffer a loss of rights in particular in the direction of demersal resources. In addition to this loss of rights, it is essential to control the exploitation of these resources in terms of transferring them to future generations.

Keywords: Shrimp trawl fishery, grid selectivity, flow channel

## **1. INTRODUCTION**

The United Nations Convention on the Law of the Sea (UNCLOS) has given the coastal States rights and responsibilities in the seas as Exclusive Economic Zone (EEZ) in terms of fish stocks. The EEZs are defined as marine areas extending up to 200 miles, and for areas below this distance, the boundaries between the states should be determined based on the coastal line. It is very difficult to determine these borders in the Eastern Mediterranean because there are disagreements and even wars between many states in the region and there has been international power show in recent years (Korman, 2010; Wakefield 2016).

There are serious international disputes in the eastern Mediterranean waters. In fact, the inability to solve the two-state problem on the island of Cyprus limited the economic use and control of these waters. With the Arab Spring and the Syrian War in recent years, the problems gained a different dimension (Rygiel, Baban, & Ilcan, 2016).

Although the Mediterranean is an terrestrial sea, it is an important military and security area since the geopolitical historical process. In addition to the coastal states in the region, Britain and the United States were still in military presence in the region, and finally, with the Syrian war, Russia has now begun to show military presence in the Eastern Mediterranean.

The discovery of oil and natural gas resources in the region reveals conflicts in terms of continental shelf and coastal states (Prontera, 2017). Cyprus as the sole ruler of the island state in the last decade, with the exception of Turkey are making bilateral agreements in this area. Turkey sees the process does not recognize this unjust, unfair, and void. This dispute, which mostly concerns continental shelf, also hinders economic zone agreements (Winrow, 2016). In this context, a number of problems in the economic zone and varid agreements and limits attached to it because it is not certain in the Eastern Mediterranean in areas outside the territorial waters of Turkey's fisheries management and control issues. In this article, problems are analyzed and evaluations are made on the basis of scientific requirements and national interests.

## 2. MATERIAL AND METHOD

Fishing activities on the map shown in the appendix were obtained by using Marine Traffic, Global Fishing Watch application. In this data, assessments are made on the basis of fishing area, fishing period, fishing method and the country to which the ship belongs. The ports of departure of each ship were also assessed. In these evaluations, fishing vessel trips were transferred to excel environment and evaluated (Merten, et al., 2016).


Figure 1. Eastern Mediterranean Fishing Vessel Movements (White dots; Fishing Vessel, Green Line; EEZ drawn by Google)

Fishing activities on the map shown in the appendix were obtained by using Marine Traffic, Global Fishing Watch application. In this data, assessments are made on the basis of fishing area, fishing period, fishing method and the country to which the ship belongs. The ports of departure of each ship were also assessed. In these evaluations, fishing vessel trips were transferred to excel environment and evaluated.

#### **3. RESULTS AND DISCUSSION**

To this day, an important part of the fishing activity in the region, Turkey was carried out by fishing vessels. To be close to them when considering the scope of the lie Turkey and the Turkish fishing fleet this is a natural result. Although trawling and purse seine fishing is the main priority in terms of fishing method, there are longline and bottom trawl nets and fishing activities. The purse seine fishing fleet is all Turkish-flagged vessels, but the ones that have Moroccan and Algerian flags, but actually belong to the Turks, have been found on fishing vessels. The purse-fishing fleet is generally large pelajik Bluefin Tuna targeted fishing boats. However, IES data are not available on ships that generally belong to Syria and North African countries. When the trawler fleet has examined, Turkish vessels have only in this region during the summer months, while Italian flagged vessels have been operating in the region for many years by taking advantage of this lack of management. This Italian ships from Cyprus by Turkey Motion fishing areas are very close to making territorial waters. This is connected Umeda Turkey should improve the management and control activities in the region in general. In particular, sensitive demersal fishing losses should be targeted in line with the principles of the United Nations Convention on the Law of the Sea. For example, the border to territorial waters can be found in the adjoining zone.

#### REFERENCES

- Korman, S. (2010). International Management of a High Sea Fishery: Political and Property-Rights Solutions and the Atlantic Bluefin. Va. J. Int'l L., 51, 697.
- Merten, W., Reyer, A., Savitz, J., Amos, J., Woods, P., & Sullivan, B. (2016). Global Fishing Watch: Bringing transparency to global commercial fisheries. arXiv preprint arXiv:1609.08756.
- Prontera, A. (2017). The New Offshore Frontiers of EU Energy Security in the Mediterranean: The Politics of Hydrocarbon Development in the Adriatic and Ionian Seas. Mediterranean Politics, 22(3), 383-406.
- Rygiel, K., Baban, F., & Ilcan, S. (2016). The Syrian refugee crisis: The EU-Turkey 'deal'and temporary protection. Global Social Policy, 16(3), 315-320.

Wakefield, J. (2016). Reforming the Common Fisheries Policy. Edward Elgar Publishing.

Winrow, G. M. (2016). The anatomy of a possible pipeline: the case of Turkey and Leviathan and gas politics in the Eastern Mediterranean. Journal of Balkan and Near Eastern Studies, 18(5), 431-447.



# Investigation on Anti-Fungal Effect of Onion (*Allium cepa*) Skin Aqueous Methanolic Extract in Rainbow Trout (*Oncorhynchus mykiss*) Eggs

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**Abstract:** In this preliminary study, it was aimed to determine formation inhibitor effects of onion (*Allium cepa*) skin extract on fungal disease (*Saprolegnia parasitica*) which occurs on rainbow trout (*Oncorhynchus mykiss*) eggs. In order to evaluate fungicidal effect on *S. parasitica*, obtained aqueous methanolic extract was applied to eggs right after fertilization via bathing method in concentrations of 0.1, 0.2, 0.4, 0.8, 1.6 and 3.2 g l<sup>-1</sup>. In the trial, experimental groups showed positive results in terms of preventing formation of the fungus in comparison with control group where, best result was seen in 0.4 g l<sup>-1</sup> group (P<0.05). According to results, we conclude that onion skin may be used in rainbow trout hatcheries as an alternative natural fungicidal.

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Keywords: Onion, Allium cepa, Oncorhynchus mykiss, Saprolegnia parasitica, disease, fungicide



# Seasonal Investigation of Heavy Metal Accumulation in Sediments of Sinop, Kastamonu and Zonguldak Coasts

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Abstract: In this work, the levels of Mn, Cd, Zn, Cu, Fe, Ni, Pb heavy metals in sediment samples obtained from Kastamonu, Sinop and Zonguldak coasts were determined and seasonal changes were investigated ICP-OES (SpectroBlue). According to results of the study, seasonal differences were observed between some heavy metals where, significant correlation was observed between Mn-Cu, Ni-Cu, Pb-Cu, Zn-Cu, Mn-Fe, Ni-Fe, Ni-Mn, Pb-Mn, Zn-Ni, Zn-Pb (p<0.05) and Pb-Ni (p<0.01). While concluding that there are similar pollutants causing heavy metals to be present, the results are generally within tolerable limits.

Keywords: Black Sea, heavy metal, pollution, sediment.

## **1. INTRODUCTION**

The use of sediment-findings in reflecting marine pollution is common in radioactive or chemical pollution studies in the marine environment (Topçuoğlu and Güngör, 1999). Sediment is an important component of the aquatic environment and is formed as a result of chemical and physical processes and the separation of stones or organic materials into small pieces. In addition, the carcasses, wastes and feces of the organisms in the sea subside due to gravity and constitute an important part of the organic part of the sediment. The contaminated sediment structure is soil, sand, organic matter and other minerals which accumulate at the bottom of the water and contain toxic or dangerous materials that may have a high impact on the environment and adverse effects on human health. In addition, sediments are areas that collect nutrients and toxic substances for marine organisms, and many types of pollutants can reach human beings through the food chain. The accumulation of pollutants in sediments adversely affects the natural life and living beings in the environment and causes environmental problems. Sediment analysis is very important for the general understanding of the distribution and transfer of toxic substances (Baran and Tarnawski, 2015).

For this purpose, in the present study, the levels of Mn, Cd, Zn, Cu, Fe, Ni and Pb heavy metals in sediment samples obtained from Kastamonu, Sinop and Zonguldak coasts were determined and seasonal changes were investigated.

## 2. MATERIALS AND METHODS

Sediment samples analyzed in the study were taken from three stations: Sinop, Kastamonu and Zonguldak for four times (December 2016, March 2017, June 2017 and September 2017). Sediment samples were taken from the sediment surface to a depth of maximum 10 cm with plastic trowel, transferred to wide-mouthed plastic containers and brought to the laboratory with cold chain and stored at +4 °C until analysis (Rios-Arana et al., 2003). Heavy metal analyzes were performed in Kastamonu University Central Research Laboratory. Briefly 0.5 g of each sample was taken and HNO<sub>3</sub> and H<sub>2</sub>O<sub>2</sub> were added. The samples were then dried under a pressure of 200 bar and 45 bar for 15 minutes and then cooled to room temperature. After cooling, ultra-pure water was added and readings of the samples were carried out using ICP-OES (SpectroBlue). For statistical analyzes were performed with SPSS (version 22.0) for Windows.

#### 3. RESULTS AND DISCUSSION

|         | Stations (Cu)                                     |   |   |  |  |
|---------|---|---|---|--|--|
| Seasons | 1 (Sinop)   | 2 (Kastamonu)                                     | 3 (Zonguldak)                                     |  |  |
|         | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X}{\pm}\sigma_{\overline{\mathrm{x}}}$ |  |  |
| Autumn  | 13,23±0,85 <sup>A</sup>                           | $7,47 \pm 0,90^{\mathrm{bB}}$                     | 13,88±0,97 <sup>A</sup>                           |  |  |
| Winter  | 14,02±0,41 <sup>A</sup>                           | $9,68 \pm 2,94^{abB}$                             | $12,74\pm1,62^{A}$                                |  |  |
| Spring  | 13,98±1,51  | 12,40±2,03 <sup>ab</sup>                          | 13,41±0,39  |  |  |
| Summer  | 16,94±1,43  | 17,17±0,81ª                                       | 13,86±1,81  |  |  |

**Table 1.** Seasonal variation of copper (Cu) in sediment samples (µg g<sup>-1</sup>)

Lowercase letters indicate the difference between seasons, uppercase letters indicate the difference between stations (p < 0.05).

It can be seen in the Table 1 that the difference between stations was significant in the autumn and winter seasons (p < 0.05), whereas other seasons were not statistically significant (p > 0.05). The difference between the seasons was significant only in Station 2 (p < 0.05), while no seasonal difference was observed in other stations. In addition, the interaction between the seasons and the stations was found to be insignificant in the statistical context. The lowest average was recorded in autumn at Station 2 with  $7.47 \pm 0.90 \ \mu g \ g^{-1}$ , while the highest average was recorded at  $17.17 \pm 0.81 \ \mu g \ g^{-1}$  in Station 2 in summer. Our results are similar with various studies on heavy metal levels in sediment in the literature. Cevik et al. (2008) reported that Cu level was between 161-6259  $\mu$ g g<sup>-1</sup> in their study where they examined heavy metal levels in sediment of Eastern Black Sea coasts. In another study conducted on the coasts of Western Black Sea, Cu levels were reported to range between 12-59.9  $\mu$ g g<sup>-1</sup> (Balkıs et al., 2007). Cu levels were reported to be 13-146.5  $\mu$ g g<sup>-1</sup> in Central Black Sea (Karaalioğlu, 2006), 1.9-107.6 µg g<sup>-1</sup> in Northwest Black Sea [Romania (Secrieru and Secrieru, 2002)], 30-117 µg g<sup>-1</sup> in North East Black Sea [Georgia (Wilson, 2008)], 11.2-64.8 µg g<sup>-1</sup> in Central Black Sea (Bakan and Özkoç, 2007) and 15-82 µg g<sup>-1</sup> in South Black Sea coasts (Yücesoy and Ergin, 1992). When the results of the Cu level in the sediment are evaluated in general, it is consistent with the literature. The second station constituting the statistical difference within the stations is a region where copper mines are located. The reason for the difference in the increase in the summer months can be explained by the intensification of mining and mineral transportation activities in these seasons. Although the other two stations appear to be high in other seasons, they are considered low compared to other studies in the literature. It is known that mining activities are also present in these stations.

|         | Stations (Zn)                                     |   |   |  |  |
|---------|---|---|---|--|--|
| Seasons | 1 (Sinop)   | 2 (Kastamonu)                                     | 3 (Zonguldak)                                     |  |  |
|         | $\overline{X}{\pm}\sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ |  |  |
| Autumn  | 51,49±1,39 <sup>A</sup>                           | $21,43 \pm 1,91^{\mathrm{bB}}$                    | 52,76±0,52 <sup>A</sup>                           |  |  |
| Winter  | 55,34±2,25 <sup>A</sup>                           | 40,33±14,63 <sup>abB</sup>                        | 51,38±6,27 <sup>A</sup>                           |  |  |
| Spring  | 49,92±7,75  | 48,77±9,07 <sup>ab</sup>                          | 45,97±1,85  |  |  |
| Summer  | 64,89±1,66 <sup>A</sup>                           | $60,76\pm1,48^{\mathrm{aAB}}$                     | $49,58{\pm}4,07^{\rm B}$                          |  |  |

Table 2. Seasonal variation of zinc (Zn) in sediment samples ( $\mu g g^{-1}$ )

Lowercase letters indicate the difference between seasons, uppercase letters indicate the difference between stations (p < 0.05).

When Table 2 is examined, it is seen that the difference between stations is significant in autumn, winter and summer seasons (p<0.05) and it is not significant in spring. On the other hand, the seasonal difference was found to be significant only in the Station 2 (p<0.05). Interaction between stations and seasons was also found to be insignificant. The lowest average was measured in autumn at the second station with an average of  $21.43 \pm 1.91 \ \mu g \ g^{-1}$ , while the highest average was recorded in the first station, which was  $64.89 \pm 1.66 \ \mu g \ g^{-1}$ . In a study conducted in Central Black Sea Sinop coasts, it was reported that the Zn level varied between  $12.9-73.8 \ \mu g \ g^{-1}$  (Türk Çulha, 2011). Topçuoğlu et al., (2002) reported that Zn amount was determined to be between  $33.9-267.4 \ \mu g \ g^{-1}$  in a study where they conducted in Black Sea coasts, whereas Cevik et al., (2008) reported it to be  $125-2344 \ \mu g \ g^{-1}$  in Eastern Black Sea coasts. Similarly Zn levels were reported to be  $24-138 \ \mu g \ g^{-1}$  in South Black Sea coasts (Yücesoy and Ergin, 1992),  $17-456.6 \ \mu g \ g^{-1}$  in All Sud South Black Sea coasts (Kıratlı and Ergin, 1996). Our study results are consistent with the results of the studies conducted in the Black Sea. In addition, the seasonal difference between the stations emerged in favor of stations 1 and 3 in autumn and winter. In the summer, Station 1 was higher than other stations. Here, it is assumed that the basic element is the sampling region. Although all three stations have pollutants in terms of metals, proximity of stations where levels were

higher to these pollutants should be considered. In the second station, higher values in summer compared to other seasons are attributed to the intensification of industrial activities and mining activities during this season. Also, the intensity of mine transport and shipyard activities increase in these months.

|         |   | Stations (Ni)                                     |   |  |
|---------|---|---|---|--|
| Seasons | 1 (Sinop)   | 2 (Kastamonu)                                     | 3 (Zonguldak)                                     |  |
|         | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ |  |
| Autumn  | 20,12±1,04 <sup>A</sup>                           | $14,24\pm1,10^{bB}$                               | 21,82±1,46 <sup>A</sup>                           |  |
| Winter  | 20,96±0,57  | $20,15\pm2,76^{ab}$                               | 19,68±0,82  |  |
| Spring  | 21,54±0,41 <sup>B</sup>                           | 21,68±0,03ªB                                      | 25,19±0,28 <sup>A</sup>                           |  |
| Summer  | 20,80±0,48  | $21,62\pm0,50^{a}$                                | 21,47±2,33  |  |

| Table 3. Seasonal variation of nicke | (Ni) in sediment | samples ( $\mu g g - 1$ ) |
|--------------------------------------|------------------|---------------------------|
|--------------------------------------|------------------|---------------------------|

Lowercase letters indicate the difference between seasons, uppercase letters indicate the difference between stations (p < 0.05).

Table 3 shows that the seasonal variation in nickel was only significant at the second station (p < 0.05). Difference between the stations in the seasons was statistically significant (p < 0.05) in transition seasons and insignificant in other seasons (p>0.05). Interaction between seasons and stations was also found to be insignificant. The lowest mean was  $14.24 \pm 1.10$  $\mu g g^{-1}$  at the second station in autumn and the highest mean was  $25.19 \pm 0.28 \mu g g^{-1}$  at the third station in spring. In a study conducted on the Black Sea coast, it was reported that the Ni level in sediment ranged between 13.5-65.2  $\mu$ g g<sup>-1</sup> (Topçuoğlu et al., 2002). In a study conducted on the Romanian coast, the amount of Ni in sediment was reported to be between 1-207  $\mu$ g g<sup>-1</sup> (Secrieru and Secrieru, 2002). Similarly, Ni amounts were reported to be 0-21.4  $\mu$ g g<sup>-1</sup> in Eastern Black Sea (Cevik et al., 2008), 11-202 µg g<sup>-1</sup> in Southern Black Sea coast (Yücesoy and Ergin, 1992), 80-134.3 µg g<sup>-1</sup> in Western Black Sea coast (Balkıs et al., 2007), 10-139 µg g<sup>-1</sup> in Eastern Black Sea coasts (Yılmaz Bayrak, 2016). The Ni data obtained in our study ranged between 14.24-25.19  $\mu$ g g<sup>-1</sup>. The data are consistent with the literature in the Black Sea. In fact, it was observed to be lower than those of reported in many studies. Seasonal difference occurred only in the second station; it was lower in autumn while it was higher in summer. This appearance is attributed to increasing industrial activities in summer. Mining and wood industry activities which are concentrated in the region reach their maximum intensiveness in summer months. On the other hand, the difference between stations is seen to be relatively high in station 3 in two seasons compared to other stations. The emergence of these, especially during transition seasons, can be explained as the mixing of the metals carried by precipitation into the water in the stations area. Because, Ni is already an abundant element in the environment that forms compounds with oxygen and sulfur, and it is genereally found in sediment and soil bound to particles that contain iron and manganase (ATSDR, 2003). Nickel can be deposited easily by moving to water sources with surface flows (Sönmez, 2011). This may be the reason for the slight increase in transition seasons.

| <b>Table 4.</b> Seasonal variation of iron (Fe) in sediment samples ( $\mu g g^{-1}$ ) |  |
|--|--|
|--|--|

|         | Stations (Fe)                            |   |   |  |  |
|---------|--|---|---|--|--|
| Seasons | 1 (Sinop)                                | 2 (Kastamonu)                                     | 3 (Zonguldak)                                     |  |  |
|         | $\overline{X} \pm \sigma_{\overline{x}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ |  |  |
| Autumn  | 9490,30±536,05 <sup>A</sup>              | $3490,54 \pm 413,74^{bB}$                         | 8594,02±165,51 <sup>A</sup>                       |  |  |
| Winter  | 9486,15±437,55                           | 7422,26±2933,73 <sup>ab</sup>                     | 7985,20±744,29                                    |  |  |
| Spring  | 10991,84±2288,14                         | 11185,38±1978,46ª                                 | 14999,06±662,21                                   |  |  |
| Summer  | 9365,64±247,14                           | 9226,90±247,34 <sup>ab</sup>                      | 11405,66±3342,52                                  |  |  |

Lowercase letters indicate the difference between seasons, uppercase letters indicate the difference between stations (p < 0.05).

As shown in Table 4, the difference between stations was significant only in autumn (p<0.05). On the other hand, the difference between seasons was found to be significant only in second station (p<0.05). The interaction between the seasons and the stations also appeared to be insignificant in the statistical context. The lowest was recorded in autumn at the second station with  $3490.54 \pm 413.74 \ \mu g \ g^{-1}$ , while the highest average was recorded in spring at Station 3 with  $14999.06 \pm 662.21 \ \mu g \ g^{-1}$ . When the previous studies on iron in sediment are examined, it was seen that it varies. In a study conducted on the Black Sea coast, Fe levels in sediment ranged between 1.3-4.3% (Kıratlı and Ergin, 1996), yet another study on the Black Sea coast reported Fe levels to be between 0.5-5.4% (Topçuoğlu et al. 2002). Similarly, Fe level in a study conducted on Southern Black Sea coast sediments was reported to range between 0.2-4.9% (Yücesoy and Ergin, 1992). It was reported to be 4.7-48.1% in Western Black Sea, 5.6-6% in Eastern Black Sea, 4.8-11% in another

study on Eastern Black Sea coast, 8.8-25.5% in Eastern Black Sea (Balkıs et al., 2007; Ergül and Topçuoğlu, 2008; Yılmaz Bayrak, 2016; Çevik et al., 2008). In our study results, Fe level was found to vary between 0.35-1.5%, which is lower than literature. In general, seasonal difference was seen in the second station in spring, which can be explained by the increase in metal levels carried by precipitation. On the other hand, higher Fe levels at the first and third stations in autumn is also related to the location of the stations and rainfall levels during the sampling season. Although some differences occur in some stations and seasons, the values are generally below acceptable and reference limits used [crust 5% (Mason and Moore, 1982), shale 4.7% (Turekian and Wedepohl, 1961), ultrabasic rock 5-7% (Aslaner, 1973), sandstone 0.9% (Turekian and Wedepohl, 1961)].

|         | Stations (Mn)                                     |   |   |  |  |
|---------|---|---|---|--|--|
| Seasons | 1 (Sinop)   | 2 (Kastamonu)                                     | 3 (Zonguldak)                                     |  |  |
|         | $\overline{X}{\pm}\sigma_{\overline{\mathrm{x}}}$ | $\overline{X}{\pm}\sigma_{\overline{\mathrm{x}}}$ | $\overline{X}{\pm}\sigma_{\overline{\mathrm{x}}}$ |  |  |
| Autumn  | 401,42±16,76 <sup>A</sup>                         | 156,83±22,39 <sup>bB</sup>                        | 351,42±5,46 <sup>A</sup>                          |  |  |
| Winter  | 416,41±16,07                                      | 358,56±54,07ª                                     | 352,75±37,31                                      |  |  |
| Spring  | 350,86±11,06 <sup>B</sup>                         | $383,10\pm9,43^{aB}$                              | 427,94±5,68 <sup>A</sup>                          |  |  |
| Summer  | 400,99±32,37                                      | 362,04±8,33ª                                      | 381,21±39,18                                      |  |  |

**Table 5.** Seasonal variation of manganese (Mn) in sediment samples ( $\mu g g^{-1}$ )

Lowercase letters indicate the difference between seasons, uppercase letters indicate the difference between stations (p < 0.05).

Manganese data are presented in Table 5. The difference between stations was significant in autumn and spring seasons (p<0.05). The difference between the seasons was significant only in the second station (p<0.05). On the other hand, it was concluded that there was a significant (p < 0.05) interaction between seasons and stations. The lowest mean was measured in autumn at the second station with  $156.83 \pm 22.39 \ \mu g \ g^{-1}$ , while the highest mean was in spring season 427.94  $\pm$  5.68 µg g<sup>-1</sup> at the third station. When the studies are examined, similar results have been obtained compared with the studies in Black Sea. In a study conducted in Eastern Black Sea, Mn levels were reported to be between 504-1674 µg g<sup>-1</sup> (Cevik et al. 2008). In another study, it was determined as  $651-1022 \ \mu g \ g^{-1}$  whilst, Yılmaz Bayrak (2016) reported that Mn levels in sediment of Eastern Black Sea were between 80 and 1334  $\mu$ g g<sup>-1</sup>. Similarly it was found to be 102-1064  $\mu$ g g<sup>-1</sup> in Southern Black Sea (Yücesoy and Ergin, 1992), 202-3696 µg g<sup>-1</sup> in Western Black Sea (Balkıs et al., 2007), 161.5-668.7 µg g<sup>-1</sup> in Central Black Sea (Bakan and Özkoç, 2007) and 355-751 µg g<sup>-1</sup> in all Black Sea coasts (Kıratlı and Ergin, 1996). Manganese data were generally low compared to the literature. In the second station, where there is a seasonal difference, a significant increase was observed in summer compared to other seasons. In the third station, the difference was observed especially in spring. This is mainly attributed to rainfall. In natural resources, the mixture of manganese to water and soil occurs through discharge of wastes and atmospheric transport. It is found naturally in rivers, lakes and underground waters, can be uptaken by aquatic plants and accumulate (ATSDR, 2010). On the other hand, the results were found lower than various geological sources used as a reference such as crust, shale and calcite (Mason and Moore, 1982; Turekien and Wedepohl, 1961; Aslaner, 1973).

|         | Stations (Cd)                                     |   |   |  |  |
|---------|---|---|---|--|--|
| Seasons | 1 (Sinop)   | 2 (Kastamonu)                                     | 3 (Zonguldak)                                     |  |  |
|         | $\overline{X}{\pm}\sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X}{\pm}\sigma_{\overline{\mathrm{x}}}$ |  |  |
| Autumn  | $0,97{\pm}0,07^{a}$                               | 0,54±0,01 <sup>ab</sup>                           | 0,56±0,16   |  |  |
| Winter  | $0,67{\pm}0,19^{abB}$                             | $1,23\pm0,39^{\mathrm{aA}}$                       | $0,37\pm0,10^{\circ}$                             |  |  |
| Spring  | $0,26{\pm}0,08^{\mathrm{bB}}$                     | $0,36\pm0,13^{bAB}$                               | $0,74{\pm}0,01^{\rm A}$                           |  |  |
| Summer  | $0,26\pm0,03^{bB}$                                | $0,64{\pm}0,02^{\rm abA}$                         | $0,43{\pm}0,06^{\rm B}$                           |  |  |

| Table 6. Seasonal variation of cadmium (Cd | l) in sediment samples ( $\mu g g^{-1}$ ) | ) |
|--|---|---|
|--|---|---|

Lowercase letters indicate the difference between seasons, uppercase letters indicate the difference between stations (p < 0.05).

When the Table 6 regarding the cadmium level in the sediment samples was examined, the seasonal difference was statistically significant at the first and second stations (p<0.05). The difference between stations was significant (p<0.05) in three seasons except autumn. In addition, the interaction between seasons and stations was observed as significant (p<0.05). The lowest Cd mean was recorded at station 1 with  $0.26 \pm 0.03 \ \mu g \ g^{-1}$  in summer, while the highest mean was recorded at station 2 with  $1.23 \pm 0.39 \ \mu g \ g^{-1}$  in winter. Turkmen (2003) reported that the lowest level of Cd was 1.56 mg kg<sup>-1</sup> in summer, whereas the highest mean was in winter with 7.33 mg kg<sup>-1</sup> in Gulf of İskenderun. In another study conducted by Riyad Çam (2016) in Gulf of İskenderun, the lowest Cd level was reported to be  $13 \pm 6 \ mg \ kg^{-1}$  in winter

and the highest level was reported to be  $207 \pm 15$  mg kg<sup>-1</sup> in spring among 10 different stations. In a study conducted in Mersey Bay in England, Cd level was between 0.01-11 ppm (Harland et al., 2000), it was reported to be between 0.05-2.5 ppm in Northwest Iberian Peninsula (Prego and Cobelo-Garcia, 2003), and 0.3-3.1 g kg<sup>-1</sup> in San Francisco Bay (Bradford and Luoma, 1980). Although there is not much literature regarding Cd level in the sediments of Black Sea coasts, our results are in line with the results of the studies conducted in other seas and even lower than some. At the second station, the difference occurred in winter, whereas it was in autumn at the first station. The difference between stations was in at station 2 in winter and at station 3 in spring. In general, Cd levels in sediment were not extremely high considering the values in water. This is attributed to location of the stations and level of exposure. In fact, cadmium is a very common element used in the industry and it is found in crust between 0.1-0.5 mg kg<sup>-1</sup>. In addition, it may be found in sedimentary rocks, marine phophates and phosphorites at higher levels (Cook and Morrow, 1995). When examined from this point of view, although there are partial differences between seasons and stations in our study, the results are accepted as normal.

|         | Stations (Pb)                                     |   |   |  |  |
|---------|---|---|---|--|--|
| Seasons | 1 (Sinop)   | 2 (Kastamonu)                                     | 3 (Zonguldak)                                     |  |  |
|         | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ | $\overline{X} \pm \sigma_{\overline{\mathrm{x}}}$ |  |  |
| Autumn  | 17,98±0,53 <sup>abA</sup>                         | 11,36±0,68 <sup>bB</sup>                          | 18,32±0,20 <sup>A</sup>                           |  |  |
| Winter  | 20,55±0,81 <sup>abA</sup>                         | $15,88 \pm 4,18^{abB}$                            | $18,75\pm1,28^{A}$                                |  |  |
| Spring  | 17,51±1,56 <sup>b</sup>                           | 16,69±1,76 <sup>ab</sup>                          | 15,32±0,41  |  |  |
| Summer  | 21,35±0,24 <sup>a</sup>                           | 21,19±0,70ª                                       | 18,32±1,74  |  |  |

**Table 7.** Seasonal variation of lead (Pb) in sediment samples ( $\mu g g^{-1}$ )

Lowercase letters indicate the difference between seasons, uppercase letters indicate the difference between stations (p < 0.05).

When Table 7 examined it can be observed that the difference between stations is significant in autumn and winter seasons (p<0.05). Seasonal differences were also significant in stations 1 and 2 (p<0.05), wheras there was no seasonal difference in station 3. Interaction between seasons and stations is also insignificant in statistical context (p>0.05). The lowest value was recorded as  $11.36 \pm 0.68 \ \mu g \ g^{-1}$  at the second station in autumn and the highest mean was recorded as  $21.19 \pm 0.70$ µg g<sup>-1</sup> at the second station in summer. Although Pb studies show a wide variation, Pb level in sediment was reported to be between 0.05-31  $\mu g g^{-1}$  in a study conducted in Black Sea coast (Topçuoğlu et al., 2002), and in another study performed at the same location, it has been reported to be between 14-35 µg g<sup>-1</sup> (Kıratlı and Ergin, 1996). In the studies carried out in Western Black Sea, the amount of Pb in sediment was reported to be between 0.1-23.5 µg g<sup>-1</sup> and 0.5-50.3 µg g<sup>-1</sup>, respectively (Balkıs et al., 2007; Secrieru and Secrieru, 2002). In various studies conducted in Central and Eastern Black Sea, Pb levels in sediment were reported to be 16.2-139.6  $\mu$ g g<sup>-1</sup> (Karaalioğlu, 2006), 2.27-16.7  $\mu$ g g<sup>-1</sup> (Türk Çulha, 2011), 41.7-355.1  $\mu$ g g<sup>-1</sup> (Cevik et al., 2008), and 14-920  $\mu$ g g<sup>-1</sup> (Yılmaz Bayrak, 2016). When the Pb data obtained from our study are evaluated in general, they correspond to the amounts stated in the literature and in the reference sources (Mason and Moore, 1982; Turekian and Wedepohlon, 1961). Seasonal difference usually occurs in the summer months with a high mean. This is considered to be normal due to increasing momentum in industrial activities and subsequent contamination in these months. Again, high mean value in stations 1 and 3 which was seen in autumn and winter seasons was attributed to the amount of rainfall in the region. As Kesler (1994) stated; Pb, which is a very common metal in the crust, is present in the soil at the level of 12.5 mg kg<sup>-1</sup> and is highly absorbed by soil sediment particles. Mine sources in the environment have a major impact on contamination and mining activities in the region also play an important role in explaining this status.

#### **Correlation of Heavy Metals Measured in Sediment**

|    | Cd     | Cu           | Fe      | Mn      | Ni           | Pb      | Zn |
|----|--------|--------------|---------|---------|--------------|---------|----|
| Cd | 1      |              |         |         |              |         |    |
| Cu | -0,175 | 1            |         |         |              |         |    |
| Fe | 0,031  | 0,366        | 1       |         |              |         |    |
| Mn | 0,169  | $0,600^{**}$ | 0,763** | 1       |              |         |    |
| Ni | 0,066  | 0,516**      | 0,881** | 0,812** | 1            |         |    |
| Pb | -0,049 | 0,899**      | 0,228   | 0,625** | $0,420^{*}$  | 1       |    |
| Zn | -0,082 | 0,939**      | 0,360   | 0,716** | $0,548^{**}$ | 0,963** | 1  |

Table 8. Correlation coefficients between elements in sediments

\*\* Correlation is significant at the level of 0.01, \* Correlation is significant at the level of 0.05

The correlations of heavy metals in sediment samples are given in Table 8. According to the table there was a significant correlation between Mn-Cu, Ni-Cu, Pb-Cu, Zn-Cu, Mn-Fe, Ni-Fe, Ni-Mn, Zn-Mn, Zn-Ni and Zn-Pb metals (p<0.01) and Pb-Ni metals (p<0.05). Similarly, in the study conducted by Yılmaz Bayrak (2016), a strong positive correlation was found between Ni-Mn and Zn-Cu metals in sediment. In another study conducted by Riyad Çam (2016) in Iskenderun Gulf, the correlation results obtained in this study correspond to similar studies. The correlations between metal levels generally vary according to the sources of contamination, relation with each other and some environmental factors. Correlation results are consistent with the heavy metal levels discussed individually.

#### 4. CONCLUSION

In this study, it was aimed to determine the concentrations of Cd, Cu, Fe, Mn, Ni, Pb, and Zn in sediment samples taken from three stations (Sinop, Kastamonu and Zonguldak) located in Western Black Sea coastline seasonally. The heavy metal data obtained from the sediment samples support each other in terms of differences and seasonal differences are generally observed during the transition seasons. Differences in terms of stations generally changed depending on the location of the station, diversity and distribution of pollutants. In addition, it was observed that the geological structure of the stations affected the heavy metal levels. The occurrence of some heavy metals above the specified limits is attributed to intensity of industry and industrial activities in that particular area. On the other hand, shipyard activities and port transportation also affect this situation adversely. In addition, annual precipitation and the heavy metals carried by the large and small streams have an impact on the present condition in some seasons. According to the results of this study, it is suggested that pollutants and mining activities in the region should be supervised, and such studies are needed to be carried out frequently and extensively.

#### REFERENCES

- Aslaner, M. (1973). Iskenderun-Kırıkhan Bölgesindeki Ofiyolitlerin jeoloji ve petrografisi: MTA Yayl., 150, 78 s., Ankara.
- ATSDR. (2003). Agency for Toxic Substances and Disease Registry, http://www.atsdr.cdc.gov/toxfaq.html
- ATSDR. (2010). Agency for Toxic Substances and Disease Registry, http://www.atsdr.cdc.gov/toxfaq.html
- Bakan, G., & Özkoç, H. (2007). An ecological risk assessment of the impact of heavy metals in surface sediment on biata from mid-Black Sea coast of Turkey. *International Journal of Environmental Studies*, 64, 45-57.
- Balkıs, N., Topcuoğlu, S., Güven, K.C., Öztürk, B., Topaloğlu, B., Kırbaşoğlu, C., & Aksu, A. (2007). Heavy metals in shallow sediments from the Black Sea, Marmara Sea and Aegean Sea regions of Turkey. J. Black Sea/Medit. Environ., 13, 147-153.
- Baran, A., & Tarnawski, M. (2015). Assessment of heavy metals mobility and toxicity in contaminated sediments by sequential extraction and a battery of bioassays. *Ecotoxicology*, 24, 1279-1293.
- Bradford, W. L., & Luoma, S. N. (1980) Some perspectives on heavy metal concentrations in shellfish and sediment in San Francisco Bay, California. In Contaminants and Sediments (Edited by Baker R. A.), Vol. 2. Ann Arbor Scientific Publishing, Ann Arbor, MI.
- Cevik, U., Damla, N., Kobya, A. I., Bulut, V.N., Duran, C., Dalgic, G., & Bozaci, R. (2008). Assessment of metal element concentrations in mussel (*M. galloprovincialis*) in Eastern Black Sea. *Turkey Journal of Hazardous Matterial*, 160, 396-401.
- Cook, M. E., & Morrow, H. (1995). Anthropogenic sources of cadmium in Canada. National workshop on cadmium transport into plants, Canadian Network of Toxicology Centres. Ottawa, Ontario, Canada, June 20–21.

- Ergül, H.A., Topcuoğlu, S., Ölmez, E., & Kırbaşoğlu, Ç. (2008). Heavy metals in sinking particles and bottom sediments from the eastern Turkish coast of the Black Sea. *Estuar. Coast. Shelf S.*, 78(2), 396-402.
- Harland, B. J., Taylor, D., & Wither, A. (2000). The distribution of mercury and other trace metals in the sediments of the Mersey Estuary over 25 years 1974–1998. *Science of the Total Environment*, 253(1-3), 45–62.
- Karaalioğlu, O. (2006). Sinop İli Kıyı Şeridinde Deniz Ortamı Kalitesinin Belirlenmesi. Yüksek Lisans Tezi. Ondokuz Mayıs Üniversitesi, Fen Bilimleri Enstitüsü, Çevre Mühendisliği Anabilim Dalı, Sinop, Türkiye, 109 s.
- Kesler, S. E. (1994). Mineral Resources, Economics and the Environment. *New York: Macmillan College Publishing Company*, Inc., s223, USA.
- Kıratlı, N., & Ergin, M. (1996). Partitioning of heavy metals in surface Black Sea sediments. Applied Geochemistry, 11, 775-788.
- Mason, B., & Moore, C. R. (1982). Principles of geochemistry p. 344. New york, N. Y.: Wiley.
- Prego, R., & Cobelo-García, A. (2003). Twentieth century overview of heavy metals in the Galician Rias (NW Iberian Peninsula). Environmental Pollution, 121(3), 425–452.
- Rios-Arana, J., Walsh, E., & Gardea-Torresdey, J. (2004). Assessment of arsenic and heavy metal concentrations in water and sediments of the Rio Grande at El Paso–Juarez metroplex region. *Environment International*, 29(7), 957–971.
- Riyad Çam, A. (2016). İskenderun Körfezinde Deniz Suyu Ve Sedimentteki Ağır Metal İçeriğinin Icp-Oes İle Belirlenmesi Ve Analitik Metotlarla Karakterizasyonu. Yüksek Lisans Tezi, Mustafa Kemal Üniversitesi, Fen Bilimleri Enstitüsü, Hatay, Türkiye.
- Secrieru, D., & Secrieru, A. (2002). Heavy Metal Enrichment of Man-made Origin of Superficial Sediment on the Continental Shelf of the North-western Black Sea. *Estuarine, Coastal and Shelf Science*, 54(3), 513–526.
- Sönmez, A. Y. (2011). Karasu ırmağında ağır metal kirliliğinin belirlenmesi ve bulanık mantıkla değerlendirilmesi. Doktora Tezi, Atatürk Üniversitesi, Fen Bilimleri Enstitüsü, Su Ürünleri Mühendisliği Anabilim Dalı, Erzurum, Türkiye.
- Topcuoğlu, S., & Güngör, N. (1999). Radionuclide concentrations in macroalgae and sediment samples from the Bosphorus. *Turkish J. Marine Sciences*, 5, 19-24.
- Topcuoğlu, S., Kırbaşoğlu, C., & Güngör, N. (2002). Heavy metal organisms and sediments from Turkish Coast of the Black Sea, 1997-1998. *Environment International*, 27, 521-526.
- Turekian, K.K., & Wedepohl, K.H. (1961). Distribution of the elements in some major units of the earth's crust. Geological Society of America Bulletin. 72(2), 175-192.
- Türk Çulha, S., (2011). Sinop İç Liman'da (Karadeniz) batırılmış uzun halat sisteminde yetiştirilen midyelerdeki (Mytilus galloprovincialis L., 1819) iz element seviyeleri, Sinop Üniversitesi / Fen Bilimleri Enstitüsü / Su Ürünleri Yetiştiriciliği Anabilim Dalı, Doktora Tezi, 210 s
- Türkmen A. (2003). İskenderun Körfezi'nde Deniz Suyu, Askıdaki Katı Madde, Sediment ve Dikenli Taş İstiridyesi'nde (Spondylus spinosus Schreibers, 1793) Oluşan Ağır Metal Birikimi Üzerine Araştırma. *Atatürk Üniversitesi, Fen Bilimleri Enstitüsü*, Doktora Tezi, pp. 152, Erzurum.
- Wilson, J.G., Komakhidze, A., Osadehaya, T., Alyomov, S., Romanov, A., & Tediashvili, M. (2008). Evaluating ecologial quality in the North-eastern Black Sea coastal zone. *Marine Pollution Bullettin*, 57, 202-207.
- Yılmaz Bayrak, E. (2016). Doğu Karadeniz Kıyısal Alanının Ağır Metal Kirliliğinin Tespiti Ve Akdeniz Midyesinin (Mytilus galloprovincialis) Cu Tutma (Adsorbsiyon) Kapasitesinin Araştırılması. Doktora Tezi, Recep Tayyip Erdoğan Üniversitesi Fen Bilimleri Enstitüsü, Rize.
- Yücesoy, F., & Ergin, M. (1992). Heavy-metal geochemistry of surface sediments from the southern Black Sea shelf and upper slope. Chemical Geology, 99,265-287.



# Investigation of Ermenek Dam in terms of Fisheries Potential

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**Abstract:** In this research, it is aimed to determine the aquaculture potential of Ermenek Dam which is located on Ermenek River in Ermenek District, Karaman Provencie. To this end, five various stations were assigned during the study from November 2013 to April 2014. Water samples were monthly collected 30 cm. below the water surface via these stations. Obtained average annual values, minimum and maximum, are as follows respectively Temperature of Water (9.8-14.5-11,7 °C), pH (7.51-8.50-8.02), free oxygen (8.01-11.18-9.50 mgL-1), conductivity (220-443-323  $\mu$ S/cm), sodium (4.54 11.04-7.69 mgL-1), potassium (1.69-2.30-2.01 mgL-1), calsium (20-45-32.36 mgL-1), magnezyum (9-23.46-15.00 mgL-1), nitrit (0.01-0.9-0.10 mgL-1), nitrat (0.03-4.20-1.81 mgL-1), amonium (0.02-0.49-0.14 mgL-1), total fosfor (0.02-0.42-0.724 mgL-1). Carrying capacity is 54 000 tonnes. Besides, fish species detected in the locality are; brown trout, Salmo trutta macrostigma (Duméril, 1858), Chub, Squalius cephalus (L., 1758), Common carp, Cyprinus carpio L., 1758, Rainbow trout, Oncorhynchus mykiss (Walbaum, 1792). Upon the findings through the study, the dam is considered as assessable in terms of aquacultural work facilities.

Keywords: Ermenek Dam, capacity, aquacultural potential, fish species

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# Embryonic and Larval Development Stage of European Lobster (*Homarus gammarus*, Linnaeus 1758)

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**Abstract:** The main aim of this study is to determine of embryonic and larval development stage of European lobster (*Homarus gammarus*, Linnaeus 1758). Adult lobsters were collected from Dardanelles Bay, Çanakkale Turkey. Larvae reared 12.7-13.4°C and feed by *Nannochloropsis* sp. during the first two days and then by *Artemia salina*. As a result, egg diameter of lobster was determined as from 2.5 to 2.7 mm. All larval development stages were monitored for 27 days. Larval length and weight reached to 11.60 mm and 0.034 g, respectively. In summary, The European lobster is a very important crustacean species all over the world. It has a high market value, three times the price of its American lobster (*H. americanus*). However, production of European lobster is done only fishing. But aquaculture activities are needed for the sustainable production of this species. These data will be strategic element advance understanding embryonic and larval rearing of European lobster. Therefore, outputs of present study have a great importance to provide knowledge for aquaculture activities and importance to ensure the sustainable production.

Keywords: European lobster, Homarus gammarus, larvae, embryonic stage, growth

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# Effects of Sweet Potato (*Ipomoea batatas*) Methanolic Extracts on Growth Performance of Nile Tilapia (*Oreochromis niloticus*)

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**Abstract:** The purpose of this study was to determine the effects of sweet potato (*Ipomoea batatas*) in diets for juvenile tilapia ( $3.4\pm0.04$  g mean initial weight) on growth performance of fish. With this aim, three different concentrations of methanolic extract of sweet potato (1, 2 and 3 g kg<sup>-1</sup>) were prepared and spreyed to the feed. Fish were fed with the diets (control, T1, T2 and T3) for 60 days At the end of the 60 day feeding trial, the highest mean individual weight gain, specific growth rates (SGR) and feed conversion ratio (FCR) of fish was found in T1 group (P<0.05). But the condition factor (CF) value was found higher in T2 group than the other groups (P<0.05). SGR values of T2 and control were lower than other groups (P<0.05). The lowest mean FCR was found in control group ( $0.97\pm0.14$ ) but not significantly different from T3 (P>0.05). The results of present study showed that methanolic extracts of sweet potato was increased the fish growth performance.

Keywords: Tilapia, sweet potato, growth performance

### **1. INTRODUCTION**

Tilapia are the most cultured freshwater fish in the Philippines and they are listed second in culture volume after milkfish. Until the early 1990's the Philippines was the globally the top producer of tilapia and most of it was produced for domestic consumption. Since then, many countries have developed their tilapia industry to take advantage of the booming international demand for filleted tilapia. The Philippines completely missed out on this opportunity; the country has not really developed or professionalized its tilapia industry and is still producing for the domestic market only (FAO, 2017).

Philippines has a very rich floral diversity and a great knowledge of folk medicines and consequently represents potential resources for such studies (Penecilla and Magno, 2011). Sweet potato is important as a cash crop with high levels of vitamins (A, B<sub>1</sub>, B<sub>2</sub>, C and E), minerals (Ca, Mg, K and Zn), antioxidants, dietary fibre and carbohydrates (Scott et al., 2000; Kim et al., 2012). It is livestock feed or for industrial input (Woolfe, 1993). Sweet potato (Ipomoea batatas) is a herbaceous creeping plant with smooth, lightly moderate green leaves sometimes with a considerable amount of purple pigmentation especially along its veins (Longe, 1986). Nowadays, medicinal plant is a growing area as an alternative medicine for human being and many manufactured drugs derived originally from plant compounds have wide-range uses. The medicinal plants are rich in a wide variety of nutrients and they may be used as chemotherapeutics and feed additives (Chang, 2000). The use of medicinal plants as a natural feed additive in fish diets is becoming useful rather than classic chemicals, which may have an accumulative effect on human and animal health.

#### 2. MATERIAL AND METHOD

#### **Experimental Design and Fish**

Tilapia, with a mean initial body weight of  $3.4\pm 0.47$  g, were obtained and stocked randomly (20 fish aquarium<sup>-1</sup>) into 12 aquarium, each with a 100 L capacity, in The Fisheries Biotechnology Center, BFAR-NFFTC, National Fisheries Research and Development Institute, Central Luzon University, Philippines. The experiment was designed as three replicates of each treatment. Before starting the experiment, fish were acclimatized to the experimental feeding regimen using a commercial diet for 2 weeks (tilapia commercial powder feed). Aqueous methanolic extracts of sweet potato (SW) were added to the fishes' basal diet at the rate of 0 (Control), 1 (SW1), 2 (SW2) and 3 (SW3) g kg<sup>-1</sup> by spraying. All groups were fed with the diets twice a day ad-libitum.

#### Preparation of Aqueous Methanolic Extract of Sweet Potato (Ipomoea batatas)

Sweet potatoas were purchased from market and dried under shade in natural conditions; extracts of sweet potato were prepared according the procedure described by Bilen et al. (2016).

#### **Growth Parameters**

Bulk fish live weight increments were measured every 2 weeks and feed intake was recorded daily throughout the study. At the end of the study, fish were taken individually weight and length for determining growth performance parameters. Growth performance measured are listed below and the calculations were according to Ricker (1979);

Weight gain (%) = [(final weight-initial weight) / initial weight]×100;

Specific growth rate (SGR) = [(ln final weight-ln initial weight) /days]×100;

Condition factor (CF) =  $100 \text{ x} [(\text{body weight } (g) / \text{length}^3 (\text{cm})];$ 

Feed conversion ratio (FCR) = wet weight gain (g) / feed intake (g);

#### **Statistical Analysis**

Statistical analyses of data was subjected to one-way ANOVA, and a subsequent comparison of means by Tukey's multiple range test. All of the above mentioned statistical analyses were performed using SPSS (23.0). Differences were considered statistically significant at P<0.05.

### 3. RESULTS AND DISCUSSION

As presented in Table 1 average of initial weight (g/fish), final body weight (g/fish), weight gain (WG) (g/fish), and specific growth rate (SGR %/day) as affected with methanolic extracts of sweet potato. Final fish weight in the W2 group was significantly higher than was that in the control and other experimental groups (P < 0.05). A significant increase in WG and SGR was observed in all experimental groups compared with that in the control group and higher levels were recorded in the W2 group. FCR was significantly increased in all treated groups compared with that in the control group, and the lowest FCR value was obtained in the control group.

**Table 1.** Values of initial and final weights, specific growth rate, feed consumption and feed conversion ratio of experimental diets<sup>1</sup>

|                             | <b>Dietary Treatments</b> |                       |                         |                              |  |
|-----------------------------|---------------------------|-----------------------|-------------------------|------------------------------|--|
|                             | Control                   | SW1                   | SW2                     | SW3                          |  |
| Initial weight (g/fish)     | 3.4±0.32                  | 3.4±0.21              | 3.4±0.71                | 3.4±0.46                     |  |
| Final weight (g/fish)       | $4.28 \pm 0.76^{\circ}$   | 4.69±0.25ª            | $4.24 \pm 0.78^{\circ}$ | $4.38{\pm}0.98^{\mathrm{b}}$ |  |
| Weight gain (%)             | 25.88±0.66°               | 37.92±0.23ª           | $24.82{\pm}0.75^{d}$    | 28,76±0.79 <sup>b</sup>      |  |
| Specific growth rate (SGR)  | $143.36{\pm}0.65^{b}$     | $152.49{\pm}0.98^{a}$ | 142.51±0.43°            | $145.62 \pm 0.56^{b}$        |  |
| Feed conversion ratio (FCR) | $0.97{\pm}0.62^{d}$       | $1.18 \pm 0.43^{b}$   | $1.21{\pm}0.56^{a}$     | $1.04{\pm}0.74^{\circ}$      |  |
| Condition factor            | 1.62±0.14°                | $1.67 \pm 0.17^{b}$   | 1.79±0.15ª              | $1.66{\pm}0.19^{b}$          |  |

<sup>1</sup>Data are reported as mean  $\pm$  SE of three replicates (n = 3). Means with different superscript letters in a row are significantly different (P < 0.05). <sup>2</sup>n = 60 × 2. SW1, SW2 and SW3 are extracts of sweet potato at 1, 2 and 3 g kg<sup>-1</sup> diet, respectively.

Plant extracts are known to promote growth, increase appetite, enhance immune ability, as well as have stress-reduction, sexual stimulation, hepato-protective effects and anti-pathogenic properties in fish (Yang et al., 2011; Reverter et al., 2014). In the study, growth performance parameters, such as final weight and SGR levels displayed. Especially, SW1 and SW3 caused dose dependent growth promotion. Similar to the present study, significantly increased final weight and SGR were reported in rainbow trout fed with capper and nettle extract, respectively (Bilen et al. 2016a; Bilen et al. 2016b). On the other hand, use of laurel caused no effects on growth of rainbow trout (Bilen and Bulut 2010). Abdelhamid (2010) was reported that the diet containing Alpinia (AM) as a medicinal plant produced the best growth performance parameters compared with other different medicinal plants (Ginger, *Cresson* and *Lpecdcuanha*). Tan et al. (2018) were reported that dietary ginkgo biloba leaf (GBE) levels did not significantly affect final weight (FW), specific growth rate (SGR), feed intake (FI), condition factor (CF) after 8 weeks of feeding (P>0.05). Fish fed Diet 6 had significantly lower weight gain (WG) and feed efficiency ratio (FER) than those of fish fed control (P<0.05). There had no significantly

differences in WG and FER values of fish fed with GBE (P>0.05). It seems that, in terms of growth performance use of sweet potato is effective for tilapia.

#### 4. CONCLUSION

In the present study, new information on sweet potato is provided by examining its effects on growth performance. Therefore, this is a new growth promoter for tilapia and could be a cost-effective application in aquaculture industry.

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#### REFERENCES

- Abdelhamid ,H.M.B. (2010). Physiology and nutritional studies on improving growth of Nile tilapia (*O. niloticus*) fry using some medicinal plants as a feed additives .MSc.Thesis ,University of Kafr El-Sheikh,Egypt.
- Chang, J. (2000). Medicinal herbs: drugs or dietary supplements? Biochemical Pharmacology 59, 211-219.
- Bilen S, Bulut M (2010). Effects of Laurel (Laurus nobilis) on the Non-Specific Immune Responses of Rainbow Trout (*Oncorhynchus mykiss*, Walbaum). J Anim Vet Adv 9:1275-1279.
- Bilen S, Altunoglu YC, Ulu F, Biswas, G. (2016a). Innate immune and growth promoting responses to caper (*Capparis spinosa*) extract in rainbow trout (*Oncorhynchus mykiss*). Fish Shellfish Immunology, 57:206-212
- Bilen S, Unal S, Guvensoy H (2016b) Effects of oyster mushroom (*Pleurotus ostreatus*) and nettle (*Urtica dioica*) methanolic extracts on immune responses and resistance to Aeromonas hydrophila in rainbow trout (*Oncorhynchus mykiss*). Aquaculture 454:90-94
- Kim, H. W., Kim, J. B., Cho, S. M., Chung, M. N., Lee, Y. M., Chu, S. M., et al. (2012). Anthocyanin changes in the Korean purplefleshed sweet potato, Shinzami, as affected by steaming and baking. *Food Chemistry*, 130(4), 966–972.
- Reverter, M., Bontemps, N., Lecchini, D., Banaigs, B. and Sasal, P. (2014). Use of plant extracts in fish aquaculture as an alternative to chemotherapy: current status and future perspectives, *Aquaculture*, 433 (2014) 50–61.
- Scott, G. J., Best, R., Rosegrant, M., & Bokanga, M. (2000). Roots and tubers in the global food system: A vision statement to the year 2020. Lima, Peru: International Potato Center.
- Tan, X., Sun, Z., Liu, Q., Ye, H., Zou, C., Ye, C., Wang, A. and Lin, H. (2018). Effects of dietary ginkgo biloba leaf extract on growth performance, plasma biochemical parameters, fish composition, immune responses, liver histology, and immune and apoptosisrelated genes expression of hybrid grouper (*Epinephelus lanceolatus* × *Epinephelus fuscoguttatus* ) fed high lipid diets, *Fish and Shellfish Immunology*, 72 (2018) 399-409.

Woolfe, J. (1993). Sweet potato: An untapped food resource. Cambridge: Cambridge University Press.

Yang, L., Wang, C., Ye, J., Li,H. (2011). Hepatoprotective effects of polyprenols from Ginkgo biloba L. leaves on CCl4-induced hepatotoxicity in rats, *Fitoterapia* 82 (2011) 834–840.



# Efficacy of Oregano (Origanum onites) Essential Oil Against Yersinia ruckeri and Aeromonas hydrophila

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**Abstract:** In this present study, antibacterial activity of essential oil of oregano (*Origanum onites*) was tested against the pathogenic bacteria in fish; *Yersinia ruckeri* and *Aeromonas hydrophila*. With this purpose, 150  $\mu$ l of bacterial strains were placed into 96-well plates and 8 different essential oil concentration (10  $\mu$ g/ml, 5  $\mu$ g/ml, 2.5  $\mu$ g/ml, 1.25  $\mu$ g/ml, 0.6  $\mu$ g/ml, 0.3  $\mu$ g/ml, 0.15  $\mu$ g/ml and 0.075  $\mu$ g/ml) were mixed with the culture in triplicate. The results of present study showed that essential oil of oregano was found effective against *Aeromanas hydrophila* and *Yersinia ruckeri* at all dose. Considering overall performance, oregano showed powerful antagonistic agent effect against *Aeromanas hydrophila* and *Yersinia ruckeri*.

Keywords: Aeromanas hydrophila, Yersinia ruckeri, MIC test, oregano, essential oil, fish pathogens



# Haematological Responses in Rainbow Trout Fed Diet with *Fumaria officinalis* Aqueous Methanolic Extract

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**Abstract:** In the present study, effect of *Fumaria officinalis* aqueous methanolic extract on haematological responses of rainbow trout were determined. With this aim, rainbow trout was fed with 4 different doses of the plant (0 (Control), 1%, 2% and 3%) during 75 days ad-libitum and every 15<sup>th</sup> day of the study, haematological indices were determined. The results of the study showed an decrease on RBC on 15<sup>th</sup> day of the study on FO3 group. HCT was also decreased on FO2 and FO3 group compare to control.30<sup>th</sup> day of the study all experimental groups RBC was significantly increased compared to control. Similar result were observed on HCT. On the 45<sup>th</sup> day of the study, RBC was decreased in all experimental groups. Similar result was observed on HCR. The highest RBC value was observed on FO3 group on 60<sup>th</sup> day of the study. However HGB was decreased in FO1 and 2 groups compare to control. At the end of the study both HGB and HCT were significantly decreased in all experimental groups compare to control in the study both HGB and HCT were significantly decreased in all experimental groups compare to control. If the study both HGB and HCT were significantly decreased in all experimental groups compare to control. Not suggest that usage of *Fumaria officinalis* in fish feed during long time may result with anaemia.

## **Keywords:**



# Properties of Medicinal Plants Used in Aquaculture as Immunostimulant

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Abstract: Increased world population and subsequent food consumption has led intensive aquaculture practices to emerge. As fish are cold blooded aquatic animals, they are strongly dependent on the environmental conditions. Hence, intensive culture practices put fish under stress most of the time, resulting in susceptibility to diseases. Although there are certain successful treatment or prevention methods currently used in aquaculture such as vaccines and antibiotics, these are often considered as expensive, labor intense or hazardous. Therefore, alternative treatment or prevention methods, especially medicinal plants have been the focus of the current studies. In this literature review, we aimed to give information about medicinal plant use in aquaculture, mechanism of action and constituents of the plants that are providing protection.

Keywords: Immunostimulant, immune response, fish, aquaculture, medicinal plants.

## **1. INTRODUCTION**

Obtaining high quality products in aquaculture is merely possible by providing optimum conditions for the cultured species and protecting it from diseases. Even though various methods such as vaccines, antibiotics or other chemical agents have been used successfully in fighting against fish diseases, these methods often have disadvantages such as high-cost, environmental pollution, difficulty in administration and residues in fish meat. Due to these disadvantages, the use of immunostimulants that activate the non-specific immune system as an alternative to chemotherapeutics and vaccines in aquaculture has become widespread (Aoki, 1992).

It has been reported by FAO that the number of herb species used for therapeutic purposes in the world is around 50.000 (Schippmann et al., 2002). In China, preparing extracts from plants and using them as medicines dates back to 2700 BC (Faydaoğlu and Sürücüoğlu, 2013). The use of medicinal plants in fish as well as in humans has become widespread in recent years. The reason for this is to obtain maximum yield per unit area during following years when humanity will be in downswing in terms of food resources. Medicinal plants are preferred by aquaculturists as they are natural products and often considered as cheap and easily accessible. In addition, one of the most important problems encountered in aquaculture is environmental pollution. The most threatening aspect of environmental pollution caused by aquaculture is perhaps the development of resistant bacteria as a result of antibiotic use. Medicinal plants have the potential to overcome this problem if they can substitute antibiotics.

It has been reported that feeding many different vertebrate and invertebrate species with natural immune stimulants such as plant extracts and probiotics are beneficial to aquaculture industry (Guardiola et al., 2016). Various plants such as seaweeds, herbs, spices or plant derived products have been studied in different aquatic animals (Hai, 2015). These products can promote growth and stimulate appetite, immune system and antioxidant enzymes. Also they can directly act as antimicrobial agents due to natural compounds they contain. Chemicals that plants produce such as emetine, quinine, berberine, terpenoid, alkaloid and flavonoids are currently being used in the treatment of various infectious diseases (Hussain et al., 2018; Faydaoğlu and Sürücüoğlu, 2013).

#### 2. IMMUNE STIMULATION

An immunostimulant is an agent that enhances immune response directly by itself or indirectly when coupled with an antigen (Murthy and Kiran, 2013; Mastan, 2015) resulting as a more disease resistant animal to grow (Citarasu, 2010;

Barman et al., 2013; Reverter et al., 2014). In general, immunostimulants are a group of biological and synthetic compounds that enhance the non-specific defense mechanisms in aquatic animals, thereby providing a broad spectrum protection (Murthy and Kiran, 2013). There are various studies that focus on the use of medicinal plants as immune modulators, treatment and prevention (Galina et al., 2009).

Primary defense reactions in fish are exhibited in skin and mucus. When a pathogen enters the body, humoral and cellular nonspecific immune responses are activated (Dügenci et al., 2003; Asim and Balkhi, 2015). These responses are performed by immunological cells such as myeloperoxidase, macrophages, etc. and stimulated by cytokines. Immunomodulating molecules interact with cytokines to form such reactions without needing any pathogen to be present. Plants are accepted as a major source of these immunomodulating molecules (Domenico et al., 2017).

Immunostimulants used in aquaculture are effective especially in cases where disease outbreaks can be predicted and cyclic (Syahidah et al., 2015). However, such molecules should be investigated extensively due to their potential to alter certain biological processes if used incorrectly (Citarasu, 2010; Barman et al., 2013). Immunostimulants mainly enhance the function of natural killer cells, lysozyme activity, antibody responses and phagocytic cells (Harikrishnan et al., 2011; Hai, 2015). This kind of protection may be efficient if the fish are raised in an environment where the pathogen is unknown or cannot be identified (Murthy and Kiran, 2013).

Following pathogens are successfully controlled by immunostimulants in fish or shrimp (Barman et al., 2013): *Ichthyopthirius multifiliis, Streptococcus spp., Yersinia ruckeri, Vibrio salmonicida, Vibrio vulnificus, Vibrio anguillarum, Edwardsiella ictaluri, Edwardsiella tarda, Aeromonas salmonicida, Aeromonas hydrophila* and some viruses (viral hemorrhagic septicemia, yellow head virus, infectious hematopoietic necrosis).

So far, bacterial cell wall components such as glucans and lipopolysaccharides (LPS) are considered as the best immunstimulants due to easy recognition by immune system. Especially, glucan which is a polymer of glucose found in the cell wall of bacteria, fungi and plants appears to be most promising among all examined fish and shrimp species with the best application route is found to be orally (Barman et al., 2013, Mastan, 2015, Domenico et al., 2017).

#### **3. PROPERTIES OF MEDICINAL PLANTS**

There are some plants used in aquaculture in the form of extract compounds (Hai, 2015). For example; anthraquinone extract (from *Rheum officinale Bail*), Scutellaria radix (from *Scutellaria baicalensis*) and Astragalus radix (from *Astragalus membranaceus*).

*Astragalus* root analysis showed that it contains alkaloids, flavonoids, monosaccharides and polysaccharides along with different trace minerals, cellulose, gum, mucoitin, various amino acids, folic acid, betaine and choline. Some of mentioned compounds such as glucosides, alkaloids, organic acids and polysaccharides are found to be an immune enhancer (Galina et al., 2009). Another example would be date fruit which contains phenolic constituents, anthocyanins, sterols, carotenoids, flavonoids, vitamins and various enzymes (Guardiola et al., 2016). Such herbal extracts may be applied individually, mixed or combined with other drugs or immunostimulants in order to boost immune resistance of fish (Raman, 2017).

Many different types of bioactive compounds (flavonoids, alkaloids, polysaccharides) of plants are widely studied in rats, humans, aquatic animals and proven to exhibit antimicrobial, antioxidant and immunstimulating activity (Galina et al., 2009; Pandey et al., 2012; Hai, 2015; Asim and Balkhi, 2015). In the following section, some of these compounds are examined.

#### **Phytochemicals of Medicinal Plants**

Pytochemicals are bioactive components of plants that are associated with the protection that plants provide. Although, phytochemical obtained from the plant will depend on the extraction method and plant species, in this review, we mentioned some well-known components which may be listed as tannins, terpenes, saponins, alkaloids and flavonoids.

#### Tannins

Tannins are polyphenolic constituents of plants that have two types; condensed and hydrolysable tannins (polyesters of gallic acids and various individual sugars). Tannins are shown to have antimicrobial activity against some fungi, bacteria and viruses and can enhance immune system in humans (Ergezer and Çam, 2008). However, tannins may alter digestion

due to their ability to combine with dietary proteins, minerals, polymers and digestive enzymes (Asim and Balkhi, 2015). Also, materials in aquariums such as wooden ornamentals or shelters may contain tannic acid (a specific form of tannin) which may cause change in pH or hardness of water, thus affecting fish adversely. Prusty et al. (2007) reported that hydrolysable tannin (tannic acid) supplementation for 60 days stimulated immune response in rohita fingerlings by increasing leucocyte count and respiratory burst activity without altering growth.

### Alkaloids

Alkaloids are herbal, alkaline, naturally occurring materials that consist of nitrogen, hydrogen carbon and generally oxygen. They are found in many plant species, mainly in some flowering plants (Hussain et al., 2018; Bakırel, 1998, Yılmaz et al., 2006) Alkaloids are colorless, odorless, sour-tasting, in liquid form under normal temperature, and crystallized except coniine and nicotine (Yılmaz et al., 2006). Some of well-known and commercial alkaloids are nicotine, caffeine and morphine (Hussain et al., 2018). Sharma et al. (2010) studied the effects of *Withania somnifera* root powder supplementation on immunological parameters and disease resistance to *Aeromonas hydrophila* in *Labeo rohita*. They observed increased oxidative radical production, phagocytic activity, total immunoglobulin count in plasma, lysozyme activity in plasma and high survival rate against *A. hydrophila* in experimental groups. Sharma et al. suggested that this beneficial effect might have happened due to alkaloids –along with other components- that *W. somnifera* plant contains.

#### Terpenes

Terpenes are the most common compounds among natural compounds and are small organic molecules with various structures. Although, there are terpenes that contain only carbon and hydrogen, they might contain molecules consisting of oxygen such as alcohol, ketone, aldehyde and acid groups. Oxygen-containing terpenes are called "terpenoids". Today, more than 20.000 terpene structures (either open-chain or cyclic structure) with various functional groups, are known (Kılıç, 2007) Terpenes are considered to be the most important group of volatile oils (Arslan and Karabulut, 2005). β-carotene and thymol are some terpenes that are synthesized by plants and known to have beneficial effects such as antimicrobial, antiseptic and antioxidant activity (Çöl, 2007).

#### Saponins

Saponins are naturally occurring surface glycosides that are synthesized by mainly plants. They can also be produced by some aquatic organisms and bacteria. To date, there are two types of saponins are identified; steroid and triterpenoid saponins. Steroid saponins are present in medicinal plants whereas triterpenoid saponins are found in legumes such as peas, beans and soybean (Asim and Balkhi, 2015). Saponins have antimicrobial effect and are reported to exhibit antioxidant, anti-inflammatory and immune enhancer properties (Kocaoğlu Güçlü and Uyanık, 2004). Considerable amount of studies indicate that saponins are toxic to fish. However, there are some studies reporting their beneficial effects such as growth promotion (Stadtlander, 2012).

#### Flavonoids

Flavonoids are the most studied compounds of the plants. They are water soluble, polyphenolic molecules that contain fifteen carbon atoms. Five major subgroups of flavonoids are Anthocyanidins, flavanols, flavanones, flavonoids and flavones. Flavonoids play major role in scavenging reactive oxygen species and exhibit high antioxidant activity by means of their hydroxyl content (Asim and Balkhi, 2015). Studies have shown that flavonoids show anti-inflammatory, antiviral, antiallergic and antithrombotic effects besides their antioxidant properties. Flavonoids, of which number of members are estimated to be over 4000, are abundant in tea, apple, onion, legumes, tomato and grape (Kahraman et al., 2002).

#### 4. CONCLUSION

Although plants and their extracts considered to be safe and used widely, there are some studies reporting their negative effects in fish culture. E.g. garlic (*Allium sativum*) was reported to exhibit beneficial effects in many studies in terms of immune response and disease control. On the other hand, it was shown to have harmful and even lethal effect on silver carp larvae. Abdel-Hadi et al. (2008) studied the effects of garlic in silver carp larvae and concluded that 4 g/L of garlic killed all larvae. Similarly, García Beltrán et al. (2017) showed that dietary dehydrated lemon (*Citrus limon*) peel in the ratio of 3% reduced weight gain and specific growth rate of fish (*Sparus aurata*) when fed for 30 days. Moreover, application method, dose and duration also play an important role on effect of the plant product.

Action mechanism and effects of herbs or their compounds, as well as immune stimulation mechanisms are not fully understood yet. However, it can be said that beneficial effects of herbal products in terms of immunity occur in two different ways: either directly by bioactive components of plant exhibiting antimicrobial, antioxidant activity; or indirectly by constituents of plant getting recognized by fish immune system as extraneous materials, resulting in activation of immune response. Furthermore, in order to maximize the benefit from medicinal plants, we suggest that future studies need to evaluate far-going use of medicinal plants of which beneficial effects have already supported with literature data, in terms of economy and long-term utilization.

#### REFERENCES

- Abdel-Hadi, Y. M., Saleh, O. A. & Akar, A. M. (2008). Study on the use of Artemisia cina L. (wormseed plants) and Allium sativum (garlic) in the control of Saprolegniosis in egg of Cyprinus carpio (common carp) and Hypophthalmichthys molitrix (silver carp). The 30<sup>th</sup> Malaysian Symposium on Microbiology (MSM). Hyatt Regency Resort, Kuantan, Malaysia, August 16-19, 2008.
- Aoki, T. (1992). Chemotherapy and drug resistance in fish farms in Japan. In: Shariff M, Subasighe RP, Arthur JR, (Eds.). Diseases in Asian Aquaculture Fish Health Section, Asian Fisheries Society, Manila, Philippines, 1: 519–529.
- Arslan, Ü., & Karabulut, Ö. A. (2005). Baharat Bitkilerinin Bitki Patojeni Funguslara Karşı Antifungal Etkisi. Atatürk Üniv Ziraat Fak Derg, 36(2):131-135.
- Asim, O. A., & Balkhi, M. H. (2015). Commonly Available Herbs in Kashmir to Be Used As Fish Feed Additive. International Technology and Innovation Research Journal, 1(2):1-5.
- Bakırel, T. (1998). Veteriner Toksiloji Yönünden Trakya Bölgesi'nin Zehirli Bitkileri Üzerine Çalışmalar. İ.Ü. Sağlık Bilimleri Enstitüsü Farmakoloji ve Toksikoloji Ana Bilim Dalı, Doktora Tezi, İstanbul.
- Barman, D., Nen, P., Mandal, S. C., & Kumar, V. (2013). Immunostimulants for Aquaculture Health Management. Marine Science Research & Development, 3(3). doi:10.4172/2155-9910.1000134
- Citarasu, T. (2010). Herbal biomedicines: a new opportunity for aquaculture industry. Aquaculture International, 18:403-414.
- Çöl, Ç. (2007). SİDERİTİS TMOLEA P. H. DAVİS BİTKİSİNİN DİTERPEN BİLEŞENLERİNİNİ İZOLASYONU VE YAPILARININ TAYİNİ. Balıkesir University, Msc. dissertation, Balıkesir.
- Domenico, J. D., Canova, R., Soveral, L. F., Nied, C. O., Costa, M. M., Rafael Frandoloso, R., & Kreutz, L.C. (2017). Immunomodulatory effects of dietary β-glucan in silver catfish (*Rhamdia quelen*). Pesq Vet Bras 37(1):73-78.
- Dügenci, S. K., Arda, N., & Candan, A. (2003). Some medicinal plants as immunostimulant for fish. Journal of Ethnopharmacology, 88:99-106.
- Ergezer, H., & Çam, M. (2008). Tanenler: Sınıflandırma, Yapıları ve Sağlık Üzerine Etkileri. Türkiye 10. Gıda Kongresi, 21-23 Mayıs 2008, Erzurum.
- Faydaoğlu, E., & Sürücüoğlu, M. S. (2013). MEDICAL AND AROMATIC PLANTS' ANTIMICROBIAL, ANTIOXIDANT ACTIVITIES AND USE OPPORTUNITIES. EÜFBED Fen Bilimleri Enstitüsü Dergisi, 6(2):233-265.
- Galina, J., Yin, G., Ardo, L., & Jeney, Z. (2009). The use of immunostimulating herbs in fish: An overview of research. Fish Physiol Biochem, 35:669-676.
- García Beltrán, J. M., Espinosa, C., Guardiola, F. A., & Esteban, M. Á. (2017). Dietary dehydrated lemon peel improves the immune but not the antioxidant status of gilthead seabream (*Sparus aurata* L.). Fish & Shellfish Immunology, 64:426-436. doi:10.1016/j.fsi.2017.03.042
- Guardiola, F.A., Porcino, C., Cerezuela, R., Cuesta, A., Faggio, C., & Esteban, M.A., (2016). Impact of date palm fruits extracts and probiotic enriched diet on antioxidant status, innate immune response and immune-related gene expression of European sea bass (*Dicentrarchus labrax*). Fish & Shellfish Immunology, 52:298-308.
- Hai, N. V. (2015). The use of medicinal plants as immunostimulants in aquaculture: A review. Aquaculture, 446: 88-96.
- Harikrishnan, R., Balasundaram, C., & Heo, M. S. (2011). Impact of plant products on innate and adaptive immune system of cultured finfish and shellfish. Aquaculture, 317:1-15.
- Hussain, G., Rasul, A., Anwar, H., Aziz, N., Razzaq, A., Wei, W., Ali, M., Li, J & Li, X. (2018). Role of Plant Derived Alkaloids and Their Mechanism in Neurodegenerative Disorders. International Journal of Biological Sciences, 14(3):341– 357. doi:10.7150/ijbs.23247

Kahraman, A., Serteser, M., & Köken, T. (2002). Flavonoidler. Kocatepe Tıp Dergisi, 3:1-8.

- Kılıç, E. (2007). *Tanacetum zahlbruckneri* (Nãb.) Grierson BİTKİSİ ÜZERİNDE FİTOKİMYASAL ARAŞTIRMALAR. Yıldız Teknik University, Msc. dissertation, İstanbul.
- Kocaoğlu Güçlü, B., & Uyanık, B. (2004). Saponinler ve Biyolojik Önemi. Erciyes Üniv Vet Fak Derg, 1(2):125-131.
- Mastan, S. A. (2015). Use of Immunostimulants in aquaculture disease management. International Journal of Fisheries and Aquatic Studies, 2(4):277-280.

- Murthy, K. S., & Kiran, B. R. (2013). Review on Usage Of Medicinal Plants In Fish Diseases. International Journal of Pharma and Bio Sciences, 4(3):975-986.
- Pandey, G., Sharma, M., & Mandloi, A. K. (2012). Medical Plants Useful In Fish Diseases: Plant Archives, 12(1):1-4.
- Prusty, A. K., Sahu, N. P., Pal, A. K., Reddy, A. K., & Kumar, S. (2007). Effect of dietary tannin on growth and haematoimmunological parameters of *Labeo rohita* (Hamilton) fingerlings. Animal Feed Science and Technology, 136(1-2):96-108. doi:10.1016/j.anifeedsci.2006.08.023
- Raman, R. P. (2017). Applicability, Feasibility and Efficacy of Phytotherapy in Aquatic Animal Health Management. American Journal of Plant Sciences, 8:257-287.
- Reverter, M., Bontemps, N., Lecchini, D., Banaigs, B., & Sasal, P. (2014). Use of plant extracts in fish aquaculture as an alternative to chemotherapy: Current status and future perspectives. Aquaculture, 433:50-61.
- Schippmann, U., Leaman, D., & Cunningham, A. B. (2002). Impact of Cultivation and Gathering of Medicinal Plants on Biodiversity: Global Trends and Issues. In book: Biodiversity and the ecosystem approach in agriculture, forestry and fisheries, Chapter: Impact of cultivation and gathering of medicinal plants on biodiversity: Global trends and issues, Publisher: FAO, Editors: FAO, pp.142-167.
- Sharma, A., Deo, A. D., Tandel Riteshkumar, S., Chanu, T. I., & Das, A. (2010). Effect of Withania somnifera (L. Dunal) root as a feed additive on immunological parameters and disease resistance to Aeromonas hydrophila in Labeo rohita (Hamilton) fingerlings. Fish & Shellfish Immunology, 29(3):508-512. doi:10.1016/j.fsi.2010.05.005
- Stadtlander, T. (2012). Effects of saponin fractions from Fenugreek (*Trigonella foenum-graecum*) and the desert date (*Balanites aegyptiaca*) in diets for Nile tilapia. Hohenheim University, PhD. dissertation, Bremen.
- Syahidah, A., Saad, C. R., Daud, H. M., & Abdelhadi, Y. M. (2015). Status and potential of herbal applications in aquaculture. A review. Iranian Journal of Fisheries Sciences. 14(1):27-44.
- Yılmaz, H., Akpınar, E., & Yılmaz, H. (2006). PEYZAJ MİMARLIĞI ÇALIŞMALARINDA KULLANILAN BAZI SÜS BİTKİLERİNİN TOKSİKOLOJİK ÖZELLİKLERİ. Süleyman Demirel Üniversitesi Orman Fakültesi Dergisi, 1:82-95.



# Investigation on Anti-Fungal Effect of Garlic (*Allium sativum*) Skin Aqueous Methanolic Extract in Rainbow Trout (*Oncorhynchus mykiss*) Eggs

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**Abstract:** In this preliminary study, it was aimed to determine formation inhibitor effects of garlic (*Allium sativum*) skin extract on fungal disease (*Saprolegnia parasitica*) which occurs on rainbow trout (*Oncorhynchus mykiss*) eggs. In order to evaluate fungicidal effect on *S. parasitica*, obtained aqueous methanolic extract was applied to eggs right after fertilization via bathing method in concentrations of 0.1, 0.2, 0.4, 0.8, 1.6 and 3.2 g l<sup>-1</sup>. In the trial, experimental groups showed positive results in terms of preventing formation of the fungus in comparison with control group where, best result was seen in 0.4 g l<sup>-1</sup> group (P<0.05). According to results, we conclude that garlic skin may be used in rainbow trout hatcheries as an alternative natural fungicidal.

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Keywords: Garlic, Allium sativum, Oncorhynchus mykiss, Saprolegnia parasitica, disease, fungicide



# Pharmacokinetic/Pharmacodynamic Integration of Marbofloxacin after Oral and Intravenous Administration in Rainbow Trout (*Oncorhynchus mykiss*)

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**Abstract:** The pharmaco-kinetic/dynamic of marbofloxacin was investigated after single intravenous (IV) and oral administration of 10 mg/kg in 192 healthy rainbow trout at  $13\pm1.2$  °C. The plasma concentrations of marbofloxacin were determined by high-performance liquid chromatography-ultraviolet detection. After IV and oral administration, the plasma concentration—time data were described by a noncompartmental analysis. The minimal inhibitory concentration (MIC) of marbofloxacin against *Yersinia ruckeri*, *Aeromonas hydrophila*, *Pseudomonas fluorescens* and *P. putida* were determined by broth dilution method at 13 °C. After IV administration, the elimination half-life ( $t_{1/25z}$ ), area under the concentration-versus time curve (AUC<sub>0-∞</sub>), apparent volume of distribution at steady-state and total body clearance of marbofloxacin were 18.05 h, 354.63 h\*µg/mL, 0.65 L/kg and 0.03 L/h/kg, respectively. After oral administration,  $t_{1/25z}$ , AUC<sub>0-∞</sub>, the peak plasma concentration, time of maximum concentration and bioavailability were 27.51 h, 135.29 h\*µg/mL, 3.74 µg/mL, 4 h and 38.15%, respectively. The respective MICs of marbofloxacin against *Y. ruckeri*, *A. hydrophila*, *P. fluorescens* and *P. putida* were determined as 0.02 µg/mL, 2.5 µg/mL, 2.5 µg/mL and 5 µg/mL, respectively. Following IV and oral administration of 10 mg/kg marbofloxacin, AUC/MIC and C<sub>max</sub>/MIC values were above the target levels for *Y. ruckeri*, while this dose was not sufficient for *A. hydrophila* and *Pseudomonas* spp. Because the pharmacokinetics and pharmacodynamics of a drug in fish are significantly affected by temperature, the dosage regimen of marbofloxacin should be modified according to temperature.

Keywords: Pharmacokinetics, pharmacodynamics, marbofloxacin, rainbow trout



# Evaluation of The Persimmon, Cherry Laurel And Likapa As A Natural Antioxidant Source

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Abstract: Oxygen is vital for living organisms. But it can be poisonous and cause mutation for living things. Consumption of oxygen by the cells causes the formation of reactive oxygen species (ROS). It is reported that the progression of this condition, which leads to an imbalance between antioxidant systems with excessive amounts of ROSs leads to oxidative stress, which leads to chronic and acute diseases such as cardiovascular diseases, cancer, diabetes, alzheimer and some immune problems and aging. When antioxidants are present at low concentration, they are a substance that retards or prevents oxidation according to oxidizable substances. Epidemiological data show that antioxidant-rich nutrition is beneficial and can play a major role in the prevention of diseases. Therefore, there are many researches on natural antioxidants.

This study was carried out to determine the antioxidant capacity of the extracts obtained from the leaves of persimmon, cherry laurel and likapa plants growing in and around Rize. For this purpose, *Diospyros kaki* (persimmon), *Prunus lauroceraus* (cherry laurel), *Vaccinium arctostaphylos* (likapa) leaves are dried at 60 ° C with ethanol (40 ° C), hot water (75 ° C) and warm water (40 ° C). extract extracts were obtained in 2.5% and 5% extractions. DPPH method was used to determine the antioxidant effect. According to the results, the rate of antioxidant activity was determined mostly in lichen leaf extract in the warm water extract and in the lowest amount of ethanol extract. It is clear that extract concentrations do not affect the antioxidant activity.

Keywords: Antioxidant, ROS, herbal extract, DPPH



# Haematological Indices in Rainbow Trout Fed with *P.latifolia* Extract

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**Abstract:** This study evaluated the effects of *P.latifolia* methanolic extracts on heamatological indices in rainbow torut (*Oncorhyncus mykiss*). The fish were fed with a diet containing 0.1, 0.5 and 1 % methanolic extract of *P. latifolia* leaves and seeds. At the end of the experimental period, hematological parameters including red blood cell (RBC), hemoglobin (Hb), heamatocrit (Hct), mean cell volume (MCV), mean cell hemoglobin (MCH) and mean cell hemoglobin concentration (MCHC). The results obtained demonstrate that fishes with the supplement of *P.latifolia* leaves and seeds showed significant immunostimulatory effect, increase in RBC, Hb, Hct, MCV, MCH and MCHC values, when compared with the control group (P<0.05).

Keywords: Haematological indices, P. latifolia, rainbow trout

## **1. INTRODUCTION**

Rainbow trout (Oncorhynchus mykiss) is the most commonly cultivated cold water fish in aquaculture industry. Owing to intensive culture practices for the increased production, disease management continues to pose a serious threat to aquaculture industry (Haghighi and Rohani, 2013). The use of immunostimulants in aqua-feed is considered as a modern and promising alternative to antibiotics and vaccines as a prophylactic measure in intensive aqua-culture (Raa, 2000). Herbal (medical and aromatic) immunostimulants are substances which activate white blood cells (WBC) and may render fishes more resistant to infectious diseases, by the stimulating phagocytic cells as well as complement lysozyme and antibody responses of fish (Secombes and Olivier, 1997). A haematological examination of intensively farmed fish is an integral part of evaluating their health status. Rainbow trout farming is among the important for aquaculture and it is important to improve continuously the methods for evaluating the health of trout (Rehlka et al., 2003). And so must be the ranges for the various haematological parameters considered to be characteristic of normal healthy fish. Intensive aquaculture conditions have placed increasing demands on the fish, which must be able to cope with numerous stress factors that affect their basic physiological functions. Assessment of the level of this exposure should be based on synthetic and analytical investigation of an extensive database of laboratory test values relating to fish populations, vitally dependent on specific environmental conditions (Bilen et al., 2016).

The aim of the present study was to evaluate the haematological effects of dietary intake of *P.latifolia* extract in rainbow trout.

## 2. MATERIAL AND METHOD

## **Experimental Design and Fish**

Juvenile rainbow trout (*Oncorhynchus mykiss*), with a mean initial body weight of  $40,99 \pm 1.69$  g, were obtained and stocked randomly (50 fish tank<sup>-1</sup>) into 12 mesh 1,5 x 1,5 m cages capacity in the Germeçtepe Freshwater Fish Production, Application and Research Center of Faculty of Fisheries, Kastamonu University, Turkey. Water temperature, dissolved oxygen and pH were measured by multiparametre. The freshwater having an average temperature of  $12.3 \pm 0.2$  °C. Dissolved oxygen was maintained around  $8.1 \pm 0.1$  mg L<sup>-1</sup>. The experiment was designed as three replicates of each treatment. Before starting the experiment, fish were acclimatized to the experimental feeding regimen using a commercial diet for 2 weeks. Aqueous methanolic extracts of *P. latifolia* leaves (PLL) and seeds (PLS) were added to the fishes' basal diet at the rate of 0 (Control), 0.1 (PLL0.1 and PLS0.1), 0.5 (PLL0.5 and PLS0.5) and 1 (PLL1 and PLS1) g kg<sup>-1</sup> by spraying. All groups were fed with the diets twice a day ad-libitum.

### **Preparation of Aqueous Methanolic Extract**

*P. latifolia* leaves and seeds were purchased from market and dried under shade in natural conditions; extracts of *P. latifolia* leaves and seeds were prepared according the procedure described by Bilen et al. (2016).

## **Haematological Parameters**

Blood was analyzed with routine methods adopted in fish hematology (Blaxhall and Daisley, 1973; Ivanova, 1993; Haghighi, 2010). All blood samples (Red blood cell, RBC; Hemoglobin, Hb; Heamatocrit, Hct; Mean cell volume, MCV; Mean cell hemoglobin, MCH and Mean cell hemoglobin concentration, MCHC) values were measured by Auto Hematology Device (BC-3000plus).

## **Statistical Analysis**

Statistical analyses of data were subjected to one-way ANOVA, and a subsequent comparison of means by Tukey's multiple range test. All of the above mentioned statistical analyses were performed using SPSS (23.0). Differences were considered statistically significant at P<0.05.

## 3. RESULTS AND DISCUSSION

Hematological indices of the fish groups at the  $15^{th}$ ,  $45^{th}$  and  $75^{th}$  days of the experiment are shown in Figure 1. Adding *P. latifolia* exracts to diets had effected on red blood cell, hemoglobin, heamatocrit, mean cell volume, mean cell hemoglobin and mean cell hemoglobin concentration (P<0.05).



Figure 1. Dynamics of the all hematological parameters of the experiment groups during 15<sup>th</sup>, 45<sup>th</sup> and 75<sup>th</sup> days.

Ahmadifar et al. (2011) studied the effects of thymol and carvacrol on growth, hematological parameters and tissue composion of juvenile rainbow trout. Their results showed that adding the diet had no effect on Ht, Hb concentration, RBC, WBC, MCV, MCH or MCHC, whereas supplementation with thymol and carvacrol caused an increase in lymphocyte percent (P<0.05).

Hematological examination showed that *P.latifolia* a certain influence on some of the blood indices studied. In addition to its importance for producingan attractive, *P.latifolia* in the diet for the rainbow trout may also significantly influence certain physiological functions, especially hematopoiesis.

#### 4. CONCLUSION

In the present study, new information on *P.latifolia* is provided by examining its effects on hematological parameters.

#### REFERENCES

- Ahmadifar, E., Falahatkar, B. and Akrami, R. (2011). Effects of dietary thymol-carvacrol on growth performance, hematological parameters and tissue composition of juvenile rainbow trout, *Oncorhynchus mykiss, Journal of Applied Ichthyology*, 27(2011), 1057-1060.
- Bilen S, Unal S, Guvensoy H (2016) Effects of oyster mushroom (*Pleurotus ostreatus*) and nettle (*Urtica dioica*) methanolic extracts on immune responses and resistance to Aeromonas hydrophila in rainbow trout (*Oncorhynchus mykiss*). Aquaculture 454:90-94.
- Blaxhall, P. C. and Daisley, K. W. (1973). Routine haematological methods for use with fish blood. *Journal of fish biology*, 5(6), 771-781.
- Haghighi, M. (2010). Laboratory Methodes of Fish Hematology. Aquaculture Science Publications, Tehran, Iran, pp. 29-71.
- Haghighi, M. and Rohani, M.S. (2013). The effects of powdered ginger (Zingiber officinale) on the haematological and immunological parameters of rainbow trout *Oncorhynchus mykiss*, Journal of Medicinal Plant and Herbal Therapy Research, 1(2013):8-12.
- Ivanova, N.T. (1993). Atlas of Fish Blood Cells. LPP, Mosacow, Russia (In Russian).
- Raa J. (2000). The use of immunestimulants in fish and shellfish feeds. In: L. E. Cruz-Suarez, D. Ricque-Marie, M. Tapia-Salazar, M.
   A. Y. Olvera-Novoa and R. Civera-Cerecedo (Eds.). Avances en Nutrición Acuícóla V. Memorias del V Simposium International de Nutrición Acuícóla. Merida, Yucatan, Mexico. pp. 47-56.
- Rehulka J. (2003) Haematological and biochemical analyses in rainbow trout, *Oncorhynchus mykiss* affected by viral haemorrhagic septicaemia (VHS). Diseases of Aquatic Organisms 56,185-193.

Secombes C.J. & Olivier G. (1997). Furunculosis. Academic Press, New York, pp. 269-296.



# Environmental Monitoring of Heavy Metals by Common Rhodophyta Species in Sinop Coastal Ecosystem, Turkey

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Abstract: Sinop, located in the northeast part of the Black Sea, is potentially threatened by domestic and industrial wastes. The aim of the present study was to monitor the heavy metal pollution in phylum Rhodophyta macroalgae species associated with sediment and water along Sinop coastal waters. Samples collected from eight different stations in 2015-2016. For this purpose, five dominant macroalgae species (*Ceramium* spp., *Corallina officinalis, Gelidium crinale, Laurencia obtusa, Vertebrata fucoides*) were sampled, and the levels Al, Mn, Fe, Co, Ni, Cu, Zn, As, Cd, Hg, Pb were measured by ICP-MS. The bio-accumulated essential Al and Fe concentrations were found highest in all species and also coastal waters (BCF≥1000), resulting from human activities. Bio-indicator elements Zn and Cd showed higher BSAF (>1) values presented increased pollution pressure in the study area. The mean concentrations of elements were found less than suggested acceptable values. However, in order to assess the impact of pollutants and take necessary actions to anthropogenic, agricultural and industrial contamination, the region need to be monitored regularly.

Keywords: Sinop Province, Rhodophyta, heavy metals, monitoring, bio-indicator

## **1. INTRODUCTION**

Sinop Province, located in the northeast part of the Black Sea, is potentially threatened by uncontrolled domestic and industrial waters (Bat et al. 2018). Biological marine indicators are routinely used for monitoring extent of pollution in aquatic environments in order to assess the impact of heavy metals and take necessary actions to ensure contaminations.

Macroalgae have been used widely to determine heavy metal pollution in marine ecosystems. Metals in marine macroalgae influence the chemical capacity of higher trophic levels and become threat to human and environmental health. Marine Strategy Framework Directive (MSFD) is aimed to ensure Good Environmental Status (GES) of the seas by 2020 in the EU, using 11 qualitative descriptors (MSFD, 2008). For this purpose, to determine toxic element pollutants is necessary in macroalgae species regionally with surrounding water and sediment.

There is no extensive literature on accumulation of heavy metals using red algae in the Black Sea. Reviews concerning the use of macroalgae including Rhodophyta phylum studies to monitor coastal waters are: Güven et al., 1992, 1998; Öztürk et al., 1994a,b, 1996; Bat and Öztürk, 1997; Topçuoğlu et al., 1998a,b,c; 2002, 2003, 2010; Kut et al., 2000; Bat et al., 2009, 2015; Türk Çulha et al., 2013; Öztekin, 2015; Bat and Arıcı, 2016; Arıcı and Bat, 2016, Arıcı 2017. In this study, heavy metal accumulation of *L. obtusa* and *V. fucoides* was reported for the first time in the Black Sea of Turkish coasts.

The levels of Al, Mn, Fe, Co, Ni, Cu, Zn, As, Cd, Hg and Pb in *Ceramium* spp., *Corallina officinalis, Gelidium crinale, Laurencia obtusa* and *Vertebrata fucoides* were determined to ensure sufficient increased pollution control.

## 2. MATERIAL AND METHODS

To assess of elemental uptakes, we investigated the concentrations of 11 heavy metals in red macroalgae, water and sediments along Sinop coasts (Figure 1). Five species of dominantly found in phylum Rhodophyta (*Ceramium* spp., *Corallina officinalis, Gelidium crinale, Laurencia obtusa* and *Vertebrata fucoides*) were analyzed during September 2015 and July 2016.



Figure 1. Study areas

Al, As, Cd, Co, Cu, Fe, Hg, Mn, Ni, Pb and Zn were examined with ICP-MS (Agilent Technologies, 7700X) as mg/kg dry wt. in algae and sediments, and ppb in water samples. Aquatic plant HPR-FO-08 method were used for microwave digestion system (Milestone Systems, Start D 260). The less than 63 µm of sediment samples were digested with Seawater Sediment HPR-EN-33 methodology (Milestone Systems, Start D 260). The accuracy and precision was tested by the Certified Reference Materials (CRMs) (BCR 279-*Ulva lactuca*) at 99% confidence level.

Variability in Biota-Sediment Accumulation Factor (BSAF) and Bio-Concentration Factor (BCF) data was evaluated in relation to metal accumulation from sediments and waters calculating the following equation: BSAF = [C] biota/ [C] sediment; BCF = [C] biota/ [C] water (Geyer et al., 2000; Kleinov et al., 2008). SPSS ver. 21.0 was used for statistical calculations, and the p value  $\leq 0.05$  was considered to indicate statistical significance.

## **3. RESULTS AND DISCUSSION**

#### Results

Predominant species in phylum *Rhodophyta* (*Ceramium* spp., *Corallina officinalis*, *Gelidium crinale*, *Laurencia obtusa* and *Vertebrata fucoides*) were sampled from different stations to be comparable along Sinop coasts are shown in Table 1.

| Species     | Stations |         |           |         |         |         |     |       |  |  |  |
|-------------|----------|---------|-----------|---------|---------|---------|-----|-------|--|--|--|
|             | Türkeli  | Ayancık | Inceburun | Akliman | Tersane | Karakum | DSI | Gerze |  |  |  |
| Ceramium    | v        | v       | v         | v       | v       | v       | v   | v     |  |  |  |
| spp.        | Λ        | Λ       | Λ         | Λ       | Λ       | Λ       | Λ   | Λ     |  |  |  |
| Corallina   |          |         |           |         |         | Y       |     |       |  |  |  |
| officinalis |          |         |           |         |         | Λ       |     |       |  |  |  |
| Gelidium    |          | v       |           | v       | v       |         | v   | v     |  |  |  |
| crinale     |          | Λ       |           | Λ       | Λ       |         | Λ   | Λ     |  |  |  |
| Laurencia   |          |         |           |         |         | v       |     |       |  |  |  |
| obtusa      |          |         |           |         |         | Λ       |     |       |  |  |  |
| Vertebrata  |          |         |           |         |         |         | v   |       |  |  |  |
| fucoides    |          |         |           |         |         |         | Λ   |       |  |  |  |

 Table 1. Sampled red algae species along Sinop coasts

Table 2 shows the mean concentrations of sea water and sediments and each sampling location compared to the relevant guideline values for marine waters and sediments.

BCF values were given in Table 3 and varied in the range between 1000 and 5000.

The results pointed out a high interspecific variability of metal levels among different species depending on on morphology, ecology, plant structure and the binding sites available on the algae (Lobban and Harrison, 1994).

Al: *L. obtusa < C. officinalis < G. crinale < V. fucoides < Ceramium* spp.

Mn: L. obtusa < C. officinalis < Ceramium spp. < V. fucoides < G. crinale

Fe: L. obtusa < C. officinalis < G. crinale < V. fucoides < Ceramium spp.

Co: *L. obtusa < C. officinalis < V. fucoides < G. crinale < Ceramium* spp.

Ni: C. officinalis < L. obtusa < V. fucoides < Ceramium spp. < G. crinale

Cu: C. officinalis < L. obtusa < V. fucoides < Ceramium spp. < G. crinale

Zn: C. officinalis < L. obtusa < Ceramium spp. < G. crinale < V. fucoides

As: *L. obtusa < C. officinalis < G. crinale < V. fucoides < Ceramium* spp.

Cd: G. crinale < C. officinalis < L. obtusa < Ceramium spp. < V. fucoides

Hg: *C. officinalis < L. obtusa < G. crinale < V. fucoides < Ceramium* spp.

Pb: L. obtusa < C. officinalis < G. crinale < Ceramium spp. < V. fucoides

Heavy metal concentrations in macroalgae species and bioaccumulation factors were given in Table 4

Al Ni Zn Cd Pb Mn Fe Co Cu As Hg Seawater Türkeli 1 251.88 457.58 6.84 3.25 1.61 0.38 0.03 2.7123.11 1.53 274.33 Türkeli 2 206.82 33.52 348.87 1.31 3.72 224.25 1.82 0.32 0.03 1.81 5.08 511.33 1.71 191.86 0.41 Ayancık 446.12 20.06 1.65 4.8 1.65 0.05 1.41 Akliman 208.55 12.91 285.13 1.53 4.23 2.88 368.58 1.56 0.46 0.04 6.2 Karakum 171.55 8.55 180.26 1.48 3.22 1.39 181.2 1.5 0.41 0.04 1.23 Tersane 258.94 13.22 260.19 4.69 17.56 508.71 1.55 0.04 1.58 0.64 6.61 1117.74 1557.78 3.46 DSI 64.76 2.14 5.62 2.04 344.46 3.56 0.44 0.27 Gerze 378.35 23.79 472.58 1.5 4.34 1.58 224.62 1.64 0.37 0.03 1.73 217.92 Inceburun 164.01 7.33 151.19 1.47 3.66 1.58 1.54 0.4 0.04 1.48 EC, 1998 200 2000 5 1 10 50 --\_ \_ \_ 200 TS-266, 2005 50 \_ 5 10 EPA, 2008 200 50 300 1000 10 5 2 15 \_ \_ WHO, 2011 100 100 300 20 2000 10 3 10 1 Sediment Türkeli 1 5724 516 10393 4.5 16.9 4.6 17.1 7.4 0.08 0.03 3.7 Türkeli 2 4321 325 3.5 18.2 9 16.7 0.05 0.02 3 7041 4.1 5205 420 4.5 7 0.05 0.02 3.7 Ayancık 10617 16.4 4.4 18.7 Akliman 3584 333 7260 2.7 7.5 4.6 21.1 5.1 0.1 0.01 39.3 0.42 Karakum 400 12.2 613.7 0.22 0.33 0.54 0.79 0.19 0.003 0.005 Tersane 1348 85.9 2533 0.9 2.9 1.9 6 2.3 0.02 0.01 2.2 DSI 2157 97.2 3783.8 2 6.5 1.5 5.4 3.6 0.02 0.01 1.9

Table 2. Mean heavy metal concentrations in seawater (ppb) and sediments (mg/kg dry wt.)

| Gerze      | 5010 | 231      | 8300 | 3.3 | 13        | 5.7    | 13.2    | 3.6    | 0.04    | 0.01      | 1.7    |
|------------|------|----------|------|-----|-----------|--------|---------|--------|---------|-----------|--------|
| NOAA, 2009 |      |          |      |     | 20,9-51,6 | 34-270 | 150-410 | 8,2-70 | 1,2-9,6 | 0,15-0,71 |        |
| CSQG, 2001 |      | 460-1100 | %2-4 |     | 16-75     | 16-110 | 120-820 |        | 0.6-33  |           | 31-250 |

\*Relevant guideline values for marine waters were given.

Table 3. An overview of regulatory bioaccumulation assessment endpoints and criteria

| Program  | Criteria                      |
|--|-------------------------------|
| Toxic Substances Control Act (TSCA) and Toxic Release Inventory (TRI) (USEPA, 1976)                        | 1000-5000 (bio-accumulative)  |
|  | ≥5000 (very bio-accumulative) |
| Canadian Environmental Protection Act (CEPA, 1999)   | ≥5000                         |
| Registration, Evaluation and Authorization of Chemicals Annex XII (European Commission 2001) (REACH, 2001) | ≥2000 (bio-accumulative)      |
|  | ≥5000 (very bio-accumulative) |
| Stockholm Convention on Persistent Organic Pollutants (UNEP, 2001)   | ≥5000                         |

#### Discussion

The mean concentrations of sea water and sediments were varied. Concentrations of Co, Ni and Pb exhibited variations depending on seasons in seawaters; and Mn, Cd, and Hg levels displayed statistical significance depending upon locations (ANOVA,  $p \le 0.05$ ). The metal concentrations of seawaters were increased in the order of Hg < Cd < Co < As = Pb < Cu < Ni < Mn < Al < Zn < Fe and the heavy metal content in sediments were Hg < Cd < Pb < Co = As < Cu < Ni < Zn < Mn < Al < Fe.

In the present study each sampling location compared to the relevant guideline values for marine waters and sediments. The concentrations of non-essential elements (As, Cd, Cu, Hg, Ni, Pb) that are able to toxicity even at lower levels are below the detection limit values (EC, 1998; CSQG, 2001; TS-266, 2005; EPA, 2008; NOAA, 2009; WHO, 2011).

In the present study BCFs were found in the range between 1000 and 5000 (see Table 3) Biota-Sediment Accumulation Factors (BSAFs) for macroalgae are in the range of 1 to 2; BSAF>2 macro concentrator, 1<BSAF<2 micro concentrator and BSAF<1 deconcentrator are named as (Nenciu et al., 2016). However, there is no criteria have been explicitly developed for evaluating the quality of Bio-Concentration Factor (BCF) by regulatory agencies (Arnot and Gobas, 2006).

When the air temperature oscillated between 15-19  $^{\circ}$ C, *G. crinale* was also found more than other seasons in Tersane station. Accumulation Mn, Ni and Cu in this species depends on high ship traffic and reflects the discharges of wastewaters into sediments. In the last decade, the population is three times higher in summer seasons. Thus, it is showed that with the increase of urban wastes, the contamination of heavy metals is also increased.

The concentrations of mean Al in filamentous macroalgae *Ceramium* spp. and *V. fucoides* had differentiation between seasons (p=0.03,  $p\leq0.05$ , ANOVA) and the highest values were found in Akliman station during winter originating from municipial wastes. The essential elements Al and Fe are exist in seawater as micronutrients for photosynthesis and these filamentous macroalgae have a linear relationship with water (BCF>5000).

The Zn accumulation capacity of *V. fucoides* is related to thallus morpology (Stengel et al., 2004). Moreover, antropogenic pressure affect Zn density, and the maximum concentration was found in DSI station in summer associated with increased population.

|                          | Al                  | Mn                   | Fe                   | Со           | Ni            | Cu                   | Zn                   | As            | Cd                  | Hg           | Pb           |
|--------------------------|---------------------|----------------------|----------------------|--------------|---------------|----------------------|----------------------|---------------|---------------------|--------------|--------------|
| <i>Ceramium</i> spp.     | 3932.1              | 115.5                | 3271.8               | 2.03         | 9.3           | 13.9                 | 114.8                | 4.9           | 0.4                 | 0.1          | 3.9          |
| BSAF<br>BCF              | 0.9<br><b>10425</b> | 0.4<br><b>5451.7</b> | 0.5<br><b>7998.4</b> | 0.6<br>934.6 | 0.9<br>1485.8 | <b>3.1</b><br>3185.5 | <b>10.6</b><br>320.4 | 0.8<br>1683.2 | <b>8.1</b><br>891.7 | 1.3<br>596.1 | 1.1<br>978.6 |
| Corallina<br>officinalis | 404.3               | 72.9                 | 466.2                | 0.3          | 5.1           | 3.4                  | 34.8                 | 1.8           | 0.2                 | 0.006        | 1.4          |

| BSAF                   | 0.1    | 0.6            | 0.1    | 0.1    | 1.5    | 0.6        | <b>4.4</b>  | 0.4    | <b>6.7</b>  | 0.1        | 0.7    |
|------------------------|--------|----------------|--------|--------|--------|------------|-------------|--------|-------------|------------|--------|
| BCF                    | 2378.2 | 9112.5         | 2590   | 200    | 1593.8 | 2428.6     | 192.1       | 1200   | 500         | 150        | 1166.7 |
| Gelidium<br>crinale    | 1061.5 | 281.9          | 875.8  | 2.6    | 10.1   | 11         | 98          | 3.5    | 0.1         | 0.01       | 2.5    |
| BSAF                   | 0.3    | 0.5            | 0.1    | 0.5    | 1.1    | <b>3.7</b> | <b>11.3</b> | 0.8    | <b>2.8</b>  | 0.9        | 0.8    |
| BCF                    | 2075.8 | 8274.2         | 1612.3 | 1184.4 | 2277.3 | 2947.4     | 354.5       | 1728.8 | 229.2       | 240.1      | 598.4  |
| Laurencia<br>obtusa    | 346.7  | 11             | 402.4  | 0.1    | 2.9    | 5.8        | 41.1        | 1.6    | 0.2         | 0.01       | 0.9    |
| BSAF                   | 0.1    | 0.1            | 0.1    | 0      | 0.9    | 1.1        | <b>5.2</b>  | 0.4    | <b>6.7</b>  | 0.2        | 0.5    |
| BCF                    | 2039.4 | 1375           | 2235.6 | 66.7   | 906.3  | 4142.9     | 226.8       | 1066.7 | 500         | 275        | 750    |
| Vertebrata<br>fucoides | 2921.2 | 120.8          | 1904.1 | 0.8    | 5.3    | 6.8        | 136.4       | 2.6    | 0.6         | 0.02       | 3.3    |
| BSAF                   | 0.9    | 0.5            | 0.3    | 0.4    | 0.8    | <b>3.1</b> | <b>16.1</b> | 0.6    | <b>10.7</b> | <b>2.2</b> | 0.5    |
| BCF                    | 4435.9 | <b>10575.8</b> | 2564.9 | 512.5  | 1247   | 3251.7     | 334.1       | 1204.9 | 1300        | 377.5      | 693.5  |

\*BSAF > 2 (macro concentrator) and BCF > 5000 (very bio-accumulative) values are shown in bold.

*L. obtusa* was the only species studied in the Black Sea of Turkish coasts. Piner Olgunoğlu and Polat (2007) studied on *L. papillosa* in Iskenderun Bay and the results indicated that a similar patterns between the heavy metals were found except Pb level.

Coralline algae are tough, resistant to abrasion and important contributors to carbon budget. In this study, C. officinalis was sampled in relatively unpolluted Karakum coast in Sinop and Fe/Zn ratio showed inverse relationship to Mn depending upon low contamination. C. officinalis could be a biomonitor for Cd, that was confirmed by a higher BSAF value.

#### 4. CONCLUSION

Results obtained in this study indicated that continuous discharge wastewater and untreated industrial effluents have increased in Sinop coastal areas along with population growth. To measure levels of elements is necessary for determining potential health and environmental risks to ensure GES in accordance with MSFD. Nutritional values of macroalgae for use as vegetables or as food ingredients make them to investigate. Little quantitative data are available on the concentration of heavy metals in common red algae to estimate future trends. In this study, two species were determined for the first time. There is no proof for significant heavy metal pollution in Sinop Province. However, the coasts of this region have to be monitored regularly to avoid increased pollutants.

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#### REFERENCES

- Arici E (2017). Using dominant macroalgae and seagrass in Sinop coastline of the Black Sea as biomonitors for determination of heavy metal pollution. PhD Thesis, Sinop University, Turkey, 161 p.
- Arici E & Bat L (2016). Using marine macroalgae as biomonitors: Heavy metal pollution along the Turkish west coasts of the Black Sea. In: 41st CIESM Congress, Rapp. Comm. Int. Mer Medit., Kiel, 41: 238.
- Bat L & Arici E (2016). Heavy metal concentrations in macroalgae species from Sinop coasts of the southern Black Sea. Journal of Coastal Life Medicine, 4(11): 841-845.
- Bat L & Öztürk M (1997). Heavy metal levels in some organisms from the Sinop Peninsula of the Black Sea. Tr. J. of Engineering Sciences, 21: 29-33.
- Bat L, Gökkurt O, Sezgin M, Üstün F & Sahin F (2009). Evaluation of the Black Sea land based sources of pollution the coastal region of Turkey. The Open Marine Biology Journal, 3: 112-124.
- Bat L, Öztekin A, Şahin F, Arıcı E & Özsandıkçı U (2018). An overview of the Black Sea pollution in Turkey. MedFAR. 1(2): 67-86.
- Bat L, Öztekin H C, Arıcı E, & Vişne A (2015) A preliminary study on the heavy metal levels of dwarf eelgrass Zostera noltii Homermann in the Black Sea. Journal of Aquaculture and Marine Biology, 4 (1): 1-5.

- CEPA (Canadian Environmental Protection Act) (1999). 286 p. Retrieved in February, 2018 from http://laws-lois.justice.gc.ca/eng/acts/c-15.31/
- CSQG (2001). 35 p. Retrieved in March, 2019 from https://www.ccme.ca/files/Resources/supporting\_scientific\_documents/pn\_1176\_e.pdf
- EC (European Commission) (1998). 23 p. Council Directive 98/83/EC of 3 November 1998 on the quality of water intended for human consumption. Official Journal of the European.
- EPA (Environmental Protection Agency) (2008). EPA's 2008 Report on the Environment. National Center for Environmental Assessment, Washington, DC; EPA/600/R-07/045F. Retrieved from the National Technical Information Service, Springfield, VA, and online at http://www.epa.gov/roe.
- Geyer H J, Rimkus G G, Scheunert I, Kaune A, Kettrup A, Zeeman M, Muir D C G, Hansen L G, & Mackay D (2000). Bioaccumulation and occurrence of endocrine-disrupting chemicals (EDCs), persistent organic pollutants (POPs), and other organic compounds in fish and other organisms including humans, in bioaccumulation new aspects and developments. In: B Beek (ed.), the Handbook of Environmental Chemistry, Springer-Verlag, Berlin Heidelberg, New York, 167 p.
- Güven K C, Okuş E, Topçuoğlu S, Esen N, Kücükcezzar R, Seddigh E & Kut D (1998). Heavy metal accumulation in algae and sediments of the Black Sea coast of Turkey. Toxicol. Environ. Chem., 67: 435-440.
- Güven K C, Topçuoğlu S, Kut D, Esen N, Erentürk N, Saygı N, Cevher E, & Güvener B, (1992) Metal uptake by Black Sea algae. Botanica Marina, 35: 337-340.
- Kleinov K N, Nichols J W, Hayton W L, McKim J M & Barron M G (2008) Toxicokinetics in fish. In: R T Di Giulio, D E Hinton (Eds.), The Toxicology of Fishes, Taylor and Francis Group LLC, Boca Raton, US, pp. 55-152.
- Kut D, Topçuoğlu S, Esen N, Küçükcezzar R & Güven K C (2000). Trace metals in marine algae and sediment samples from the Bosphorus. Water, Air, and Soil Pollution, 118: 27-33.
- Lobban, C S & Harrison, P J (1994). Seaweed Ecology and Physiology. Cambridge University Press, Cambridge, 384 p.
- MSFD (2008). Official Journal of the European Union. Directives Directive 2008/56/EC of the European Parliament and of the Council of 17 June 2008 Establishing a Framework for Community Action in the Field of Marine Environmental Policy. Marine Strategy Framework Directive, L 164, 19-40.
- NOAA (National Oceanographic and Atmosphere Administration) (2009) Proceedings of the International Research Workshop on the Occurrence, Effects and Fate of Microplastic Marine Debris, Silver Spring. Retrieved in January, 2017 from http://www.noaa.gov/
- Öztekin H C (2015). Heavy metal levels in fish, invertebrates, zooplankton, sea grass and sediment from Sinop coasts of the Black Sea. Master Thesis, Sinop University, 75 p.
- Öztürk M, Bat L, & Öztürk M (1994a). A study on heavy metal levels in *Ulva lactuca* (L.) Le Jolis 1863, collected from Sinop coast of Black Sea, Turkey. E.U. Fen Fak. Der., 16(1): 187-195 (in Turkish).
- Öztürk M, Bat L & Öztürk M (1994b). Heavy metal levels in bioindicator organisms collected from Sinop bay and harbour. In: XII. National Biology Congress, Edirne. p. 20-25.
- Öztürk M, Bat L & Öztürk M (1996). The heavy metal levels in *Palaemon elegans* Rathke 1837, collected from Sinop coast of the Black Sea. Tarım ve Çevre İlişkileri Sempozyumu, 13-15 May, Mersin, pp. 366-373.
- Phillips D J H (1990). Use of macroalgae and invertebrates as monitors of metal levels in estuaries and coastal waters. In: R W Furness, P S Rainbow (Eds.), Heavy Metals in the Marine Environment, CRC Press, Boca Raton, pp. 81-99.
- Piner Olgunoğlu M P & Polat S (2007). Seasonal changes of heavy metals in two macroalgae species [*Cystoseira corniculata* (Phaeophyta), *Laurencia papillosa* (Rhodophyta)] in the Iskenderun Bay. E.U. Journal of Fisheries & Aquatic Sciences 24 (1-2): 25-30.
- REACH (Registration, Evaluation and Authorization of Chemicals) (2006). Regulation (EC) No 1907/2006 of the European Parliament and of the Council of 18 December 2006, Annex II. 2006; 396-849.
- Stengel D B, Macken A, Morrison L & Morley N (2004). Zinc concentrations in marine macroalgae and a lichen from western Ireland in relation to phylogenetic grouping, habitat and morphology. Mar. Pollut. Bull., 48 (9-10): 902-909.
- Topçuoğlu S, Esen N, Eğilli E, Güngör N & Kut D (1998b). Trace elements and 137 Cs in macroalgae and mussel from the Kilyos in Black Sea. IAEA-SM354/30P 283-284.
- Topçuoğlu S, Güven K C, Balkıs N, & Kırbaşoğlu C (2003). Heavy metal monitoring of marine algae from the Turkish coast of the Black Sea, 1998-2000. Chemosphere 52: 1683-1688.
- Topçuoğlu S, Güven K C, Okuş E, Esen N, Güngör N, Eğilli E, Kut D, Doğan E & Unlu S (1998a). Metal contents of algae and sediments of Turkish coasts in the Black Sea (1979-1989 and 1991-1993). The Proceedings of the First International Symposium on Fisheries and Ecology, 2-4 September, Trabzon, pp. 437-439.
- Topçuoğlu S, Kılıç O, Belivermiş M, Ergül H A & Kalaycı G (2010). Use of marine algae as biological indicator of heavy metal pollution in Turkish marine environment. J. Black Sea/Mediterranean Environment, 16(1): 43-52.
- Topçuoğlu S, Kırbaşoğlu C & Güngör N (2002). Heavy metals in organisms and sediments from Turkish Coast of the Black Sea, 1997-1998. Environment International, 27: 521-526.

- Topçuoğlu S, Kut D, Esen N, Güngör E, Seddigh E, & Küçükksezzar R (1998c). Trace elements and radionuclides in sediments and biota from the Küçükcekmece Lake. Rapp. Comm. int. Mer Médit., 35: 294-295.
- TSI (Turkish Standards Institute), 2005. TS-266 Standards for Drinking and Utilization Water.
- Türk Çulha S, Koçbaş F, Gündoğdu A & Çulha M (2013). Heavy metal levels in marine algae from the Black Sea, Marmara Sea and Mediterranean Sea. Rapp. Comm. int. Mer Medit., 40: 827-828.
- UNEP (2011). Climate change and POPS: predicting the impacts. Report of the UNEP/AMAP Expert Group Secretariat of the Stockholm Convention, Geneva, Switzerland.
- USEPA (1976). Quality criteria for water. Office of Planning and Water, US Environmental Protection Agency, Washington DC. EPA-440-9-76-023.
- Wahbeh M I & Mahasneh, D M (1985). Concentrations of zinc, manganese, copper, cadmium, magnesium and iron in ten species of algae and sea water from Aqaba, Jordan. Mar. Environ. Res., 16: 95-102.
- WHO (2011). DDT in indoor residual spraying: Human health aspects. World Health Organization, Geneva.



# The Effectiveness of Different Treatments in Preventing Melanosis in Shrimp

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Abstract: In this study, the effectiveness of different treatments to prevent melanosis in fresh deepwater pink shrimp (*Parapenaeus longirostris*) was evaluated. Fresh shrimps were collect from local market and transferred to the Çukurova University Fisheries Faculty Research Laboratory under cold chain. Fresh shrimps were divided into six groups after that immediately were dipped in tap water and acetic acid (control group 1 and 2), and solutions containing chitosan with high deacetylation degree and low deacetylaton degree, sodium metabisulphite, citric acid for ten minutes. After treatment the shrimp samples were packed and stored at  $+4^{\circ}$ C. Melanosis index, sensory characteristics and colour parameters were determined periodically during the cold storage. Sodium metabisulphite showed strong properties of delaying melanosis development during cold stogare. The results of this study indicated that treatment with chitosan solutions did effect a statistically significant difference (p<0.05) In addition, the results showed that chitosan from shrimp wastes could be easily used instead of sodium metabisulphite.

Keywords: Melanosis, shrimp, chitosan, deepwater pink shrimp, Parapenaeus longirostris

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# Sensory Changes of Rainbow Trout (*Oncorhynchus mykiss*) Fed with Different Plant Oils Supplemented Feeds during Storage in Refrigerator Conditions

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**Abstract:** In this study, it was aimed to determine the preservation time of the serving size rainbow trout (*Oncorhynchus mykiss*) fed with feeds supplemented with different herbal oils as additives. The fish were cleaned after harvesting and stored without any treatment. Oils obtained from black mustard (*Brascica nigra*), stinging nettle (*Urtica dioica*), milk thistle (*Silybum marianum*) and cannabis (*Cannabis sativa*) herbs were added to commercial trout feeds at the rates of 5, 10 and 20 ml kg<sup>-1</sup>. The trial was carried out with 3 replications of 50 individuals in each tank and fish were fed with these prepared feed for a total of 60 days. At the end of the experiment, the fish were killed by anesthetic and they were placed on a styrofoam plates in a refrigerator bag after cleaning internal organs and stored at +4 °C for 24 days. Changes in sensory qualities of all treatment groups were determined in comparison with control group according to raw fish sensory evaluation form. Ultimately, preservation durations of trouts fed with different plant oils supplemented feed were determined according to sensory analysis results. The results of this study indicated that general preference criteria such as appearance, odour, texture and colour were chosen for the sensorial analysis, which were rated by the panellists. According to the results of overall appearance control group was spoilaged at 18th day wherease all consantrations of stinging nettle plant oil added groups fishes were stored longer than other groups and control group.

Keywords: Black mustard, cannabis, stinging nettle, milk thistle, rainbow trout, sensory evaluation.



# ORAL PRESENTATION

# Assessing the Water Quality of River Kızılırmak (Sivas)

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**Abstract:** It was aimed to seasonally monitor the water quality of River Kızılırmak, which is used for irrigating the agricultural land around Sivas Province, for a year, to record the seasonal changes by using the water quality data, and to establish a database to be used in further studies to be carried out on River Kızılırmak, on which no study has been carried out before.

This study, which was started in August 2011, was carried out by taking water samples in all 4 seasons. The water samples to be used in analyzing the water quality parameters were collected on monthly basis. 1 day before the field works, the equipment, devices, and sampling bottles were disinfected. The water samples were collected by sinking the 3L bottles into 15cm depth from the surface and in the direction opposite to the water flow. The parameters examined are dissolved oxygen (DO), salinity, pH, temperature, and electrical conductivity (EC), which were analyzed on-site, and the suspended solid matters (SSM), chemical oxygen demand (COD), biological oxygen demand (BOD), nitrite, nitrate, phosphate, sulfide, sulfate, chloride, sodium, potassium, ferrous, lead, copper, nickel, zinc, cobalt, and cadmium, which were taken to Sivas Province Food Control Laboratory within 4 hours and analyzed in the same day.

In the present study, which was carried out on the Brook Kızılırmak that constitutes an important water source for Sivas city center and nearby villages, the chemical and physical analyses of water samples were performed and the seasonal changes were recorded. The water quality classes were determined according to the actual Surface Water Quality Management Regulation's Intra-Continental Water Classes, and the upper and lower limit values were compared. In conclusion, it was determined that the studies on improving the actual water quality of River Kızılırmak, in which many fishes and aquatic organisms are known to live, should be immediately started, and that the importance of this brook should be taken into consideration for the sustainability of ecological balance and the irrigation of nearby agricultural lands. Moreover, it is necessary to repeat the water quality studies on a regular basis for determining the contribution to water quality of brook, to prevent the release of insufficiently filtered water into brook, to prevent the leakage of domestic and animal wastes through precipitation water or underground water, and to take measures and precautions in order to improve the actual water quality of River Kızılırmak.

Keywords: Water Quality, Water Pollution, Sivas, River Kızılırmak



# Antibacterial Activity of Chitosan, Some Plant Seeds Extracts and Oils Against Pathogenic Organisms *Escherichia coli* and *Staphylococcus aureus*

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Abstract: In this study, the antibacterial activity of chitosan, three different plants seeds and oils were used. With this aim, three different aqueous methanolic extracts which are black seed (*Nigella sativa* L), Flax seeds (*Linum Usitatissimum*) and Chaste Tree seeds (*Vitex agnus-castus* L.), four different extracted chitosan which were shrimp (*Parapenaeus longirostris*), crayfish (*Astacus leptodactylus*), Blue Crab (*Callinectes sapidus*) and Pavurya Crab (*Eriphia verrucosa*) chitosan prepared and concentrations were adjusted 0.5%, 1% and 2%, respectively. And two of the same concentrations of cold press oils which were black seed (*Nigella sativa* L) and Flax seeds (*Linum Usitatissimum*) oils prepared same three different concentrations (0.5%, 1% and 2%,), as well as Eritromisin (15µg/ml) and florfenicol (30µg/ml), was used as a positive control to determine the sensitivity of microbial strains and cotton oil and acetic acid as negative controls. For evaluating its effect on the growth of one gram-negative bacteria (*Escherichia coli*) and one gram-positive bacteria (*Staphylococcus aureus*). by disc diffusion method.

Antimicrobial activity of chitosan is affected by different intrinsic and extrinsic factors. Source of chitosan, molecular weight, deacetylation degree, viscosity and solvent material could be stated as intrinsic factors, while the extrinsic factors are related with pH, temperature, ionic strength, metal ions, presence of EDTA organic matters and bacteria cultures. In the present study, all four extracted chitosans were observed to have different antimicrobial effects on two different type of bacteria, on the other hand there are not any significant antibacterial effect of methanolic extracts and oils of the three plants that used in this study.

Keywords: Chitosan, Nigella sativa, Linum Usitatissimum, Vitex agnus-castus



# Feeding Factors Reducing the Level of the Total Ammonia-Nitrogen Excretion in Freshwater Fish

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Abstract: In this study, feeding factors that reduce the total ammonia-nitrogen (TAN) level for cultured and ornamental freshwater fish species are reviewed. Lysine included plant-based ingredients (soybean and wheat meals) in rainbow trout (*Oncorhynchus mykiss*) feeds are decreased the TAN level. Mojave yucca (*Yucca schidigera*), bamboo (*Dracaena sanderiana*) charcoal, and soapbark (*Quillaja saponaria*) included diets are reduced the total ammonia-nitrogen (TAN) excretion in channel catfish (*Ictalurus punctatus*), hyprid tilapia (*Oreochromis mossambicus*  $\times$  *O. niloticus*), and striped catfish (*Pangasianodon hypophthalmus*). Spirulina (*Arthrospira platensis*) inclusion, feed type, and protein level are affected to TAN level in kenyi (*Maylandia lombardoi*), auratus (*Melanochromis auratus*), and yellow tail (*Pseudotropheus acei*) cichlids, respectively. In conclusion, ammonia-nitrogen level is important parameter in aquaculture industry and it should be emphasized with different freshwater fish species in future studies.

Keywords: Feeding regime, aquafeeds, feed additives, toxic effects, ammonium

## **1. INTRODUCTION**

Ammonia is one of the most harmful pollutants because of its toxic feature. Its unionized  $NH_3$  form is highly toxic to the fish species and aquatic environment. It can be lead to a decrease in both growth and reproduction performances and even it causes to death of aquatic organisms (Sergeant, 2014). The dangerous effects of ammonia are put it a remarkable point for aquaculture. Decreasing the level of ammonia in the water volume is important for aquaculture (Francis-Floyd et al., 2012). For this reason, some managements are applied for reduction the level of ammonia. Feeding regimes are one of the important factors amongst the such managements. Feeding managements, including optimum feeding rates, feeding frequencies, dietary additives or protein / lipid levels are reported as an ammonia reducer in previous studies.

# Water Management in Aquaculture

Because water is an essential requirement for fish farming, any properly prepared management plan for aquaculture must describe the quality and quantity of water available for the proposed facility. Water quality determines to a great extent the success or failure of a fish cultural operation (Alam and Al-Hafedh, 2006). Well-managed water parameters, including temperature, pH, dissolved oxygen, carbon dioxide, alkalinity, and ammonia improve the efficiency in aquaculture operations.

#### Nitrogenaus Compounds

Nitrogen can be an important factor controlling algal growth when other nutrients, such as phosphate, are abundant. Nitrogenous compounds in water volume are classified as;

| Dissolved gas =    | Nitrogen (N).                                       |
|--------------------|---|
| Dissolved solids = | Ammonium (NH4) and its unionized form, Ammonia (NH2 |
|                    | Nitric Oxide (NO),                                  |
|                    | Nitrite (NO <sub>2</sub> ),                         |
|                    | Nitrate (NO <sub>3</sub> ).                         |

Ammoniacal nitrogen (NH<sub>3</sub>-N), is a ration for the quantity of ammonia, a toxic compound often established in waste products, including sewage, liquid manure and other liquid organic wastages.

#### **Effects of Ammonia in Aquaculture Operations**

Ammonia can be converted to nitrite  $(NO_2)$  and nitrate  $(NO_3)$  by bacteria, and then used by plants. Nitrate and ammonia are the most common forms of nitrogen in aquatic systems (Quirós, 2003). Ammonia is one of the most harmful pollutants because of its toxic feature (Guthrie et al., 2018). The major source of ammonia in a water of a heavily stocked culture pond or in the effluent of a raceway is from excretion of fish, mostly via their gills. Total ammonia-nitrogen (TAN) concentration must be less than 0.5 mg/L in aquaculture operations. The level of 2 mg/L is caused lethal effect thorough the fish metabolism (Sergeant, 2014).

#### Feeding Factors as a Reducer of Total Ammonia-Nitrogen Level

Lysine included plant-based ingredients (soybean and wheat meals) in rainbow trout (*Oncorhynchus mykiss*) feeds are decreased the TAN level (Cheng et al., 2003). Mojave yucca (*Yucca schidigera*) inclusion to the feeds of channel catfish (*Ictalurus punctatus*) and hyprid tilapia (*Oreochromis mossambicus* × *O. niloticus*) is affected to decrease the ammonia excretion (Kelly and Kohler, 2003). Ammonia-nitrogen level is reduced by mojave yucca (*Y. schidigera*) addition to the striped catfish (*Pangasianodon hypophthalmus*) diets (Güroy et al., 2014). Ammonia-nitrogen level is reduced by *Nutrofito Plus* (yucca and soapbark extract) addition to the feeds of the same species (Güroy et al., 2016). Bamboo charcoal is positively affected on the reducing the level of TAN in the culture of this catfish (*Quaiyum et al., 2014*). *Spirulina (Arthrospira platensis)* inclusion to the diets of kenyi cichlids (*Maylandia lombardoi*) and different feeding frequencies are reduced the TAN level (Karadal et al., 2017). An ornamental fish feed is caused to decrease the TAN as compared with the salmon feed in auratus cichlids (*Melanochromis auratus*) (Karadal et al., 2018). Increasing of dietary protein level of yellow tail cichlid (*Pseudotropheus acei*) diet is led to higher concentrations of the TAN (Güroy et al., 2012).

### 2. CONCLUSION

The lethal effects of the ammonia in the aquaculture operations in both freshwater farms and ornamental fish facilities are tended to investigate the reducers of the TAN in the water volume. In conclusion, the plant proteins and plant-based feed additives are mainly used as a reducer of total ammonia-nitrogen excretion in the aquaculture. However, optimizing the feeding frequency and portion should be needed as a regulator of the water parameters.

#### REFERENCES

- Alam, A., and Al-Hafedh, Y. S., 2006. Diurnal dynamics of water quality parameters in an aquaculture system based on recirculating green water technology. Journal of Applied Sciences and Environmental Management 10(2): 19-21.
- Cheng, Z. J., Hardy, R. W., and Usry, J. L., 2003. Plant protein ingredients with lysine supplementation reduce dietary protein level in rainbow trout (*Oncorhynchus mykiss*) diets, and reduce ammonia nitrogen and soluble phosphorus excretion. Aquaculture 218(1-4): 553-565.
- Francis-Floyd, R., Watson, C., Petty, D., and Pouder, D.B., 2012. Ammonia in aquatic systems. University of Florida IFAS Extension Publication, No: FA16, 4 p.
- Guthrie, S., Giles, S., Dunkerley, F., Tabaqchali, H., Harshfield, A., Ioppolo, B., and Manville, C., 2018. The Impact of Ammonia Emissions from Agriculture on Biodiversity: An Evidence Synthesis. Rand Europe Publishing, 76 p.
- Güroy, B., Mantoğlu, S., Kayalı, S., and Şahin, İ., 2014. Effect of dietary Yucca schidigera extract on growth, total ammonia–nitrogen excretion and haematological parameters of juvenile striped catfish Pangasianodon hypophthalmus. Aquaculture Research 45(4): 647-654.
- Güroy, B., Mantoğlu, S., Merrifield, D. L., and Guroy, D., 2016. Effects of dietary *Nutrafito Plus* on growth, haemotological parameters and total ammonia-nitrogen excretion of juvenile striped catfish *Pangasianodon hypophthalmus*. Aquaculture Research 47(6): 1770-1777.
- Güroy, D., Sahin, I., Güroy, B., Altin, A., and Merrifield, D. L., 2012. Effect of dietary protein level on growth performance and nitrogen excretion of the yellow tail cichlid, *Pseudotropheus acei*. The Israeli Journal of Aquaculture Bamidgeh 64: 6 p.
- Karadal, O., Güroy, D., and Türkmen, G., 2018. Effects of feed type and feeding frequency on growth performance, reproductive efficiency and skin coloration of auratus cichlids (*Melanochromis auratus*). Aquaculture Studies 18(2): 135-144.

- Karadal, O., Güroy, D., and Türkmen, G., 2017. Effects of feeding frequency and *Spirulina* on growth performance, skin coloration and seed production on kenyi cichlids (*Maylandia lombardoi*). Aquaculture International 25(1): 121-134.
- Kelly, A. M., and Kohler, C. C., 2003. Effects of *Yucca shidigera* extract on growth, nitrogen retention, ammonia excretion, and toxicity in channel catfish *Ictalurus punctatus* and hybrid tilapia *Oreochromis mossambicus* × *O. niloticus*. Journal of the World Aquaculture Society 34(2): 156-161.
- Quaiyum, M. A., Jahan, R., Jahan, N., Akhter, T., and Sadiqul, I. M., 2014. Effects of bamboo charcoal added feed on reduction of ammonia and growth of *Pangasius hypophthalmus*. Journal of Aquaculture Research & Development 5(6): 1000269.

Quirós, R., 2003. The relationship between nitrate and ammonia concentrations in the pelagic zone of lakes. Limnetica 22(1-2): 37-50.

Sergeant, C., 2014. The management of ammonia levels in an aquaculture environment. Water/Wastewater 2 p.



# Dietary Amino Acids Affecting the Immune Status and Blood Parameters of Marine Fish

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Abstract: Amino acids are essential constituents for the aquafeeds which are developed to meet requirements of the fish species. In this study, dietary amino acids affecting the health status of various cultured marine fish species are reviewed. Methionine and tryptophan are improved the immune status of European sea bass (*Dicentrarchus labrax*). Lysine, methionine, tryptophan, and aspartate supplementation to the diets of meagre (*Argyrosomus regius*) is enhanced the haematocrit level and bactericidal activity against *Vibrio anguillarum* and *Photobacterium damselae*. Dietary glutamine is improved the glutathione content and superoxide dismutase activity in the intestine of gilthead sea bream (*Sparus aurata*). Arginine addition to the feeds of turbot (*Scophthalmus maximus*) is decreased serum cortisol. In conclusion, supplementation of the essential or non-essential amino acids to the marine fish diets is important for the health status of cultured species.

Keywords: Fish nutrition, dietary protein, feed additives, health parameters, immunostimulants

# **1. INTRODUCTION**

There are vast differences in the essential amino acid requirements of marine fish species when stated as a proportion of the aquafeeds (Güroy et al., 2017). Dietary amino acids are needed to recover of body amino acids lost with some metabolic managements, including intestine activities, amino acids oxidation, nitrogen molecules conversion, and protein turnover (Li et al., 2009). Developing suggestions from studies with aquatic organisms presents that many amino acids achieve key metabolic lines that are critical to growth, reproduction, health, and immune status. In this study, dietary amino acids affecting the blood parameters and immune status of several economic marine fish are reviewed.

#### **Aquafeeds and Feed Additives**

Aquafeeds are made by grinding and mixing together ingredients such as fishmeal, vegetable proteins and binding agents such as wheat products. Nutritionists who formulate aquafeeds have to account for about 40 essential nutrients needed by the fish (NOAA, 2018). The essential nutrients contain vitamins, minerals, amino acids (the building blocks of protein), and some fats. These important ingredients are supplemented to the aquafeeds as feed additives. These additives added to aquafeeds are aimed to increase the voluntary intake, palatability, and digestibility of the fish diets. Also, the feed additives play roles for fish gaining resistance against diseases.

#### **Amino Acids**

Amino acids are important molecules for the protein structures, such as enzymes, hormones, peptides, and cofactors, which are significant in the metabolism of the fish species (Güroy et al., 2017). There are 22 important amino acids for the vital requirements of the fish species and these amino acids are divided into three sub-groups, including essential, non-essential, and conditionally essential (Table 1). Deficiency of the most essential amino acids in diet results in a reduced weight gain, poor feed efficiency, and diminished disease resistance in fish (Kiron, 2012; Yaghoubi et al., 2018).

Table 1. Important amino acids for the aquatic organisms (Li et al., 2009)

| Non-essential Amino Acids | Conditionally Essential Amino Acids   |
|---------------------------|---|
| Alanine                   | Cysteine  |
| Asparagine                | Glutamine   |
| Aspartate                 | Hydroxyproline  |
| Glutamate                 | Proline   |
| Glycine                   | Taurine   |
| Serine                    |   |
| Tyrosine                  |   |
|                           |   |
|                           |   |
|                           |   |
|                           | Non-essential Amino Acids<br>Alanine<br>Asparagine<br>Aspartate<br>Glutamate<br>Glycine<br>Serine<br>Tyrosine |

# Fish Health

Aquafeeds contain all the essential nutrients needed to keep them healthy and growing. A deficiency of any nutrient can affect fish health directly by impairing metabolic functions, or indirectly by making fish more susceptible to opportunistic disease-causing agents.

Presence or absence of amino acids in the diets of fish can directly / indirectly affect to the blood parameters of fish. Fish have a well-developed immune system, composed by innate mechanisms which are responsible for resistance to diseases. In marine fish aquaculture operations, water moves freely between farms and the ocean. Risks lead to the transmission of disease from wild to farmed fish.

#### Effects of Amino Acids on Blood and Immune Parameters of Marine Fish

Methionine is improved the immune status of European sea bass (*Dicentrarchus labrax*) and this situation is led to enhance immune response and plasma parameters of this species (Machado et al., 2018). Arginine (1 and 2% supplementation) is inhibated the immune defences and disease resistance of this species (Azeredo et al., 2015). Tryptophan is affected to immune system as an immunostimulant in 275 g weight of the same species (Machado et al., 2015). Lysine and methionine supplementation to the diets of meagre (*Argyrosomus regius*) is positively impacted to the haematocrit and haemoglobin levels in 44.4 g of this species (Güroy et al., 2017). Existing of tryptophan and aspartate in meagre diets is enhanced the bactericidal activity against *Vibrio anguillarum* and *Photobacterium damselae* (Gonzalez-Silvera et al., 2018). Dietary glutamine is improved the glutathione content and superoxide dismutase activity in the intestine of 13 g gilthead sea bream (*Sparus aurata*) (Coutinho et al., 2016). Arginine addition to the 8.5 g olive flounder (*Paralichthyes olivaceus*) diets is improved the innate immunity of the fish (Rahimnejad and Lee, 2014).

#### 2. CONCLUSION

In conclusion, nutrition-based health approach is a new trend in modern aquaculture. Addition of the amino acids to the marine fish aquafeeds is enhanced the health status, such as blood, serum, and immune parameters. This study provides a basic information about the nutritional effects of amino acids in aquafeeds on the health parameters of cultured marine fish species. Further studies should be subjected on the effects of different amino acids on haematological and plasma parameters, immune responses, immune related gene expression of economic marine and freshwater fish species.

#### REFERENCES

- Azeredo, R., Pérez-Sánchez, J., Sitjà-Bobadilla, A., Fouz, B., Tort, L., Aragão, C., Oliva-Teles, A., and Costas, B., 2015. European sea bass (*Dicentrarchus labrax*) immune status and disease resistance are impaired by arginine dietary supplementation. PLoS One 10(10): e0139967.
- Costas, B., Rego, P. C. N. P., Conceicao, L. E. C., Dias, J., and Afonso, A., 2013. Dietary arginine supplementation decreases plasma cortisol levels and modulates immune mechanisms in chronically stressed turbot (*Scophthalmus maximus*). Aquaculture Nutrition 19: 25-38.
- Coutinho, F., Castro, C., Rufino-Palomares, E., Ordóñez-Grande, B., Gallardo, M. A., Oliva-Teles, A., and Peres, H., 2016. Dietary glutamine supplementation effects on amino acid metabolism, intestinal nutrient absorption capacity and antioxidant response

of gilthead sea bream (*Sparus aurata*) juveniles. Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology 191: 9-17.

- Gonzalez-Silvera, D., Herrera, M., Giráldez, I., and Esteban, M., 2018. Effects of the dietary tryptophan and aspartate on the immune response of meagre (*Argyrosomus regius*) after stress. Fishes 3(1): 6.
- Güroy, D., Karadal, O., Güroy, B., Mantoğlu, S., Çelebi, K., Şimşek, O., Eroldoğan, O.T., Genç, M.A., and Genç, E., 2017. The effects of dietary protein levels with amino acid supplementation on the growth performance, haematological profile and histology of meagre (*Argyrosomus regius*) in two different size classes. Aquaculture Research 48(12): 5751-5764.
- Kiron, V., 2012. Fish immune system and its nutritional modulation for preventive health care. Animal Feed Science and Technology 173(1- 2): 111-133.
- Li, P., Mai, K., Trushenski, J., and Wu, G., 2009. New developments in fish amino acid nutrition: towards functional and environmentally oriented aquafeeds. Amino Acids 37(1): 43-53.
- Machado, M., Azeredo, R., Díaz-Rosales, P., Afonso, A., Peres, H., Oliva-Teles, A., and Costas, B., 2015. Dietary tryptophan and methionine as modulators of European seabass (*Dicentrarchus labrax*) immune status and inflammatory response. Fish & Shellfish Immunology 42(2): 353-362.
- Machado, M., Azeredo, R., Fontinha, F., Fernández-Boo, S., Conceição, L. E., Dias, J., and Costas, B., 2018. Dietary methionine improves the European seabass (*Dicentrarchus labrax*) immune status, inflammatory response, and disease resistance. Frontiers in Immunology 9: Article 2672.
- NOAA, 2018. Feeds for Aquaculture. National Oceanic and Atmospheric Administration, Published in April 06, 2018 and Retrieved in March 25, 2019 from https://www.fisheries.noaa.gov/insight/feeds-aquaculture/
- Rahimnejad, S., and Lee, K. J., 2014. Dietary isoleucine influences non-specific immune response in juvenile olive flounder (*Paralichthys olivaceus*). Turkish Journal of Fisheries and Aquatic Sciences 14(4): 853-862.
- Yaghoubi, M., Mozanzadeh, M. T., Safari, O., and Marammazi, J. G., 2018. Gastrointestinal and hepatic enzyme activities in juvenile silvery-black porgy (*Sparidentex hasta*) fed essential amino acid-deficient diets. Fish Physiology and Biochemistry 44(3): 853-868.



# KASTAMONU BELEDIYESI

# **Crayfish Diseases**

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**Abstract:** Accurate and rapid diagnosis, prevention and treatment of diseases are the issues that need to be emphasized for cultivation, conservation and sustainability of aquatic animals containing high amount of protein such as crayfish. In this review, it is aimed to give information about crayfish diseases encountered across the world and their etiology, epizootiology, pathology, diagnosis and treatment methods. According to the literature data, it was evaluated that the most jeopardous disease is crayfish plague, whereas the second one is white spot syndrome virus. In conclusion, while studies on the detection and distribution of diseases are continuing, treatment studies of crayfish diseases, especially crayfish plague, need to be intensified.

Keywords: Crayfish, crawfish, disease, plague.

## **1. INTRODUCTION**

There are more than 400 crayfish species distributed worldwide. However, only 10 different species of them are of economic importance (Huner 1994, 1995). Four of these species belong to Cambaridae (*Procambarus clarkii, P. acutusacutus, P. zonangulus* and *Orconectes immunis*) family; three belong to the family of Astacidae (*Astacus astacus, A. leptodactylus* and *A. pacifastacus*) and the other three belong to the family of Parastacidae (*Cherax quadricarinatus, C. tenuimanus* and *C. destructor*) (Huner 1995; Mazlum and Yılmaz 2006).

The most common crayfish species in Turkey is Turkish crayfish (*Astacus leptodactylus*) which is also known as narrowclawed crayfish (Akhan et al., 2014; Kalaycı and Akhan, 2016). In turkey, crayfish are consumed as food and mainly exported to abroad. Its capture amount was recorded as 524 metric tons in 2018 according to Turkish Statistical Institute (TÜİK) whereas, production by culture is not available. Even though there is no total data for world crayfish production, in 2016 China produced 899100 tons crayfish alone where, 95% of which was by aquaculture (FAO, 2017).

First disease diagnosis from crayfish was performed in 1850s where *Psorospermium haeckeli* was reported. Although, crayfish plague which is the most important disease in crayfish was emerged in the 1860s, researchers were able to identify causative agent of it in 1934 as *Aphanomyces astaci* (Cornalia 1860; Alderman 1996). In the following years, the diagnosis of other diseases in crayfish continued. Accurate and rapid diagnosis, prevention and treatment of diseases is important for the production, protection and sustainability of crayfish populations. In order to achieve this, it is also necessary to know causative agents and their effects well. Within this scope, in this literature review, it is aimed to inform the reader about bacterial, viral, fungal and parasitic diseases in economically important crayfish species.

# **Crayfish Plague**

Crayfish plague is the most dangerous infectious disease affecting crayfish, resulting up to 100% mortality in an infected individual. Although *Aphanomyces astaci*, the causative agent of the disease, is pathogenic to European, Asian and Australian species; it has little or no effect on American crayfish species (Unestam 1969, 1972, 1975, Unestam and Weiss 1970).

It is estimated that the disease was first transported to Europe from North America around 1860 (Cornalia 1860, Alderman 1996). *Aphanomyces astaci* belongs to the Saprolegniales order of the Oomycete class. Oomycete class (commonly

referred as "water molds") is more closely related to brown algae and diatoms than it is to Eumycota which are real fungi. For this reason, *Aphanomyces astaci* is not accepted as real fungi taxonomically (Stephens 2005; Bower 2012).

The symptoms seen during the gross examination may vary, whereas in some crayfish the symptoms may not be visible. The most obvious symptom in European crayfish species is white abdomen. In some cases, browning (melanization) of cuticle and muscle tissue and *A. astaci* hyphae may appear in this area. It is molecularly diagnosed by PCR and there is no known treatment (Buller 2008).

Susceptible species are: Astacus astacus, Astacus leptodactylus, Astacopsis gouldi, Austropotamobius pallipes, Austropotamobius torrentium, Cambaroides japonicus, Cherax quadricarinatus, Cherax destructor, Cherax papuanus. Also; Orconectes limosus, Pacifasticus leniusculus and Procambarus clarkii species are native to North America and are carriers of Aphanomyces astaci and they do not exhibit any signs of disease other than intensive culture (Buller 2008).

# White Spot Syndrome (WSS)

White spot syndrome is one of the deadliest diseases seen in crayfish. Mortality is usually very high and it has been stated that the cumulative mortality reaches up to 100% between 3 and 10 days (Chou et al. 1995, Wongteerasupaya et al. 1995). White spot syndrome was reported from many Asian countries in the early 1990s (Inouye et al. 1994, Momoyama et al. 1994, Nakano et al. 1994, Chou et al. 1995, Flegel et al. 1995, Huang et al. 1995, Wongteerasupaya et al. 1995, Mohan et al. 1998, Park et al. 1998, Wang et al. 1999), and from South America and southeastern coast of USA more recently (Lightner et al. 1997, Lo et al. 1999, Wang et al. 1999, 2000, Jiravanichpaisal et al. 2001).

White spot syndrome virus (WSSV) is a rod-shaped crustacean virus with a wide geographic distribution (Wang et al. 1999). This virus is a double-stranded DNA virus which is in the first group according to the Baltimore classification system whereas, according to International Committee on Taxonomy of Viruses (ICTV) classification system, it belongs to Nimaviridae family. It is mostly transmitted horizontally through swallowing of water and vertically from the infected mother in hatcheries (Shi et al. 2000).

In the host, this virus infects many cells of ectodermal and mesodermal origin. Histological changes are seen in gill epithelium, gastric sac, hematopoietic tissue, nerve tissue, connective tissue and intestinal epithelial tissue (Wongteerasupaya et al. 1995). White spots appear on the shell of the infected crayfish as ellipses with a diameter of 0.3-3 mm. The clinical diagnosis is made by the appearance of white spots formed as described above where, molecular diagnosis is made quickly and specifically by using nested or quantitative PCR. There is no known treatment for this disease yet (Longshaw 2011).

Susceptible species are reported as; Astacus astacus, Cherax destructor albidus, Cherax quadricarinatus, Pasifastacus leniusculus and Procambarus clarkii (Longshaw 2011).

#### Intranuclear Basiliform Viruses (IBV)

These viruses are in the double-stranded DNA group. Due to the lack of immunological, biochemical and molecular information, the taxonomic position is unknown (Stentiford et al. 2004). IBVs are limited to the liver-pancreas and stomach. The strains seen in crayfish are: *Astacus astacus* bacilliform virus (AaBV) (Edgerton et al. 1996b), *Austropotamobius pallipes* bacilliform virus (ApBV) (Edgerton et al. 2002), *Cherax destructor* bacilliform virus (CdBV) (Edgerton 1996), *Cherax quadricarinatus* bacilliform virus (CqBV) (Anderson and Prior 1992) and *Pasifactacus leniusculus* bacilliform virus (PlBV) (Hauck et al. 2001) groups.

**Birnaviridae:** double stranded RNA viruses. It is an important fish pathogen, and crayfish are only responsible for carrying this virus (Halder and Ahne 1988). *Nimaviridae*: Known as white spot syndrome virus. *Parvoviridae*: single-stranded DNA virus. Some parvoviruses are seen in Australian Cherax species. These are: *Cherax quadricarinatus* parvo-like virus (CqPIV), *Cherax quadricarinatus* parvovirus of Bowater et al. (2002) and *Cherax destructor* systemic parvo-like viruses (CdSPV). *Picornavirida*: single-stranded RNA viruses. Edgerton (1999); Jones and Lawrence (2001) reported this virus-related mortality in Western Australia. However, its prevalence is less than 5%. *Reoviridae*: is a double-stranded RNA virus. It is a low-risk infection. It was considered to be relatively benign (Edgerton et al. 2000, Bowater et al. 2002, La Fauce and Owens 2007). *Totiviridae*: double-stranded RNA virus. It was reported from *Cherax quadricarinatus*. According to Owens and McElnea (2000) it causes death, albeit limited.

## **Unidentified Viral Infections**

A lot of viruses have been reported from crayfish to date. However, some have been described very poorly or not classified. There is therefore a need for proper identification and proper diagnosis of viruses in crayfish.

## Bacteria

Bacterial crayfish diseases were evaluated by Longshaw (20011) and major bacterial crayfish diseases reported as follows;

*Coxiella cheraxi*: It is a systemic intracellular bacterial infection encountered in *Cherax quadricarinatus*. Tan and Owens (2000) identified it by molecular techniques. The infection caused mortality in the host, which was thought to be systemic by the authors. However, in later studies it was reported to infect the gill tissue first, and in severe infections, the liver and the pancreas at an advanced stage (La Fauce and Owens, 2007).

*Nocardia*: Members of this genus (at least 80 species) have a wide range of hosts (humans, fish, mussels). To this date, it was merely reported from one crayfish individual (Alderman et al. 1986).

*Spiroplasma*: This bacterium, especially in insects, is often known as a male-killing organism and affects the sex ratio of some populations (Enigl and Schausberger, 2007; Kageyama et al., 2007). To date, there is no evidence that Spiroplasma affects the sex ratio in crayfish populations.

*Vibrio*: Several cases have been reported associated with Vibrio species. Although *V. cholerae* is held responsible for the deaths of *P. clarkii*, albeit to a small extent, it is usually *V. mimicus* (Thune et al., 1991) that causes terminal disease in crayfish. Infections are systemic and frequently present in the hemolymph and show no symptoms other than indolence (Eaves and Ketterer, 1994; Thune et al., 1991).

Aeromonas: Aeromonas hydrophila is generally known to cause disease in fish, mussels and humans and is a species of bacteria commonly found in fresh water (Tulsidas et al., 2008). This bacterium was isolated from crayfish which were apparently healthy but it was predicted that it would cause disease under culture conditions (Quaglio et al., 2006a).

## Fungi

Oomycetes, *Aphanomyces spp., Saprolegnia spp.* and Saprolegnia species are generally benign and infect dead eggs and dying crayfish larvae (Herbert, 1987; Royo et al., 2002; Vey, 1979, 1981). They can also infect healthy crayfish. Infestation starting from the wounded cuticle significantly increases fungal spread and mortality (Diéguez-Uribeondo et al., 1994).

Sedariomycetes: Fusarium (Plectosporium) has been reported to cause disease in several crayfish of this class, but has not been identified clearly on the basis of species (Quaglio et al., 2006a; 2006b). However, *F. avenaceum* reported as causative agent of burn spot disease by Arlı et al. (2017) and *F. oxysporum* was reported as causative agent of black gill disease reported by Taştan et al. (2017) in narrow clawed crayfish (*Astacus leptodactylus*).

*Microsporidia:* Microsporidia, traditionally classified as protista, are under fungi taxonomically (Fischer and Palmer, 2005; Gill and Fast, 2006; Hirt et al., 1999). The most important genus of this group in terms of crayfish diseases is Thelohania. Thelohania species are usually found in muscle tissue in the crayfish they infect, and produce an opaque or whitish color there. This disease is called 'Porcelain Disease'. There are doubts about the current phylogenetic position of the genus Thelohania. Thelohania species present in crayfish have a close affinity with Vairimorpha and Nosema seen in other insect and crustacean hosts (Brown and Adamson, 2006). Susceptible species are known as: *Astacus fluviatilis, Austropotamobius pallipes, Astacus astacus, Astacus leptodactylus, Pacifastacus leniusculus, Orconectes limosus* (Dunn et al., 2009; Edgerton et al., 2002a).

*Mesomycetozoea:* There are two orders defined in Mesomycetozoea class; Ichthyophonida and Dermocystida (Ragan et al., 1996). *Psorospermium spp.* which belong to Ichtyhophonida order, are parasite species that affect mot crayfish in Astacidae, Cambaridae and Parastacidae families (Vogt and Rug, 1999). According to Bangyeekhun et al. (2001), several types of *Psorospermium morphotypes* have been defined according to size, morphology, host and geographical locations. The most important of these is the *Psorospermium haeckeli* species found in Europe. This parasite emerges especially during moulting of crayfish and causes death. It is commonly found in gills, connective and nerve tissues, hepatopancrease, and entrance of stomach (Herbert, 1987; Edgerton and Owens, 1999; Jones and Lawrence, 2001).

Susceptible species are known as: Astacus astacus, Astacus leptodactylusi, Austropotamobius torrentium, Procambarus clarkii, Procambarus zonangulus, Pacifastacus leniusculus and Orconectes limnosus (Vogt and Rug, 1995).

#### Protista

*Ciliata*: Even though ciliata is associated with crayfish, it is not considered as a problem in the wild. Mortality usually occurs in culture conditions where there is poor water quality, unstable temperatures and high stocking density (Morado, 1995). They usually reproduce by fussion or budding. Most ciliata are found on the outer surfaces of crayfish such as pleopod, periopod, telson, fin and carapax (Sprague and Couch, 1971).

**Branchiobdella:** Branchiobdella, an annelid worm, is seen as ectocommensal or ectosymbiotic on crayfish. Branchiobdelides are found on the outer parts of the host crayfish such as gills and carapace; depending on the season and the presence of other Branchiobdelid species (Edgerton et al., 2002a). The inter-individual transmission takes place through contact of the host with another crayfish. There are several arguments regarding that branchiobdelides are pathogenic (based on the fact that some species cause reactions such as melanization in the gills of host individual) (Alderman and Polglase, 1988). Although only one mortality associated with Branchiobdelid has been reported, many bacteria and other pathogens have also been found in the dying individual and thought to be more likely to cause death (Hubault, 1935). Keller (1992); Brown et al. (2002) and Lee et al. (2009) on the other hand, concluded that Branchiobdelids improved growth rate of the host individual by repelling fouling organisms.

#### 4. CONCLUSION

In order to understand a disease, its etiology, epizootiology, pathology and diagnosis should be well-known. Treatment studies can be conducted only after knowing these factors. According to the data examined in this study, we suggest that studies on treatment or prevention of the crayfish plague and white spot syndrome diseases should be carried out. On the other hand, other diseases should be investigated in detail, and unspecified factors need identification.

#### REFERENCES

- Akhan, S., Bektas, Y., Berber, S., Kalaycı, G. (2014). Population structure and genetic analysis of narrowclawed crayfish (*Astacus leptodactylus*) populations in Turkey. *Genetica*, 142: 381-395.
- Alderman, D.J. (1996). Geographical spread of bacterial and fungal diseases of crustaceans. Revue Scientifique et Technique Office International des Épizooties, 15(2): 603-632.
- Alderman, D.J., Feist, S.W., Polglase, J.L. (1986). Possible nocardiosis of crayfish, Austropotamobius pallipes. J. Fish Dis., 9, 345– 347.
- Alderman, D.J., Polglase, J.L. (1988). Pathogens, parasites and commensals. In: Holdich, D.M., Lowery, R.S. (Eds.), Freshwater Crayfish: Biology. Management and Exploitation. Croom Helm, London, pp. 167–212.
- Anderson, I.G., Prior, H.C. (1992). Baculovirus infections in the mud crab, *Scylla serrata*, and a freshwater crayfish, *Cherax quadricarinatus*, from Australia. *J. Invertebr. Pathol.*, 60, 265–273.
- Arli, B., Akhan, S. (2017) Molecular identification of burn spot diseases (*Fusarium sp.*) from Turkish crayfish stocks by PCR method. I<sup>st</sup> International Symposium on Limnology and Freshwater Fisheries, Isparta, Turkey, 4-6 Ekim 2017, pp.200.
- Bangyeekhun, E., Ryynanen, H.J., Henttonen, P., Huner, J.V., Cerenius, L., Söderhäll, K. (2001). Sequence analysis of the ribosomal internal transcribed spacer DNA of the crayfish parasite *Psorospermium haeckeli*. *Dis. Aquat. Org.*, 46, 217–222.
- Bowater, R.O., Wingfield, M., Fisk, M., Kelly, M.L.C., Reid, A., Prior, H., Kulpa, E.C. (2002). A parvo-like virus in cultured redclaw crayfish *Cherax quadricarinatus* from Queensland, Australia. *Dis. Aquat. Org.*, 50, 79–86.
- Bower, S.M. (2012): Synopsis of Infectious Diseases and Parasites of Commercially Exploited Shellfish: Crayfish Plague ("Fungus" Disease).
- Brown, A.M.V., Adamson, M.L. (2006). Phylogenetic distance of *Thelohania butleri* Johnston, Vernick, and Sprague, 1978 (Microsporidia; Thelohaniidae), a parasite of the smooth pink shrimp *Pandalus jordani*, from its congeners suggests need for major revision of the genus Thelohania Henneguy, 1892. J. Eukaryot. Microbiol., 53, 445–455.
- Brown, B.L., Creed, R.P., Dobson, W.E. (2002). Branchiobdellid annelids and their crayfish hosts: are they engaged in a cleaning symbiosis? *Oecologia*, 132, 250–255.
- Chou, H.Y., Huang, C.Y., Wang, C.H., Chiang, H.C., Lo, C.F. (1995). Pathogenicity of a baculovirus infection causing white spot syndrome in cultured penaeid shrimp in *Taiwan. Dis. Aquat. Org.*, 23: 165–173.
- Cornalia, E. (1860). [English title not available]. (Sulla malattia dei gamberi.) *Atti della Società Italiana di Scienze Naturali*, II: 334-336.

- Diéguez-Uribeondo, J., Cerenius, L., Söderhäll, K. (1994). Saprolegnia parasitica and its virulence on three different species of crayfish. Aquaculture, 120, 219–228.
- Dunn, J.C., McClymont, H.E., Christmas, M., Dunn, A.M. (2009). Competition and parasitism in the native white clawed crayfish Austropotamobius pallipes and the invasive signal crayfish Pacifastacus leniusculus in the UK. Biol. Invasions. 11, 315–324.
- Eaves, L.E., Ketterer, P.J. (1994). Mortalities in red claw crayfish *Cherax quadricarinatus* associated with systemic *Vibrio mimicus* infection. *Dis. Aquat. Org.*, 19, 233–237.
- Edgerton, B. (1996). A new bacilliform virus in Australian *Cherax destructor* (Decapoda: Parastacidae) with notes on *Cherax quadricarinatus* bacilliform virus (= *Cherax baculovirus*). *Dis. Aquat. Org.*, 27, 43–52.
- Edgerton, B.F., Paasonen, P., Henttonen, P., Owens, L. (1996b). Description of a bacilliform virus from the freshwater crayfish, *Astacus astacus. J. Invertebr. Pathol.*, 68, 187–190.
- Edgerton, B.F., Owens, L. (1999). Histopathological surveys of the redclaw freshwater crayfish, *Cherax quadricarinatus*, in Australia. *Aquaculture*, 180, 23–40.
- Edgerton, B.F. (2000). A compendium of idiopathic lesions observed in redclaw freshwater crayfish, *Cherax quadricarinatus* (von Martens). *J. Fish Dis.*, 23, 103–113.
- Edgerton, B.F. (2002). Hazard analysis of exotic pathogens of potential threat to European freshwater crayfish. *Bull. Fr. Pêche Piscic.*, 367, 813–820.
- Edgerton, B.F., Evans, L.H., Stephens, F.J., Overstreet, R.M. (2002a). Synopsis of freshwater crayfish diseases and commensal organisms. *Aquaculture*, 206, 57–135.
- Enigl, M., Schausberger, P. (2007). Incidence of the endosymbionts Wolbachia, Cardinium and Spiroplasma in phytoseiid mites and associated prey. *Exp. Appl. Acarol.*, 42, 75–85.
- FAO 2017. FAO Yearbook of Forest Products.
- Fischer, W.M., Palmer, J.D. (2005). Evidence from small-subunit ribosomal RNA sequences for a fungal origin of Microsporidia. *Mol. Phylogenet. Evol.*, 36, 606–622.
- Flegel, T.W., Fegan, D.F., Sriurairatana, S. (1995) Environmental control of infectious disease in Thailand. In: Browdy CL, Hopkins JS (eds) Swimming through troubled water. Proceeding of special session on shrimp farming, Aquaculture '95. World Aquaculture Society, Baton Rouge, LA, p 65–79.
- Gill, E.E., Fast, N.M. (2006). Assessing the microsporidia–fungi relationship: combined phylogenetic analysis of eight genes. *GENE*. 375, 103–109.
- Haeckel, E. (1857) Ueber die Gewebe des Flusskrebses Arch Anat Physiol Med (Miiller's Archiv) 24:469-568
- Halder, M., Ahne, W. (1988). Freshwater crayfish Astacus astacus a vector for infectious pancreatic necrosis virus (IPNV). Dis. Aquat. Org., 4, 205–209.
- Hauck, A.K., Marshall, M.R., Li, J.K.K., Lee, R.A. (2001). A new finding and range extension of bacilliform virus in the freshwater red claw crayfish in Utah, USA. J. Aquat. Anim. Health., 13, 158–162.
- Herbert, B. (1987). Notes on diseases and epibionts of *Cherax quadricarinatus* and *C. tenuimanus* (Decapoda: Parastacidae). *Aquaculture*, 64, 165–173.
- Hirt, R.P., Logsdon, J.M., Healy, B., Dorey, M.W., Doolittle, W.F., Embley, T.M. (1999). Microsporidia are related to Fungi: evidence from the largest subunit of RNA polymerase II and other proteins. *Proc. Natl. Acad. Sc.*, 96, 580–585.
- Huang, J., Song, X.L., Yu, J., Yang, C.H. (1995) Baculoviral hypodermal and hematopoietic necrosis-study on the pathogen and pathology of the explosive epidemic disease of shrimp. *Mar. Fish Res.*, 16:1–10.
- Hubault, E. (1935). Une epizootie sur Potamobius pallipes Lereboullet. Ann. Parasitol. Hum. Comp., 2, 109-112.
- Huner, J.V. (1994). Freshwater Crayfish Aquaculture in North America, Europe, and Australia: Families Astacidae, Cambaridae, and Parastacidae, *Food Products Press*, s. 312.
- Huner, J.V. (1995). An overview of the status of freshwater crayfish culture. J. Shellfish Res., 14(2): 539-543.
- Inouye, K., Miwa, S., Oseko, N., Nakano, H., Kimura, T., Momoyama, K., Hiraoka, M. (1994) Mass mortalities of cultured kuruma shrimp *Penaeus japonicus* in Japan in 1993: electron microscopic evidence of the causative virus. *Fish Pathol.*, 29:149–158.
- Jiravanichpaisal, P., Eakaphun, B., Kenneth, S., Irene S. (2001) Experimental infection of white spot syndrome virus in freshwater crayfish Pacifastacus leniusculus. *Dis Aquat Org.*, 47: 151–157.
- Jones, J.B., Lawrence, C.S. (2001). Diseases of yabbies (Cherax albidus) in Western Australia. Aquaculture, 194, 221-232.
- Kageyama, D., Anbutsu, H., Shimada, M., Fukatsu, T. (2007). Spiroplasma infection causes either early or late male killing in Drosophila, depending on maternal host age. *Naturwissenschaften*, 94, 333–337.
- Kalaycı, G. and Akhan, S. (2016). Molecular Identification of *Astacus leptodactylus* and *Austropotamobius torrentium* Using mtDNA-RFLP Method. *Turkish Journal of* Fisheries and Aquatic Sciences, 16: 789-795.
- Keller, T.A. (1992). The effect of the branchiobdellid annelid *Cambarincola fallax* on the growth rate and condition of the crayfish *Orconectes rusticus. J. Freshwat. Ecol.*, 7, 165–171.

- La Fauce, K., Owens, L. (2007). Investigation into the pathogenicity of *Penaeus merguiensis* densovirus (PmergDNV) to juvenile *Cherax quadricarinatus. Aquaculture*, 271, 31–38.
- Lee, J.H., Kim, T.W., Choe, J.C. (2009). Commensalism or mutualism: conditional outcomes in a branchiobdellid-crayfish symbiosis. *Oecologia*, 159, 217–224.
- Lightner, D.V., Redman, R.M., Poulos, B.T., Nunan, L.M., Mari, J.L., Hasson, K.W. (1997) Risk of spread of penaeid shrimp viruses in the Americas by the international movement of live shrimp for aquaculture and frozen shrimp for commodity markets. *Rev. Sci. Tech. Off Int. Epizoot.*, 16:146–160.
- Lo, C.F., Ho, C.H., Chen, C.H., Liu, K.F., Chiu, Y.L., Yeh, P.Y., Peng, S.E., Hsu, H.C., Liu, H.C., Chang, C.F., Su, M.S., Wang, C.H., Kou, G.H. (1997). Detection and tissue tropism of white spot syndrome baculovirus (WSBV) in captured brooders of *Penaeus* monodon with a special emphasis on reproductive organs. *Dis. Aquat. Org.*, 30, 53–72.
- Longshaw, M. (2011). Diseases of crayfish: A review. Journal of Invertebrate Pathology, 106: 54-70.
- Mazlum, Y. and Yılmaz. E. (2006). Türkiye'de Önemli Kerevit Türlerinin Yetiştiriciliği. E.Ü. Su Ürünleri Dergisi, 23(1-2): 201-205.
- Mohan, C.V., Shankar, K.M., Kulkarni, S, Sudha, P.M. (1998) Histopathology of cultured shrimp showing gross signs of yellow head and white spot syndrome during 1994 India epizootics. *Dis. Aquat. Org.*, 34:9–12.
- Momoyama, K., Hiraoka, M., Nakano, H., Inouye, K., Oseko, N. (1994) Mass mortalities of cultured kuruma shrimp, *Penaeus japonicus*, in Japan in 1993: histopathological study. *Fish Pathol.*, 29:141–148.
- Morado, J.F. (1995). Ciliate parasites and related diseases of Crustacea: a review. Rev. Fish. Sci., 3, 275-354.
- Nakano, H., Koube, H., Umezawa, S., Momoyama, K., Hiraoka, M., Inouye, K., Oseko, N. (1994) Mass mortalities of cultured Kuruma shrimp, *Penaeus japonicus*, in Japan in 1993: epizootiological survey and infection trials. *Fish Pathol.*, 29:135–139.
- Nicky, B. (2008). Crayfish Plague Australia and New Zealand Standard Diagnostic Procedure. Sub-Committee on Animal Health Laboratory Standards.
- Owens, L., McElnea, C. (2000). Natural infection of the redclaw crayfish *Cherax quadricarinatus* with presumptive spawner-isolated mortality virus. *Dis. Aquat. Org.*, 40, 219–223.
- Park, J.H., Lee, Y.S., Lee, S., Lee, Y. (1998) An infectious viral disease of penaeid shrimp newly found in Korea. *Dis. Aquat. Org.*, 34:71-75.
- Quaglio, F., Morolli, C., Galuppi, R., Bonoli, C., Marcer, F., Nobile, L., De Luise, G., Tampieri, M.P. (2006a). Preliminary investigations of disease-causing organisms in the white-clawed crayfish *Austropotamobius pallipes* complex from streams of northern Italy. *Bull. Fr. Pêche Piscic.*, 380–381, 1271–1290.
- Quaglio, F., Morolli, C., Galuppi, R., Tampieri, M.P., Bonoli, C., Marcer, F., Rotundo, G., Germinara, G.S. (2006b). Sanitarypathological examination of red swamp crayfish (*Procambarus clarkii*, Girard 1852) in the Reno Valley. *Freshwater Crayfish*, 15, 1–10.
- Ragan, M.A., Goggin, C.L., Cawthorn, R.J., Cerenius, L., Jamieson, A.V.C., Plourde, S.M., Rand, T.G., Söderhäll, K., Gutell, R.R. (1996). A novel clade of protistan parasites near the animal–fungal divergence. *Proc. Natl. Acad. Sci.*, 93, 11907–11912.
- Royo, F., Gonzales-Cienfuegos, G., Muzquiz, J.L. (2002). Preliminary observation of occasional mortality in the population of freshwater crayfish in the Picos de Europa (Asturias, Spain). Bull. Fr. Pêche Piscic., 367, 935–941.
- Shi, Z., Huang, C., Zhang, J., Chen, D., & Bonami, J. R. (2000). White spot syndrome virus (WSSV) experimental infection of the freshwater crayfish, *Cherax quadricarinatus. Journal of Fish Diseases*, 23(4), 285–288.
- Stentiford, G.D., Bateman, K., Feist, S.W. (2004). Pathology and ultrastructure of an intranuclear bacilliform virus (IBV) infecting brown shrimp *Crangon crangon* (Decapoda: Crangonidae). *Dis. Aquat. Org.*, 58, 89–97.
- Stephens, F., Buller N., Alderman D., Cameron A., DoroudiM., Suijdendorp P., Deveney M. & Bowate R. (2005). Disease strategy: Crayfish plague (Version 1.0). In: Australian Aquatic Veterinary Emergency Plan(AQUAVETPLAN), Edition 2. Australian Government Department of Agriculture, Fisheries and Forestry, Canberra, ACT, Australia, Available at: http://www.agriculture.gov.au/animal/aquatic/aquavetplan/crayfish-plague.
- Tan, C.K., Owens, L. (2000). Infectivity, transmission and 16S rRNA sequencing of a rickettsia, *Coxiella cheraxi sp.* nov., from the freshwater crayfish *Cherax quadricarinatus*. Dis. Aquat. Org., 41, 115–122.
- Taştan, Y., Çağatay, İ.T., Akhan, S., (2017) Tatlısu ıstakozundaki (*Astacus leptodactylus*) hastalık etkeni *Fusarium oxysporum*'un teşhisi", II. Uluslararasi Turizm ve Mikrobiyal Gıda Güvenliği Kongresi, Antalya, Türkiye, 13-14 Aralık 2017, pp61-62.
- Thune, R.L., Hawke, J.P., Sebeling, R.J. (1991). Vibriosis in the red swamp crawfish. J. Aquat. Anim. Health, 3, 188-191.
- TÜİK (2018). Su ürünleri istatistikleri 2018. https://biruni.tuik.gov.tr/medas/?kn=97&locale=tr.
- Tulsidas, H., Ong, Y.Y., Chan, K.C. (2008). Aeromonas hydrophila bacteraemia and portal pyaemia. Singapore Med. J., 49, 346-348.
- Unestam, T. (1969). Resistance to the crayfish plague in some American, Japanese and European crayfishes. *Rep. Inst. Freshw. Res. Drottningholm*, 49: 202–209.
- Unestam, T. (1972). On the host range and origin of the crayfish plague fungus. Rep. Inst. Freshw. Res. Drottningholm, 52: 192–198.

- Unestam, T. (1975). Defence reactions in and susceptibility of Australian and New Guinean freshwater crayfish to European-crayfishplague fungus. *Aust. J. Exp. Biol. Med. Sci.*, 53: 349–359.
- Unestam, T., and Weiss, D.W. (1970). The host-parasite relationship between freshwater crayfish and the crayfish disease fungus *Aphanomyces astaci*: Responses to infection by a susceptible and a resistant species. *J. Gen. Microbiol.*, 60: 77–90.
- Vey, A. (1979). Infections fongiques chez l'ecrevisse Astacus leptodactylus Esch. Freshwater Crayfish, 4, 403.
- Vey, A. (1981). Les maladies des écrevisses, leur reconnaissance et la surveillance sanitaire des populations astacicoles. Bull. Fr. Pêche Piscic., 223–236.
- Vogt, G., Rug, M. (1995). Microscopic anatomy and histochemistry of the crayfish parasite *Psorospermium haeckeli*. *Dis. Aquat. Org.*, 21, 79–90.
- Vogt, G., Rug, M. (1999). Life stages and tentative life cycle of *Psorospermium haeckeli*, a species of the novel DRIPs clade from the animal–fungal dichotomy. J. Exp. Zool. Part A, 283, 31–42.
- Wang, Q., White, B.L., Redman, R.M., Lightner, D.V. (1999) Per os challenge of *Litopenaeus vannamei* postalarvae and *Farfantepenaeus duorarum* juveniles with six geographic isolates of white spot syndrome virus (WSSV). Aquaculture, 170:179–194.
- Wang, Q., Nunan, L.M., Lightner, D.V. (2000) Identification of genomic variations among geographic isolates of white spot syndrome virus using restriction analysis and Southern blot hybridization. *Dis. Aquat. Org.*, 43:175–181.
- Wongteerasupaya, C., Vickers, J.E., Sriurairatana, S., Nash, G.L., Akarajamorn, A., Boonsaeng, V., Panyim, S., Tassanakajon, A., Withyachumnarnkul, B., Flegel, T.W. (1995). A nonoccluded, systemic baculovirus that occurs in cells of ectodermal and mesodermal origin and causes high mortality in the black tiger prawn, *Penaeus monodon. Dis.Aquat. Org.*, 21: 69–77.



# Effects of Broad Bean Meal Usage in Fish Feed on Growth, Feed Evaluation and Body Composition in Sea Bass (*Dicentrarchus labrax*)

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Abstract: In this study, feeding experiments were carried out with different percentages (10-20-30-50%) of broad bean meal (Vicia faba L.) in sea bass fingerling feeds.  $1.33 \pm 0.2$  grams sea bass were used in the experiment. Sea bass fingerlings were fed for 45 days with this diet and the experiment was conducted as triplicates. At the end of the experiment, growth performance, body composition and some biochemical parameters of these fish were investigated. In the study, a control group and four groups of diet containing different rates of broad bean meal (10-20-30-50%) were used instead of fish meal. At the end of the research, the best growth and feed conversion were observed in the control group. Among the experimental groups, the best growth was observed in the group using 30% broad bean meal. Similarly, the feed conversion rate was found to be higher in the groups fed with 20-30% experimental feed than the others. There was no difference in body composition between the groups. At the end of the trials, protein, moisture, lipid and ash ratios were not different between the experimental groups. As a result, it was determined that 20-30% broad bean meal can be used in sea bass diets when the cost of the feed used in the feeding experiment and growth performance of fish and body composition were examined

Keywords: Sea bass, broad bean, feed, growth parameters

Acknowledgments: This study is a part of MSc thesis of Hüseyin SAKA (Effects of Broad Bean Meal Usage in Fish Feed on Growth, Feed Evaluation and Body Composition in Sea Bass (*Dicentrarchus labrax*)) from Graduate School of Natural and Applied Sciences, Department of Aquaculture, at Çanakkale Onsekiz Mart University.



# Determination of Genetic Structures of Ruditapes Genus in from Musakca in the South of Marmara Sea

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**Abstract:** Turkish waters have many bivalve species with high economic value. However, as a result of uncontrolled fishing and variable environmental factors, and the invasion of invasive species, the distribution and quantities of local endemic species are under significant threat. Turkey is hosting two economically important bivalve species. One of them is a native clam species, *Ruditapes decussatus* (Linnaeus 1758), and the other is an alien species, *Ruditapes philippinarum* (Adams and Reeve 1850), which entered Turkish waters from Japanese waters by different means. *R. philippinarum*, which is resistant to wide temperature and diseases, salinity intervals, is the most suitable for aquaculture. Despite it has three times lower value than the native clam, alien species distributed at different point of Turkey. Particularly it is more common in the Sea of Marmara. Species-specific molecular markers (ITS1, ITS2 and 16SmtDNA) were used to determine the species of collected clam individuals from the coast of Musakca in the south of Marmara Sea. The results of band sizes showed that collected individuals were *Ruditapes philippinarum*. Also, genotypic descriptions were found similarly with morphologically observations.

Keywords: Ruditapes philippinarum, Ruditapes decussatus, ITS1, ITS2, 16SmtDNA

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# Bioaccumulation of Toxic Metals in *Rapana venosa* and Sediments from Sinop Shores of the Black Sea

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**Abstract:** The Black Sea ecosystem for remediation and protection, thus metal data from this region is needed. In the present study, toxic heavy metals (Cd, Hg and Pb) in *Rapana venosa* and sediments in Sinop coasts of the Black Sea were investigated. Toxic metals analysis was performed by ICP-MS. The heavy metal levels in sediments occurred in descending order of Pb > Hg > Cd during sampling period. The same pattern was observed in sea snail. Cd and Hg levels in sea snail were higher than those in sediment, whereas Pb levels were high in sediment. The levels of toxic metal levels in sediments of Sinop shores of the Black Sea were much lower than the amounts pointed out the sediment quality. The concentration ranges found in sea snails for these toxic elements, when expressed on a wet weight basis, were as follows: not detected (ND) to 0.0125 mg kg<sup>-1</sup> for Cd, ND to 0.0141 mg kg<sup>-1</sup> for Hg and ND to 0.705 mg kg<sup>-1</sup> for Pb. The levels of these toxic elements in *R. venosa* were relatively low when compared to those studied in other parts of the Black Sea coasts and is able to meet the legal standards set by Turkish Food Codex and the European Commission Regulation using the maximum levels for certain contaminants in foodstuffs. The amounts of toxic metals in sea snail were found to be within the safe limits for people consumption.

Key words: Black Sea, Sinop Peninsula, Akliman, toxic metals, Rapana venosa, sediment

# **1. INTRODUCTION**

The Black Sea is connected to the Aegean Sea and Mediterranean Sea by the Strait of Bosporus. The Black Sea is a semiclosed formation and heavy discharges of the surrounding domestic and agriculture and industries via major rivers have been ongoing for many decades. Sediments are major sinks for various contaminants especially heavy metals. In recent review, *R. venosa* has the capability of accumulating heavy metals (Bat and Arici, 2018) and is thus a suitable bio-indicator for environmental metal pollution (Bat et al., 2000; 2016; 2018a; Bat and Öztekin, 2016). Heavy metals especially nonessential ones have a particular important in ecotoxicology, since they are highly persistent and all have the potential to be toxic to living organisms even at very low levels.

Over the past few decades, heavy metals have been discharged into the Black Sea as a result of the rapid industrialization and agricultural activities (Bat et al., 2018b). The effects of heavy metals in the environment depend to a large extent on whether they occur in forms that can be taken up by biota. Therefore, this study was undertaken to evaluate the toxic elements in the sediments and sea snails.

# 2. MATERIAL AND METHODS

Sea snails and surficial sediments were collected from Akliman of Sinop shores in the Black Sea in summer months (June, July and August) of the year 2016. Akliman is located in Sinop coasts (Figure 1) and there is no any industry in this region. People activities here include only local fisheries and touristic activities.



Figure 1. Study area

Sea snails were collected by SCUBA diver. After sampling, sea snails were cleaned well by rinsing with bi-distilled water to remove debris, plankton and other external adherent. Then they were put in an aquarium with seawater from collection site for 24 hours to allow clean for gut content (Bat et al., 2000, 2016, 2018a; Bat and Öztekin, 2016). Only the edible portions of sea snails were measured. Ten individuals of each sampling months were used for analysis. Homogeneous samples of biological sample for analysis were rinsed with deionized water. The samples digested with Suprapur® HNO<sub>3</sub> (using a microwave digestion system (Milestone Systems, Start D 260).

The sediment samples were dried at 105 °C for 24 hours and 0.25 g of the dried sediment were digested with Seawater Sediment HPR-EN-33 methodology (Milestone Systems, Start D 260).

Toxic metals were determined with ICP-MS (Agilent Technologies, 7700X), used three replicates and results of mean concentrations were detected as mg/kg dry wt. in sediments and mg/kg wet wt. sea snail samples. When metals levels in sediment compared with the sea snails, dry wt. was converted to wet wt.

#### **Statistical Analysis**

Data were analysed using the one-way analysis of variance (ANOVA) and group means were compared using Duncan's multiple range test. P values < 0.05 were considered significant. In addition the weekly intake levels set by international organizations for allowable values were calculated using the maximum toxic metal amounts in sea snail. Estimated Weekly Intakes (EWI) = maximum levels of Cd, Hg and Pb (mg/kg wet wt.) multiplied by seafood consumption (kg/70 kg body weight/week).

# 3. RESULTS AND DISCUSSION

#### Results

The certified reference material used for the determination of the limits of detection of cadmium, mercury and lead was 2702 (marine sediment) and 2976 (mussel tissue) from National Institute of Standards and Technology (NIST) for the sea snail samples. The recoveries of the toxic elements found in certified reference materials were 103% for Cd, 106% for Hg and 96% for Pb where the relative standard deviations were less than 6%, indicating good agreement and all analyses were considered satisfactory.

The results showed that concentrations of the toxic metals in sediments were higher than those in sea snails. Pb was the highest concentration in samples, followed by Hg and least was Cd (Figure 2).



Figure 2. Pb, Cd and Hg concentrations (mean ± SD) in sediment and sea snail (mg / kg dry wt.)

The amounts of toxic metals in sea snail were found to be within the accepted safe limits for people consumption (Table 1).

Table 1. Accepted safe levels of exposure to toxic metals

| Metals                                    | Standard                                  | Reference                |  |  |
|---|---|--------------------------|--|--|
| Cd  | PTWI of 0.007 mg per kg body wt. per week | WHO, 1989; WHO, 2004     |  |  |
| Hg  | PTWI of 0.004 mg per kg body wt. per week | FAO/WHO, 2011            |  |  |
| Pb  | PTWI of 0.025 mg per kg body wt. per week | WHO, 2000; FAO/WHO, 2010 |  |  |
| DTWL Provisional Talarahla Waaldy Intalya |   |                          |  |  |

PTWI: Provisional Tolerable Weekly Intake

Table 2 shows the calculation PTWI values of Cd, Hg and Pb for a 70 kg reference person by the Joint Food and Agricultural Organization for the United Nations (FAO) / World Health Organization (WHO) Expert Committee on Food Additives (JECFA) (FAO/WHO, 2010).

**Table 2.** PTWI values of Cd, Hg and Pb for a 70 kg adult person and Estimated Weekly Intakes (EWI) of these toxic metals in tissue of, R. venosa.

| Metals | Standard                                   | EWIs (maximum) |
|--------|--|----------------|
| Cd     | Maximum allowable PTWI value of 0.49 mg Cd | 0.0000875      |
| Hg     | Maximum allowable PTWI value of 0.28 mg Hg | 0.0000987      |
| Pb     | Maximum allowable PTWI value of 1.75 mg Pb | 0.0049350      |

#### Discussion

Sediment quality guideline values (SQGV) are one of the most significant tools to protect marine ecosystem from damage effects. The results of this study compared to those obtained by the recommended SQGV (Simpson and Batley, 2016). The levels of toxic metal levels in sediments of Akliman shores of Sinop Peninsula were much lower than the amounts pointed out the sediment quality. SQGV and SQGV-high for Hg, Cd and Pb are 0.15-1.0, 1.5-10 and 50-220 mg/kg dry wt., respectively (Simpson and Batley, 2016).

Sea snails are carnivore and spend more time in sediments where metals present. They are generally feed on mussels in the Black Sea, thus expected to concentrate more metals than other biota. Concentrations of toxic heavy metals in seafood are of particular interest because of the potential risk to people who consume them.

The concentration ranges found in sea snails for these toxic elements, when expressed on a wet weight basis, were as follows: not detected (ND) to 0.0125 mg / kg for Cd, ND to 0.0141 mg / kg for Hg and ND to 0.705 mg / kg for Pb. The levels of these toxic elements in *R. venosa* were relatively low when compared to those studied in other coasts of the Turkish Seas (Bat and Arici, 2018) and is able to meet the legal standards set by Turkish Food Codex (2002) and the European Commission Regulation (2006) using the maximum levels for certain contaminants in foodstuffs. The amounts of toxic heavy metals in *R. venosa* from Akliman shores of Sinop were found to be within the accepted safe limits for human consumption.

The Provisional Tolerable Weekly Intake (PTWI) value is an estimate of the amount of a contaminant that can be consumed by human over a lifetime without appreciable risk. In the present study the calculation PTWI values of Cd, Hg and Pb for a 70 kg reference person by the Joint Food and Agricultural Organization for the United Nations (FAO) / World Health Organization (WHO) Expert Committee on Food Additives (JECFA) (FAO/WHO, 2010) was calculated. The average daily Mollusca consumption in Turkey is 1 g per person, which are equivalent to 7 g/week for Turkey (FAO, 2010). As a result, the weekly intakes of the studied toxic metals in *R. venosa* in Akliman per kg of body amounts not exceeded the Provisional Tolerable Weekly Intake (PTWI).

#### 4. CONCLUSION

Biomonitoring studies can be employed to demonstrate geographical and temporal variations in the bio-availabilities of toxic elements in the marine ecosystem. Sea snails have been already used as bio-indicator as it contains variable levels of the heavy metals.

The data of toxic metal levels in sea snails are very important with respect to protect the marine ecosystem, human consumption of these species and to determine the well-nigh useful biomonitor species.

Overall, it can be concluded from this study that the tissues of sea snails contain toxic metals less than the sediments and is safe for people consumption according to EU and Turkish regulations.

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#### REFERENCES

- Bat L & Arici E (2018) Heavy Metal Levels in Fish, Molluscs, and Crustacea from Turkish Seas and Potential Risk of Human Health. In: Food Quality: Balancing Health and Disease, 159-196.
- Bat L, Gönlügür G, Andaç M, Öztürk M, & Öztürk M (2000). Heavy metal concentrations in the sea snail *Rapana venosa* (Valenciennes, 1846) from Sinop coasts of the Black Sea. Turkish J. Mar. Sci., 6(3): 227-240.
- Bat L, Arıcı E, Sezgin M, & Şahin F (2016). Heavy metals in edible tissues of benthic organisms from Samsun coasts, South Black Sea, Turkey and their potential risk to human health. Journal of Food and Health Science, 2(2): 57-66.
- Bat L, & Öztekin H C (2016). Heavy metals in *Mytilus galloprovincialis, Rapana venosa* and *Eriphia verrucosa* from the Black Sea coasts of Turkey as bioindicators of pollution. Walailak Journal of Science and Technology, 13(9): 715-728.
- Bat L, Şahin F, & Öztekin A (2018a). Toxic elements in edible mollusks from Igneada coasts of the Black Sea, Turkey. Korean Journal of Food & Health Convergence, 4(3): 22-31.
- Bat L, Öztekin A, Şahin F, Arıcı E, & Özsandıkçı U (2018b). An overview of the Black Sea pollution in Turkey. MedFAR., 1(2): 67-86.
- FAO (2010). The food consumption refers to the amount of food available for human consumption as estimated by the FAO Food Balance Sheets.
- FAO/WHO (2010). Summary report of the seventy-third meeting of JECFA. Joint FAO/WHO Expert Committee on Food Additives Geneva.
- FAO/WHO (2011). Food standards programme codex committee on contaminants in foods. Fifth Session Codex Alimentarius Commission. The Hague, The Netherlands.
- European Commission Regulation (2006). Setting maximum levels for certain contaminants in foodstuffs. No 1881/2006 of 19 December 2006.
- Simpson S L, & Batley G E (2016). Sediment quality assessment: a practical guide, Second Edition, CSIRO Publishing Australia.
- Turkish Food Codex (2002). Official Gazette of Republic of Turkey. Notifications about determination of the maximum levels for certain contaminants in foodstuffs of Turkish Food Codex. (Notification No: 2002/63), Issue: 24885.
- WHO (1989). Evaluation of certain food additives and contaminants. Report of the Thirty-Third of the Joint FAO/WHO Expert Committee on Food Additives. Technical Report Series No. 776 Geneva.
- WHO (2000). Evaluation of certain food additives and contaminants. Report of the Fifty-Third of the Joint FAO/WHO Expert Committee on Food Additives. Technical Report Series No. 896 Geneva.
- WHO (2004). Evaluation of certain food additives and contaminants. Report of the Sixty-First of the Joint FAO/WHO Expert Committee on Food Additives. Technical Report Series No. 922 Geneva.



# Health Risk Assessment of Heavy Metals in Scomber japonicus Houttuyn, 1782

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Abstract: Toxic metals (cadmium, mercury and lead) and essential trace elements (copper and zinc) were analysed in liver and edible tissues and entire gills of Chub mackerel collected from Sinop shores of the Black Sea using ICP-MS (Inductively Coupled Plasma – Mass Spectrometer) after wet digestion. The heavy metal amounts ranged from 0.022 to 0.058, 0.029 to 0.094, 0.058 to 0.21, 0.39 to 2.1 and 9 to 27 mg kg<sup>-1</sup> wet wt. for Cd, Hg, Pb, Cu and Zn, respectively. In general, the liver samples had the highest level of Cu and Zn, while gills samples showed the highest content of Pb, Hg and Cd as compared with muscle samples. Heavy metal levels in muscle tissues were less than the Turkish and European standard limits. The estimation of human health risk for adults revealed an estimated daily intake (EDI) value of muscle below threshold of oral reference dose (R<sub>f</sub>D) for all metals analysed. The hazard index (HI) and Target Hazard quotient (THQ) for all the analysed metals were below 1, demonstrating that human health risk through consumption of fish muscles is not possible.

Keywords: Sinop, heavy metal, Scomber japonicus, Black Sea, hazard index, target hazard quotient

# **1. INTRODUCTION**

Heavy metals are high density non-biodegradable metallic elements owning enduring toxicant impacts. They result from in the coastal environment are transferred to the marine biota thanks to different ways and in the end accumulate in food chains. This may reason physiological and morphological changes both in biota and in people via consumption. These can cause carcinogenic, cytotoxic and mutagenic effects or even death. Fish are major component of marine food chains which accumulate big levels of heavy metals from polluted waters. Heavy metals being a major pollution of marine coastal ecosystem have already become a serious problem in the Black Sea (Bat and Arici, 2018). Turkey, like other the Black sea countries, is facing severe problem of coastal pollution from anthropogenic activities as the industrial waste water discharges via rivers and domestic wastes without any treatment (Bat et al., 2018). On one hand, the catchment of commercially important fish are steadily declining from the Black Sea coasts due to pollution, overfishing and other anthropogenic activities while on other hand the quality of fish is increasingly becoming inclined to heavy metal, plastics and other chemicals contamination (Bat et al., 2017; Bat et al., 2018). With the in any way increasing pollution in marine coastal areas, the risk of fish contaminated with heavy metal is increasing day-to-day (Bat, 2014; Bat, 2017). Accumulation of heavy metals in pelagic fish seems to be a region specific occurrence, depending upon some aquatic components like water column, plankton and suspended matters. Many rivers bring a lot of metal contaminants and other untreated industrial and domestic wastes from cities to the Black Sea coasts. Thus, there is a necessity in order to update data on contaminant levels in fish of Turkey. This work supplies info on the metal levels in muscle liver and entire gills of S. japonicus in Sinop coasts of the Black Sea coast of Turkey, and compares the results with national and international regulations. The heavy metals investigated include zinc, copper, cadmium, lead and mercury because they are known to accumulate in the environment and in fish, and are known to cause adverse health effects if consumed in sufficient quantities.

# 2. MATERIALS AND METHODS

The accumulations of Zn, Cu, Pb, Hg and Cd were studied in dorsal muscles, liver and gills of *Scomber japonicus* (Figure 1) collected from Sinop coasts of the Black Sea in November and December of 2018 (Figure 2). Fish samples were cleaned with bi-distilled water and then dissected. Muscle, entire gills and liver tissues of the fish were prepared for analysis according to the method described by Bernhard (1976). Sample preparation and analytical determination of heavy metals was made following the recommendations given by the UNEP (1984) and UNEP (1985).



Figure 1. Scomber japonicus Houttuyn, 1782



Figure 2. Sampling region

Heavy metals (Hg, Cd, Pb, Cu and Zn) in edible tissues were determined by ICP-MS (Agilent 7700x). A certified reference material (TORT-3 lobster hepatopancreas) was processed along with samples to determine the accuracy of the method and the results were compared to the acceptable limits. The recovery of the studied heavy metals was ranged between 97 and 105%.

#### Statistical analysis

Analysis of variance (ANOVA) was performed for heavy metals amounts, followed by the post-hoc Tukey multiple comparison test (p<0.05) when appropriate (Zar, 1999), using the SPSS 21 software. The estimation was carried out at significant level of 0.05. The measured concentrations are expressed in mg kg<sup>-1</sup> wet wt.

Target hazard quotient (THQ), which is a ratio of the estimated daily intake (EDI) to the oral reference dose (Rf.D), is used to assess the potential non-carcinogenic risk of the consumers of the perceived contaminated fish as food.

The EDI of the heavy metals was found employing the equation:

$$EDI = \frac{C_{metal} \times IR}{b.wt}$$

Where: C metal is the amounts of the metals in edible tissues of fish; IR appears for the daily mean intake; b. wt. is the body weight of 70 kg for adults.

THQ was estimated as following the equation:

$$THQ = \frac{C_{metal} \times IR \times 10^{-3} \times EF \times ED}{Rf.D \times BW \times ATn}$$

Where, C <sub>metal</sub> is the metal concentration in fish (mg kg<sup>-1</sup>), IR is the fish ingestion rate (g day<sup>-1</sup>), EF is the exposure frequency (day year<sup>-1</sup>) or number of exposure events per year of exposure, ED is the exposure duration, total for adult

(year), Rf.D is the reference dose mg kg<sup>-1</sup> day<sup>-1</sup>, BW is the body weight (adult, kg), and ATn is the averaging time, non-carcinogens (day year-1).

The Rf. D for Hg, Cd, Pb, Cu and Zn are 0.0003, 0.001, 0.002, 0.04 and 0.3 (mg kg-1) day-1, respectively (U.S. EPA, 2016). Health risk assessment of consumers from the intake of metal-contaminated fish was characterized by using the THQ. THQ < 1 means the exposed population is unlikely to experience some non-carcinogenic adverse effects during the life time. Otherwise, THQ > 1 means that there is a chance of non- carcinogenic effects, with an increasing probability as the value increases. The hazard index (HI) from THQs can be expressed as the sum of the hazard quotients (USEPA, 2011). It is calculated as following the equation:

$$HI = THQ_{Zn} + THQ_{Cu} + THQ_{Pb} + THQ_{Ha} + THQ_{Cd}$$

The annual quantity of fish consumed is 7.6 kg person-1 in 2018 (TUIK, 2019), which is approximately to 20.8 g day-1 for Turkey. The body weight of adult person is 70 kg.

#### 3. RESULTS AND DISCUSSION

#### Results

Mean amounts of studied heavy metal (mg kg<sup>-1</sup>  $\pm$  SD wet wt.) found in fish species are given in Figure 3. In gills, liver and edible tissues Zn is found in highest concentrations, followed by Cu. Metal accumulations were higher in liver and gills of Chub mackerel compare to edible tissues. The concentration of Cd, Hg and Pb were highest in gills; whereas Cu and Zn were higher in liver.



**Figure 3.** Mean levels and standard deviation of Zn, Cu, Pb, Hg and Cd found in gills, liver and muscle of *Scomber japonicus* from Sinop shores of the Black Sea in 2018.

#### Discussion

The results of this study are generally supported in the literature. Liver has been considered to be the inner organs for Cd and Pb accumulation in fish. Bat (2014) reported highest levels of the heavy metals were found in the liver tissues in many fish species. The higher amounts of these metals in liver relative to edible tissues were attributed to the affinity to metallothionein protein (Kotze et al., 1999; Ahmed et al., 2017). On the other hand low levels of the heavy metals in edible tissues of fish appear to be due to low levels of binding proteins in those tissues. The higher heavy metals levels in the gills could be owing to the fact that these organs serve as the respiratory in fishes through which metal ions are absorbed. The gills are directly touch with the polluted surrounding water and have the finest epithelium of all the organs and metals can pass through the fine epithelia cells (Bebianno et al. 2004). Usually gills have high activities and could be much sensitive tissues in reflecting the general oxidative stress from the surrounding water contaminated with heavy metal. The levels of heavy metals in gills may reflect their levels in the surrounding water of the fish habitats. In the present study, heavy metal amounts were higher in entire gills and liver tissues compare to muscles of fish.

Fish are constantly at the upper food chains in marine ecosystem and may concentrate high amounts of some metals from the water, particles and sediment. The bioaccumulation and their risk assessment of heavy metals from Turkish waters have been reviewed (Bat, 2014; Bat, 2017; Bat and Arici, 2018). Mean levels of Cd, Hg, Pb and Cu were lower in Sinop coasts when compared with other localities, while Zn individually had similar accumulation levels in all costs with no significant differences.

The THQs of Cd, Hg, Pb, Cu and Zn were given in Table 1. The observed values of THQs were lower than the safe standard and HI of these metals was 0.0442, far from 1 in *S. japonicus*, indicated that ingestion of this fish from Sinop shores of the Black Sea did not result in overexposure of these metals. Nonetheless, the estimation of THQs conducted in the present study showed that adverse health effects may not occur when considering fish consumption patterns.

| Metals             | Rf. D. EDI EWI                                   |  |  |             |
|--------------------|--|--|--|-------------|
|                    | (mg kg <sup>-1</sup> body wt.) day <sup>-1</sup> | (mg kg <sup>-1</sup> body wt.) day <sup>-1</sup> | (mg kg <sup>-1</sup> body wt.) day <sup>-1</sup> | ТНQ         |
| Cd                 | 0.001  | 0.0004576  | 0.0032032  | 0.006537143 |
| Hg                 | 0.0003   | 0.0006032  | 0.0042224  | 0.017234286 |
| Pb                 | 0.002  | 0.0012064  | 0.0084448  | 0.008617143 |
| Cu                 | 0.04   | 0.008112   | 0.056784   | 0.002897143 |
| Zn                 | 0.3  | 0.1872   | 1.3104   | 0.008914286 |
| Hazard index (HI)= |  |  |  | 0.0442      |

Table 1. Maximum metal levels (mg kg-1 wet wt.) in the muscles of S. japonicus and EDIs and HIs values for consumers.

#### 4. CONCLUSION

Understanding of heavy metal levels in fish is considerable with regard to marine environment health, wellbeing of biota and people consumption of fish. Consequently effect on all aspects of heavy metal levels and their impact on fish should be monitored occasionally to ahead of understand the effects of heavy metals on fish development and the present conditions of population dynamics.

This study provides new information on the concentration of heavy metals in Chub mackerel from Sinop shores of the Black Sea Chub mackerel had metal levels below the guideline values established by the Turkish Food Codex (2002) and European Commission Regulation (2006).

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#### REFERENCES

- Ahmed Q, Bat L & Ali Q M (2017). Bioaccumulation of nine heavy metals in some tissues of Anodontostoma chacunda (Hamilton, 1822) in the Arabian Sea coasts of Pakistan. Natural and Engineering Sciences, 2(3): 79-92.
- Bat L (2014). Heavy metal pollution in the Black Sea. In: Düzgüneş E, Öztürk B, Zengin M. (Eds.). Turkish Fisheries in the Black Sea. Published by Turkish Marine Research Foundation (TUDAV), Publication number: 40, ISBN: 987-975-8825-32-5 Istanbul, Turkey, p. 71-107.
- Bat L (2017). The Contamination Status of Heavy Metals in Fish from the Black Sea, Turkey and Potential Risks to Human Health. In: Sezgin, M., Bat, L., Ürkmez, D., Arıcı, E., Öztürk, B. (Eds.) Black Sea Marine Environment: The Turkish Shelf. Turkish Marine Research Foundation (TUDAV), Publication No: 46, ISBN- 978-975-8825-38-7, Istanbul, TURKEY, pp. 322-418.
- Bat L, Öztekin A & Arıcı E (2017). Marine Litter Pollution in the Black Sea: Assessment of the Current Situation in Light of the Marine Strategy Framework Directive. In: Sezgin M, Bat L, Ürkmez D, Arıcı E & Öztürk B (Eds.) Black Sea Marine Environment: The Turkish Shelf. Turkish Marine Research Foundation (TUDAV), Publication No: 46, ISBN- 978-975-8825-38-7, Istanbul, TURKEY, pp. 476-494
- Bat L & Arici E (2018). Chapter 5. Heavy Metal Levels in Fish, Molluscs, and Crustacea From Turkish Seas and Potential Risk of Human Health. In: Holban AM, Grumezescu AM. (Eds.) Handbook of Food Bioengineering, Volume 13, Food Quality: Balancing Health and Disease. Elsevier, Academic Press, ISBN: 978-0-12-811442-1, pp. 159-196. http://dx.doi.org/10.1016/B978-0-12-811442-1.00005-5

Bat L, Öztekin A, Şahin F, Arıcı E & Özsandıkçı U (2018). An overview of the Black Sea pollution in Turkey. MedFAR., 1(2): 67-86.

- Bebianno M J, Géret F, Hoarau P, Serafim M A, Coelho M R, Gnassiabarelli M & Roméo M (2004). Biomarkers in Ruditapes decussatus: a potential bioindicator species. Biomarkers 9(4–5):305–330.
- Bernhard M (1976). Manual of Methods in the Aquatic Environment Research. FAO Fisheries Technical Paper FIRI/T no.158, Food and Agriculture Organisation, Rome.
- European Commission Regulation (2006). Setting maximum levels for certain contaminants in foodstuffs. No 1881/2006 of 19 December 2006.
- Kotze P H, Preez H H & Van Vuren J H J (1999) Bioaccumulation of copper and zinc in Oreochromis mossambicus and Clarias garicpinus, from the Olifants River, Mpumalanga, South Africa. Water SA 25: 99-110.
- TUIK (2019). Turkish Fishery Statistics. http://www.tuik.gov.tr/
- Turkish Food Codex (2002). Official Gazette of Republic of Turkey. Notifications about determination of the maximum levels for certain contaminants in foodstuffs of Turkish Food Codex. (Notification No: 2002/63), 2002; Issue: 24885.
- UNEP (1984). Determination of total Cd, Zn, Pb and Cu in selected marine organisms by flameless AAS. Reference Methods for Marine Pollution Studies, 11 Rev 1.
- UNEP (1985). GESAMP Cadmium, lead and tin the Marine Environment. UNEP Regional Seas Reports and Studies, No 56.
- U.S. EPA (2011). USEPA (United States Environmental Protection Agency) Regional Screening Level (RSL) Summary Table. http://www.epa.gov/regshwmd/risk/human/Index.htm
- U.S. EPA (2016). United States Environmental Protection Agency Regional Screening Levels (RSLs). https://www.epa.gov/risk/regional-screening-levels-rsls-generic-tables-may-2016



# Assessment of Heavy Metal Pollution and Potential Ecological Risk in Sediments of Sinop Shores of the Black Sea

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**Abstract:** Aim of this study was to assess the current heavy metals in sediments of Sinop shores of the Black Sea. Enrichments of the heavy metals were detected on the surface sediments of Sinop shores in the Black Sea. For this purposes, Hg, Cd, Pb, Cu, Zn and Fe levels in surface sediments were studied from five stations in Sinop shores of the Black Sea. The concentrations of metals in the surface sediments of Sinop shores varied from 0.028 to 0.065 mg kg-1 dry wt. for Cd; 0.015 to 0.061 mg kg-1 dry wt. for Hg; 4.91 to 8.03 mg kg-1 dry wt. for Pb; 5.85 to 13.49 mg kg-1 dry wt. for Cu; 13.72 to 33.25 for Zn; and 11340 to 18800 mg/kg for Fe mg kg-1 dry wt. Heavy metal levels were compared to the Sediment Quality Criteria for surface sediments. The levels of the metals were much lower than the levels recommended the sediment quality guideline values (SQGV).

Ranges of metal enrichment factors for metals in the sediments of Sinop shores are as follows: 0.69 to 1.54 for Cd, 0.44 to 2.21 for Hg, 0.69 to 0.91 for Pb, 0.98 to 1.74 for Cu, and 0.64 to 1.05 for Zn. Mostly the EF values for heavy metals were less than 1, may be come from crustal materials or natural weathering processes, indicate no enrichment. However, some the results of EF values were between 1 and 2.21 indicate minor enrichment. These values have not been considered significant. The  $I_{geo}$  values also suggest that surface sediments in Sinop shores of the Black Sea is unpolluted.

Keywords: Sinop, heavy metal, Sediment Quality Guidelines, Enrichment Factor, Geo-accumulation Index, Pollution Load Index

# **1. INTRODUCTION**

The growth in coastal areas of the Black Sea has increased the anthropogenic effects on these systems due to increased amounts of land-based nutrients and other contaminants such as heavy metals (Bat et al., 2018). Heavy metals continue to be reach into coastal ecosystems in the Black Sea through major rivers, runoff and land-based sources (Bat et al., 2009). This has resulted to accumulation of contaminants especially heavy metals in the coastal water. When heavy metals enter into the marine coasts eventually settle and become incorporated into sediments and can affect the health of the marine ecosystem. These sediments bound metals can also be released into the water again under variable conditions. Thus, the metal concentrations in surface sediments can reflect the pollution level and geochemical behaviour of the metals. Therefore, heavy metal contamination is one of the important environmental problems in the Black Sea coasts.

In this study, the amounts of heavy metals in surface sediments of Sinop shores of the Black Sea. The aims of the study were: (1) to determine amounts of heavy metals (Cd, Hg, Pb, Cu, Zn and Fe); (2) to assess the degree of pollution with these metals; (3) to compare heavy metal concentrations with Sediment Quality Guidelines (SQGs).

# 2. MATERIAL AND METHODS

Field sampling was carried out in between June and July of the year 2017. Five sampling stations in Sinop shores were selected to sample surface sediments (< 5 cm) which were sampled using core sampler with 10 cm in length with a diameter of 4 cm. The water depth was less than 20 meters for all stations. The sampling stations are shown in Figure 1. The samples were placed into sealed polyethylene bags, carried to the laboratory in an ice box and stored at  $-21^{\circ}$ C till metal analysis.

Metal analysis (except Hg) in all subsampling pulverized to 85% passing 200 mesh was performed using 4 acid digestion and ultra-trace ICP-MS method by accredited ACME Analytical Laboratories Ltd. (Vancouver, Canada). Ultra-trace

Aqua Regia digestion method for Hg analysis was also used by accredited ACME Analytical Laboratories. The average values of duplicates were analysed for each determination.



#### Figure 1. Study area.

#### **Enrichment Factor (EF)**

In this study, Fe was used as reference element to differentiate natural from anthropogenic components, EF was calculated according to the following equation:

$$EF = \frac{\left(\frac{Me}{Fe}\right)_{sample}}{\left(\frac{Me}{Fe}\right)_{background}}$$

(Me/Fe) <sub>sample</sub> is the ratio of metal and Fe concentrations of the sample and (Me/Fe) <sub>background</sub> is the ratio of background metal and Fe concentrations. Also, the average crust concentrations of elements (Wedepohl, 1995) were taken as background.

#### Geo-accumulation Index (Igeo)

The data set has been subjected to correlation and factor analysis for elucidating the relationships between the heavy metals and geochemical characteristics. The geo-accumulation index is calculated to determine metals contamination in sediments of Sinop shores as the following equation:

$$I_{geo} = log_2 \left[ \frac{C_{sample}}{1.5 x C_{background}} \right]$$

In which, C sample is the measured concentration of samples; C background is the background concentration of sediments and factor 1.5 is the possible variations of background data due to anthropogenic impacts.

#### **Pollution Load Index (PLI)**

Pollution load index, defined as the n<sup>th</sup> root of the multiplicity of the presumed contaminants, is used to determine the quality of the sediment samples. The PLI shows how polluted the sediment is an indication of the overall toxicity of the sediment (Tomlinson et al., 1980). PLI is calculated according to the following equation:

$$PLI = \sqrt[n]{CF_1 \ x \ CF_2 \ x \ \dots \ x CF_n}$$

Where CF is the ratio between the mean concentrations of each metals determined and their background concentrations in the sediment. n= number of CF values. In this study, n, which represents for number of metal, is five.

Pollution Load Index for zone is calculated as the following equation:

 $PLI = \sqrt[n]{Station_I \ x \ Station_{II} \ x \ \dots \ x \ Station_n}$ 

Contamination factors for Cd, Hg, Pb, Cu and Zn are used to produce 5 stations and zone indices, and these indices are then multiplied and five rooted to give the Pollution Load Index for Sinop shores.

# 3. RESULTS AND DISCUSSION

## Results

The concentrations of metals in the surface sediments of Sinop shores varied from 0.028 to 0.065 mg kg<sup>-1</sup> dry wt. for Cd; 0.015 to 0.061 mg kg<sup>-1</sup> dry wt. for Hg; 4.91 to 8.03 mg kg<sup>-1</sup> dry wt. for Pb; 5.85 to 13.49 mg kg<sup>-1</sup> dry wt. for Cu; 13.72 to 33.25 for Zn; and 11340 to 18800 mg kg<sup>-1</sup> dry wt. for Fe (Figure 2).



Figure 2. Mean concentrations of heavy metals in sediments of Sinop shores in the Black Sea

In Figure 3 EFs for the sediments taken from all the stations between June and July of the year 2017 are presented. Ranges of metal enrichment factors for metals in the sediments of Sinop shores are as follows: 0.69 to 1.54 for Cd, 0.44 to 2.21 for Hg, 0.69 to 0.91 for Pb, 0.98 to 1.74 for Cu, and 0.64 to 1.05 for Zn.



Figure 3. Mean values of enrichment factor for 5 elements in the sediments of Sinop shores in the Black Sea

In the present study Igeo was calculated using geochemical background values for all stations of Sinop shores and given in Figure 4.



Figure 4. Mean values of geo-accumulation index for 5 elements in the sediments of Sinop shores in the Black Sea

In this study PLI values were given in Figure 5.



Figure 5. Pollution load index values for 5 elements in the sediments of Sinop shores in the Black Sea

## Discussion

Sediment quality guideline values (SQGV) are very important and useful tools to protect and evaluate marine ecosystem from adverse effects. The results compared to those determined by the recommended SQGV (Simpson and Batley, 2016). In this study, the results were compared with SQGV to see data on the metal levels in sediments of Sinop shores of the

Black Sea. The levels of the metals were much lower than the amounts pointed out the sediment quality. SQGV and SQGV-high for Hg, Cd, Pb, Cu and Zn are 0.15-1.0, 1.5-10, 50-220, 65-270 and 200-410 mg kg-1 dry wt., respectively (Simpson and Batley, 2016). EF values were interpreted as suggested by Birch (2003) where EF<1 indicates no enrichment;  $1 \le EF \le 3$  is minor;  $3 \le EF \le 5$  is moderate;  $5 \le EF \le 10$  is moderately severe;  $10 \le EF \le 25$  is severe;  $25 \le EF \le 50$  is very severe and EF > 50 is extremely severe.

The normalization of the metal concentrations to Fe or Al has become an accepted practice in environmental science for evaluating the metal levels in natural waters due to anthropogenic activities. Fe is a common element in the structure of clay minerals and is also associated with particle surfaces such as oxide coatings. Fe and Al are often used to calculate metal enrichment factors. Fe in estuarine sediments primarily comes from natural weathering processes. The enrichment factor (EF) can be used to determine whether the metals are from the natural weathering processes of rocks or whether metals are from anthropogenic sources and reflect the status of environmental contamination. The assessment criteria are generally based on the EF values. In this study EF values were between 0.44 and 2.21. In this study, mostly the EF values for heavy metals were less than 1, may be come from crustal materials or natural weathering processes, indicate no enrichment. However, some the results of EF values were between 1 and 2.21 which indicate minor enrichment.

Müller (1969) has classified  $I_{geo}$  in relation to contamination levels into following classes; class 0, uncontaminated ( $I_{geo} \le 0$ ), class 1 from uncontaminated to moderately contaminated ( $0 < I_{geo} < 1$ ), class 1 moderately contaminated ( $1 < I_{geo} < 2$ ), class 3 from moderately to strongly contaminated ( $2 < I_{geo} < 3$ ), class 4 strongly contaminated ( $3 < I_{geo} < 4$ ), class 5 from strongly to extremely contaminated ( $4 < I_{geo} < 5$ ) and class 6, extremely contaminated ( $I_{geo} > 5$ ). In the present study Igeo was calculated using geochemical background values for all stations of Sinop shores (see Figure 4). I<sub>geo</sub> was calculated using strong values. I<sub>geo</sub> values for Hg, Cd, Pb, Cu and Zn were less than 0 at all sites, suggesting that the Sinop shores is not polluted by these metals.

PLI was defined by Tomlinson et al. (1980) for estuarine sediments in Ireland based on baseline metal amounts. In this study, Upper Continental Crust values were used (Wedepohl, 1995) as background value for those metals. The PLI is proposed as a standardized system for detecting pollution which permits a comparison of pollution levels between different stations. The Tomlinson pollution index indicates how much a sample exceeds the levels of heavy metals for natural environments and gives an assessment of the overall toxicity status for a sample. On the basis of Tomlinson et al. (1980), PLI value of zero suggests absence of background metals, a value of one suggests that only background levels of metals are present, and value larger than one would suggests progressive deterioration of quality. PLI values ranged between 0.366 (Station III) and 0.522 (Station I), indicating that Sinop shores were less impacted by these heavy metals. PLI value for zone was found as 0.442.

# 4. CONCLUSION

The concentration range of Fe in the surface sediment was found to be the highest at 18800 ppm among the studied heavy metals followed by Zn, Cu, Pb, Cd and Hg. In this study, the accumulated heavy metal levels in sediment from Sinop shores did not exceed the standard maximum limits given by Simpson and Batley (2016). The EF may reflect the real impact of human activities on the environment. In this study the EF was calculated. EF could be a useful criterion in evaluating the impact of the anthropogenic origins of heavy metals. In this study, EF values less or equal to 1, show that there is no contamination at all, the concentration is within the prediction range for the natural background. However, EF values from 1 to 3 show a minor degree of pollution. The I<sub>geo</sub> analyses showed that Sinop shores sediments Hg, Cd, Pb, Cu and Zn levels at all sites were classed as unpolluted; however, PLI values indicated that Sinop shores were less impacted by these heavy metals. The PLI values cannot provide data on the effect of the combination of metals, it can provide the public with same understanding of the quality of a component of their environment, and it can indicate trends over time and region (Tomlinson et al. 1980).

In this study, heavy metals were not significantly enriched in the surface sediments of Sinop shores and did not show a serious harm to the ecosystem. The results of this study may be employed by basis for the government agencies in forming essential regulations to control the transport and disposal of toxic and risky substances. In addition results can be employed to strengthen and implement available public policies in protecting the marine coastal environment.

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#### REFERENCES

Bat L, Gökkurt O, Sezgin M, Üstün F & Sahin F (2009). Evaluation of the Black Sea land based sources of pollution the coastal region of Turkey. The Open Marine Biology Journal 3: 112-124.

Bat L, Öztekin A, Şahin F, Arıcı E & Özsandıkçı U (2018). An overview of the Black Sea pollution in Turkey. MedFAR 1(2): 67-86.

Birch G (2003). A scheme for assessing human impacts on coastal aquatic environments using sediments. in: Woodcoffe, C.D., Furness, R.A. (Eds.), Coastal GIS 2003. Wollongong University Papers in Center for Maritime Policy, 14, Australia.

Müller G (1969). Index of geoaccumulation in the sediments of the Rhine River. Geojournal 2: 108-118.

- Simpson SL, Batley GE (2016). Sediment quality assessment: a practical guide, Second Edition, CSIRO Publishing Australia.
- Tomlinson DL, Wilson JG, Harris CR & Jeffrey DW (1980). Problems in the assessment of heavy metal levels in estuaries and the formation of a pollution Helgoläende Meeresuntersuchungen 33:566–575.

Wedepohl KH (1995). The composition of the continental crust. Geochimica et Cosmochimica Acta 59(7):1217-1232.



# The Use of some Medicinal and Aromatic Plants in Aquaculture

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**Abstract:** Fisheries are facing a number of problems, especially recently and due to intensive farming practices the infectious diseases pose a major problem in aquaculture industry, causing heavy loss to farmers. The use of medicinal and aromatic plants as an alternative to the drugs, chemicals and antibiotics currently being used to control fish diseases and improving the production in fish culture and it is attracting the attention of many researchers. In this context, the medicinal and aromatic plants have been used in many ways for several purposes in aquaculture. The aim of this paper is to review several research that conducted on the use of medicinal and aromatic plants as immunostimulants, antibacterial, antiviral, antifungals and anti-stress.

Keywords: Aquaculture, medicinal and aromatic plants, immunostimulants, plant extracts.

## **1. INTRODUCTION**

The aquaculture industry is one of the fastest growing food-producing sectors and playing an important role for food procurement (FAO 2016). It is expected that world aquaculture trade will be reached up to 9.7 billion by 2050. The value of aquaculture production is increasing year by year but the infectious diseases seem the main problem that restrict the production rate. The fish farmers are generally using antibiotics and chemicals for curing but environmental impact and antibiotic resistance appeared as a main problem (Harikrishnan et al., 2010). Antibiotics are also have many side effects (Atal, 1982).

Immunostimulants seem focus point in aquaculture industry with its protection, and growth promoting properties. Especially medicinal plants are capable of improving defense mechanism against infectious diseases (Sahu et al., 2006; Sahu, 2004, Rao et al, 2006). In some conditions, immunostimulants also used as a therapeutic (Bilen and Elbeshti 2019; Bilen et al 2019a; Bilen et al 2014). Medicinal plants play an important role as an immunostimulantwith its alkaloids, flavanoids, pigments, phenolics, terpenoids, steroids and essential oils (Citarasu et al., 2002; Sivaram et al. 2004; Dulger et al 2005).

Most effective way of losing fish is to improve and strengthen of the immune system (Robertsen, 1999; Raa et al., 1992; Bilen et al 2018). So that in the present review we try to explain the use medicinal and aromatic plants and their compounds and extracts in the health aspects of aquaculture especially as immunostimulants, antibacterial antiviral, antifungals and anti-stress.

#### Use of Medicinal and Aromatic Plants as Immunostimulants

Several studies have been conducted on the use of medical and aromatic plants as immunostimulant of fish immune system. It is well known that fish treated with immunostimulants show increased phagocytosis as well as respiratory burst activity (Bilen et al 2019b; Amhamed et al 2018; Mohamed et al 2018; Almabrok et al 2018; Bilen et 1 2019c ).

Some of the medicinal plants such as *Emblica officinalis*, *Cynodon dactylon* and *Adathoda vasica* showed an increased immune response in *Poecilia sphenops* (Magdelin 2005) and goldfish *Carassius auratus* (Minomol, 2005). Increased immune activity was also observed in *Carassius auratus* fed with nettle, in jian carp (*Cyprinus carpio var. Jian*) fed with *Astragalus* root (Jian and Wu, 2003) and in yellowcroaker (*Pseudosciaena crocea*) *Angelica* root (*Radix Angelicae sinensis*). Also, thyme, rosemary and fenugreek showed immunostimulant effects (Yilmaz et al. 2011). Some different herbal mixtures used as an immunostimulant in aquaculture (Harikrishnan et al. 2011).

Some of the ingredients of the medicinal plants are well know that eugenol, methyl eugenol, and caryophyllene (Chopra et al., 1956), saponin (Ninomiya et al., 1995), glycyrrhizin (Jang et al., 1995), aloe (Kim et al., 1999), azadirachtin (Logambal and Michael, 2000; Logambal and Michael, 2001; Harikrishnan et al., 2009a; Harikrishnan et al., 2009b) and glycyrrhizin (Wada et al. 1987; Zhang et al. 1990). *Quillaja saponin* in yellowtail (Ninomiya et al., 1995), *Astragalus polysaccharides* also showed immunostimulant effects (Jian and Wu, 2003; Tzianabos, 2000). Immunostimulant effects of the some medicinal plants were demonstrated by many different scientist (Gopalakannan and Arul, 2006; Venkatalakshmi and Michael, 2001; Sharifpour, 1997; Logambal et al., 2000; Logambal and Michael, 2001; Chakrabarti and Rao, 2006; Jang et al., 1995; Rao and Chakrabarti, 2005; Kim et al., 1999). Also, in shrimps (Luo, 1997), in tilapia (Chansue et al., 2000), in *Epinephelus tauvina* when fed with *Ocimum sanctum*, *Withania somnifera*, and *Myristica fragrans methanolic* extracts (Sivaram et al., 2004), in rainbow trout fed with *Viscum album*, *Urtica dioica*, and *Zingiber officinale* (Dügenci et al., 2003), in *Labeo rohita* fed with *Catharanthus roseus* plant extract (Nguyen et al., 2002) shwed an increased non-specific immune responses.

Medicinal plants extracts have been used in aquaculture to enhance non-specific immunity. For instance, acetone extract of *Ocimum sanctum* (Hemapriya, 1997), water extract of *Ocimum sanctum* (Logambal et al., 2000), acetone extract of *Phyllanthus emblica* (Balasubramani and Michael 2002) showed different immune responses.

Medicinal palnts also used șn vaccineation of the fish as adjuvant (Jeney and Anderson, 1993a; Jeney and Anderson, 1993b). administration of medicinal plants as an immunostimulant in finfish akso showed protection agains *Aeromonas salmonicida* (Ninomiya et al., 1995), *Aeromonas hydrophila* (Logambal et al., 2000; Bilen et al 2019).

## Use of Medicinal and Aromatic Plants as Antibacterial

The medicinal plants also take into consideration with their antibacterial usage in aquaculture (Shangliang et al., 1990). *Stella aquatica, Ocimum sanctum, Rosmarinus officinalis, Psidium guajava*, (Shangliang et al., 1990; Logambal et al., 2000; Abutbul et al., 2004). Direkbusarakom (2004) showed oxytetracycline could be used more effectively used with guava.

#### Use of Medicinal and Aromatic Plants as Antivirals

A few herbs have been determined to have antiviral effects against fish viruses in vivo (Direkbusarakom et al. 1996a), and some have been found for their ability on viruses that kill shrimps (Direkbusarakom et al., 1995). Some of Indian medicinal plants such as *Aegle marmelos*, *Cynodon dactylon*, *Lantana camara*, *Momordica charantia* and *Phyllanthus amarus* showed strong antiviral activity against WSSV. *Cynodon dactylon* extracts is very effective against WSSV (Balasubramanian et al., 2007). *Lonicera japonica* extract showed inhibitory effects against IPN and IHN. Against IHN *Artemisia vulgaris* and *Stellaria aquatic* extractalso found effective (Shagnliang et al., 1990). Yellow head virusin shirpm as found sensitive Ethanol of *Clinacanthus nutans* (Direkbusarakom et al., 1996b). Salmonid rhabdovirus and viral haemorrhagic septicaemia virus (VHSV) could be cured with *Olea europaea* leaf (Micol et al., 2005). Also Salmonid rhabdo virus and viral haemorrhagic septicaemia virus of five different herbal medicinal plants like *Cynodon dactylon*, *Aegle marmelos*, *Tinospora cordifolia*, *Picrorhiza kurooa* and *Eucalyptus alba* were selected and prepared as a diet for WSSV-infected shrimps. Yogeeswaran (2007), demonstrated that *Aclypha indica*, *Cynodon dactylon*, *Picrorhiza kurooa*, *Withania somnifera* and *Zingiber officinalis* methanolic extracts are effective against WSSV

#### Use of Medicinal and Aromatic Plants as Antifungals

The main antifungal structure of the medicinal plants is methylethyl pentanoate (Dabur 2004) has showed anti-Aspergillus properties and against 10 clinical isolates of Candida, 19 clinical isolates of Aspergillus and a few marine fungi (Citarasu, 2010). *Tamarix dioica, Aspergillus flavi* and *Rhazya stricta* determined as antifungal against *Trichophyton longifusis, Aspergillus flavus, Candida albicans* and *Fusarium solani* (Khan et al., 2004). *Terminalia catappa*, extract can reduce the fungal infection in tilapia eggs (Chitmanat et al., 2005).

Diethanol-amide extracted from coconut, *Azadirachta siamensis* and *Melaleuca alternifloria* showed fungicidal activity against hyphae of the fish-pathogenic *Oomycete fungus*, *Aphanomyces invadans*.

### Use of Medicinal and Aromatic Plants as Anti-stress

The medicinal plant mixture (HM), *Massa medicata fermentata*, *Crataegi fructus*, *Artemisia capillaries*, and *Cnidium officinale*, is useful to improve growth, fatty acid utilization, and stress recovery in the Japanese flounder (Ji et al., 2007).

Herbal compounds have the ability to inhibit the generation of oxygen anions and to scavenge free radicals. The herbal antioxidant effect has been shown to be similar to that of superoxide dismutase, metal-ion chelators and xanthine oxidase inhibitors. The best example is the herb *Picrorhiza kurroa* used as an antistress compound for shrimps. The products Stresstol (*Withania somnifera, Ocimum sanctum*, purified Silajit, *Tinospora cordifolia, Picrorhiza kurvoa, Eclipta erecta, Ipomea digitata, Vernonia cinera*) and *Tefroli (Tephrosia purpurea, Eclipta alba, Phyllanthus niruri, Andrographis paniculata, Ocimum sanctum* and *Terminalia chebula*) had a good influence on shrimp post-larvae. The postlarvae fed with the above products were resistant to osmotic pressure, temperature and pH stress.

Some medicinal plant extracts have been used in aquaculture as an anti-stress, such as herbal extracts, *Andrographis paniculata, Portulaca oleracea* and *Flavescent sophora* (Wu et al. 2007). Rutin is a bioflavonoid extracted from *Toona sinensis* with strong antioxidant and antistress activity in crustaceans. Rutin has improved the biochemical, immunological and haematological parameters in *Litopenaeus vannamei* during the stress conditions by *Vibrio alginolyticus* (Hsieh et al. 2008).

#### 2. CONCLUSION

The application of antibiotics and chemicals in culture is often expensive and undesirable since it leads to antibiotic and chemical resistance and consumer reluctance (Harikrishnan et al., 2011). The use of medicinal and aromatic plants as an alternative to the drugs, chemicals and antibiotics currently being used to control fish diseases in aquaculture is attracting the attention of many researchers. In this context, many have focused on the use of medicinal plants and its products as potential therapeutic measures for improving and protect the aquaculture industry. Medicinal plants and their products are cheaper source for therapeutics, have greater accuracy than chemotherapeutic agents, and offer a viable solution for most problems which aquaculture faces today. Several medicinal and aromatic plants and their compounds have been successfully in aquaculture as immunostimulants, antibacterial, antiviral, antifungals and anti-stress.

#### REFERENCES

- Almabrok, A. A., Amhamed, I. D., Mohamed, G. A., Bilen, S., & Altief, T. A. S., 2018. Effect of *Tilia tomentosa* methanolic extract on growth performance, digestive enzyme activity, immune system and haematological indices of common carp (*Cyprinus carpio*). Marine Science and Technology Bulletin, 7(1), 12-20.
- Abutbul, S., Golan-Goldhirsh, A., Brazani, O., Zilberg, D., 2004. Use of *Rosmarinus officinalis* as a treatment against *Streptococcus iniae* in tilapia (*Oreochromis* sp.). Aquaculture 238, 97–105.
- Amhamed, I. D., Mohamed, G. A., Almabrok, A. A., Altief, T. A. S., & Bilen, S., 2018. Efficacy of Dietary *Chenopodium album* Extract on Some Health Parameters, Digestive Enzymes and Growth Performance in Juvenile *Cyprinus carpio*. Alinteri Zirai Bilimler Dergisi, 33(2), 165-176.
- Atal, C.K., 1982. Chemistry of some biological active Indian medicinal plants. Proc Indian Natl Sci Acad 48(Suppl 1):99–121.
- Balasubramanian, G., Sarathi, M., Rajesh Kumar, S., Sahul Hameed, A.S., 2007. Screening the antiviral activity of Indian medicinal plants against white spot syndrome virus in shrimp. Aquaculture 263:15–19. doi: 10.1016/j.aquaculture.2006.09.037
- Balasubramani, S.P., Michael, R.D., 2002. Immunomodulation by the fruit extract of Indian Gooseberry, *Phyllanthus emblica* (Linn). in *Oreochromis mossambicus* (Peters). MSc thesis. The American College, Madurai.
- Bilen, S., Elbeshti, H. T. A. G. 2019. A new potential therapeutic remedy against *Aeromonas hydrophila* infection in rainbow trout (*Oncorhynchus mykiss*) using tetra, *Cotinus coggygria*. Journal of Fish Diseases.
- Bilen, S., Sirtiyah, A. M. A., & Terzi, E. 2019a. Therapeutic effects of beard lichen, *Usnea barbata* extract *against Lactococcus garvieae* infection in rainbow trout (*Oncorhynchus mykiss*). Fish & Shellfish immunology, 87, 401-409.
- Bilen, S., Kenanoglu, O. N., Terzi, E., Ozdemir, R. C., & Sonmez, A. Y. 2019b. Effects of tetra (*Cotinus coggygria*) and common mallow (*Malva sylvestris*) plant extracts on growth performance and immune response in Gilthead Sea bream (*Sparus aurata*) and European Sea bass (*Dicentrarchus labrax*). Aquaculture, 734251.
- Bilen, S., Soydaş, E., & Bilen, A. M. (2014). Effects of methanolic extracts of nettle (*Urtica dioica*) on non-specific immune response of gold fish (*Carassius auratus*). Alinteri Zirai Bilimler Dergisi, 27, 24-29.
- Bilen, S., Filogh, A. M., Ali, A. B., Kenanoğlu, O. N., & Zoral, M. A. (2019). Effect of common mallow (*Malva sylvestris*) dietary supplementation on growth performance, digestive enzyme activities, haemotological and immune responses of common carp (*Cyprinus carpio*). Aquaculture International, 1-12.
- Bilen, S., Özkan, O., Alagöz, K., & Özdemir, K. Y. (2018). Effect Of dill (*Anethum graveolens*) and garden cress (*Lepidium sativum*) dietary supplementation on growth performance, digestive enzyme activities and immune responses of juvenile common carp (*Cyprinus carpio*). Aquaculture, 495, 611-616.
- Chakrabarti, R., Rao, Y.V., 2006. Achyranthes aspera stimulates the immunity and enhances the antigen clearance in Catla catla. Int. Immunopharmacol. 6, 782–790.
- Chansue, N., Ponpornpisit, A., Endo, M., Sakai, M., Satoshi, Y., 2000. Improved immunity of tilapia *Oreochromis niloticus* by C-UP III, a herb medicine. Fish Pathol. 35, 9–90.
- Chitmanat, C., Tongdonmuan, K., Khanom, P., Pachontis, P., Nunsong, W., 2005. Antiparasitic, antibacterial, and antifungal activities derived from a *terminalia catappa* solution against some tilapia (*Oreochromis niloticus*) pathogens. Acta Hortic 678:179–182 ISHS.
- Chopra, R.N., Nayyar, S.L., Chopra, I.C., 1956. Glossary of Indian Medicinal Plants. Council of Scientific and Industrial Research, New Delhi, India, p. 160.
- Citarasu, T., 2010. Herbal biomedicines: a new opportunity for aquaculture industry. Aquacult Int 18, 403-414.
- Citarasu, T., Sekar, R.R., Babu, M.M., Marian, M.P., 2002. Developing Artemia enriched herbal diet for producing quality larvae in Penaeus monodon. Asian Fish Sci 15:21–32.
- Dabur, R., 2004. A novel antifungal pyrrole derivative from Datura metel leaves. Pharmazie 59:568–570.
- Direkbusarakom, S., Herunsalee, A., Boonyaratpalin, S., Danayadol, Y., Aekpanithanpong, U., 1995. Effect of *Phyllanthus* spp. against yellow-head Baculovirus infection in Black Tiger Shrimp, *Penaeus monodon*. In: Shariff M, Arthur JR, Subasinghe RP (eds) Diseases in Asian aquaculture II. Fish Health Section, Asian Fisheries Society, Manila, pp 85–92.
- Direkbusarakom, S., Herunsalee, A., Yoshimizu, M., Ezura, Y., 1996a. Protective efficacy of *Clinacanthus nutans* on yellow-head disease in Black Tiger Shrimp (*Penaeus monodon*). Fish Pathol 33(4):404–410.
- Direkbusarakom, S., Runghomnerdwong, S., Herunasalee, A., Rungpan, L., 1996b. Screening of Thai traditional herbs against shrimp pathogenic bacteria. NICA Technical paper, No. 7, p. 13.
- Direkbusarakom, S., 2004. Application of medicinal herbs to aquaculture in Asia, Walailak. J Sci Tech1(1):7-14.
- Dügenci, S.K., Arda, N., Candan, A., 2003a. Some medicinal plants as immunostimulant for fish. J. Ethnopharmacol. 88, 99–106.
- Dulger, B., Gonuz, A., Bilen, S., & Jäger, A. K. 2005. Antimicrobial studies on three Hypericum species from Turkey. South African Journal of Botany, 71(1), 100-103.
- FAO, 2016. The State of World Fisheries and Aquaculture. Contributing to food security and nutrition for all. Rome. 200 pp.
- Gopalakannan, A., Arul, V., 2006. Immunomodulatory effects of dietary intake of chitin, chitosan and levamisole on the immune system of *Cyprinus carpio* and control of *Aeromonas hydrophila* infection in ponds. Aquaculture 255, 179–187.
- Harikrishnan, R., Balasundaram, C., Dharaneedharan, S., Moon, Y.G., Kim, M.C., Kim, J.S., Heo, M.S., 2009a. Effect of plant active compounds on immune response and disease resistance in *Cirrhina mrigala* infected with fungal fish pathogen, *Aphanomyces invadans*. Aquacult. Res. 40, 1170–1181.
- Harikrishnan, R., Balasundaram, C., Kim, M.C., Kim, J.S., Han, Y.J., Heo, M.S., 2009b. Innate immune response and disease resistance in *Carassius auratus* by triherbal solvent extracts. Fish Shellfish Immunol. 27, 508–515.
- Harikrishnan, R., Balasundaram, C., Heo, M., 2011. Impact of plant products on innate and adaptive immune system of cultured finfish and shellfish. Aquaculture 317, 1–15.
- Harikrishnan, R., He, J., Balasundaramb, C., Kim, M.C., Kim, J.C., Han, Y.J., M.S. Heo, M.S., 2010. Effect of *Punica granatum* solvent extracts on immune system and disease resistance in *Paralichthys olivaceus* against lymphocystis disease virus (LDV), Fish and Shellfish Immunology, 29, 668-673.
- Hemapriya, V.S., 1997. Immunostimulatory effect of leaf extracts of few medicinal plants in *Oreochromis mossambicus* (Peters), MSc thesis. The American College, Madurai.
- Hsieh, T.J., Chyi Wang, J., Hu, C.Y., Li, C.T., Ching-Ming, K., Hsieh, S.L., 2008. Effects of Rutin from *Toona sinensis* on the immune and physiological responses of white shrimp (*Litopenaeus vannamei*) under *Vibrio alginolyticus* challenge. Fish Shellfish Immunol 25(5):581–588. doi:10.1016/j.fsi.2008.07.014
- Jang, S.I., Marsden, M.J., Kim, Y.G., Choi, M.S., Secombes, C.J., 1995. The effect of glycyrrhizin on rainbow trout, Oncorhynchus mykiss (Walbaun), leucocyte responses. J. Fish Dis. 18, 307–315.
- Jeney, G., Anderson, D.P., 1993a. Enhanced immune response and protection in rainbow trout to *Aeromonas salmonicida* bacterin following prior immersion in immunostimulants. Fish Shellfish Immunol. 3, 51–58.
- Jeney, G., Anderson, D.P., 1993b. Glucan injection or bath exposure given alone or in combination with a bacterin enhance the nonspecific defence mechanisms in rainbow trout (*Oncorhyncus mykiss*). Aquaculture 116, 315–329.
- Jian, J., Wu, Z., 2003. Effects of traditional Chinese medicine on nonspecific immunity and disease resistance of large yellow croaker, *Pseudosciaena crocea* (Richardson). Aquaculture 218,1–9.
- Ji, S.C., Jeong, G.S., Im, G.S., Lee, S.W., Yoo, J.H., Takii, K., 2007. Dietary medicinal herbs improve growth performance, fatty acid utilization, and stress recovery of Japanese flounder. FISHERIES SCIENCE 73, 70–76.
- Khan, S., Khan, G.S., Mehsud, S., Rahman, A., Khan, F., 2004. Antifungal activity of *Tamarix dioica*—an in vitro study. Gomal J Med Sci 2:2.

- Kim, K.H., Hwang, Y.J., Bai, S.C., 1999. Resistance to Vibrio alginolyticus in juvenile rockfish (*Sebastes schlegeli*) fed diets containing different doses of aloe. Aquaculture 180, 13–21.
- Logambal, S.M., Michael, R.D., 2000. Immunostimulatory effect of Azadirachtin in *Oreochromis mossambicus* (Peters). Indian J. Exp. Biol. 38, 1092–1096.
- Logambal, S.M., Michael, R.D., 2001. Azadirachtin an immunostimulant for *Oreochromis mossambicus* (Peters). J. Aquac. Trop. 16, 339–347.
- Logambal, S.M., Venkatalakshmi, S., Michael, R.D., 2000. Immunostimulatory effect of leaf extract of *Ocimum sanctum* Linn. in *Oreochromis mossambicus* (Peters). Hydrobiologia 430, 113–120.
- Luo, R., 1997. Induction of immunity substance in *Penaeus Chinensis* by Chinese herbal medicine. Oceanlogia et Limnologia Sinica 28, 573–577 (in Chinese).
- Magdelin, S.M., 2005. Culture of ornamental fish, Black molly (*Poecilia sphenops*) using medicinal plants having immunostimulant characteristics. M.Phil Dissertation, Manonmaiam Sundaranar University, India.
- Micol, V., Caturla, N., Perez-Fons, L., Mas, V., Perez, L., Estepa, A., 2005. The olive leaf extract exhibits antiviral activity against viral haemorrhagic septicaemia rhabdovirus (VHSV). Antiviral Res. 66, 129–136.
- Minomol, M., 2005. Culture of Gold fish *Carassius auratus* using medicinal plants having immunostimulant characteristics. M.Phil Dissertation, MS University, India.
- Mohamed, G. A., Amhamed, I. D., Almabrok, A. A., Barka, A. B. A., Bilen, S., & Elbeshti, R. T., 2018. Effect of celery (*Apium graveolens*) extract on the growth, haematology, immune response and digestive enzyme activity of common carp (*Cyprinus carpio*). Marine Science and Technology Bulletin, 7(2), 51-59.
- Nguyen, T.T.T., Mukherjee, S.C., Pani, P.K., 2002. Studies on the immunostimulatory effect of certain plant extracts on fish. Abstracts: AH-13, The Sixth Indian Fisheries Forum, Mumbai, India, p. 153.
- Ninomiya, M., Hatta, H., Fujiki, M., Kim, M., Yamamoto, T.R., 1995. Enhancement of chemotactic activity of yellowtail (*Seriola quinqueradiata*) leucocytes by oral administration of Quillaja saponin. Fish Shellfish Immunol. 5, 325–327.
- Raa, J., Rorstad, G., Engstad, R.E., Robertson, B., 1992. The use of immunostimulants to increase resistance of aquatic organisms to microbial infections. In: Shariff, M., Subasinghe, R.P., Arthur, J.R. (Eds.), Disease in Asian Aquaculture. Proceedings of the First Symposium on Diseases in Asian Aquaculture. Asian Fisheries Society, Philippines, pp. 39–50. 1.
- Rao, Y.V., Chakrabarti, R., 2005. Stimulation of immunity in Indian major carp *Catla catla* with herbal feed ingredients. Fish Shellfish Immunol. 18, 327–334.
- Rao, Y.V., Das, B.K., Pradhan, J., Chakrabarti, R., 2006. Effect of Achyranthes aspera on the immunity and survival of Labeo rohita infected with Aeromonas hydrophila. Fish and Shellfish Immunology, 20, 263-273.
- Robertsen, B., 1999. Modulation of the non-specific defence of fish by structurally conserved microbial polymers. Fish Shellfish Immunol. 9, 269–290.
- Sahu, S., 2004. Antibacterial activity of plant extracts on fish microbial pathogens, M.F. Sc. Dissertation thesis, CIFA, Kausalyaganga, Bhubaneswar, India. 237.
- Sahu, S., Das, B.K., Mishra, B.K., Pradhan, J., Sarangi, N., 2006. Effect of *Allium satium* on the immunity and survival of *L. rohita* infected with *A. hydrophila*, Journal of Applied Ichthyology, 22, 1-6.
- Shangliang, T., Hetrick, F.M., Roberson, B.S., Baya, A., 1990. The antibacterial and antiviral activity of herbal extracts for fish pathogens. *J Ocean University of Qingdao*; 20: 53-60.
- Sharifpour, I., 1997. Histology of inflammatory response of carp (*Cyprinus carpio L*.) to various stimuli. Ph.D. thesis, Institute of Aquaculture, University of Stirling, Scotland.
- Sivaram, V., Babu, M.M., Citarasu, T., Immanuel, G., Murugadass, S., Marian, M.P., 2004. Growth and immune response of juvenile greasy groupers (*Epinephelus tauvina*) fed with herbal antibacterial active principle supplemented diets against Vibrio harveyi infections. Aquaculture 237:9–20. doi:10.1016/ j.aquaculture.2004.03.014.
- Tzianabos, A.O., 2000. Polysaccharide immunomodulators as therapeutic agents: structural aspects and biologic function. Clin. Microbiol. Rev. 13, 523–533.
- Venkatalakshmi, S., Michael, R.D., 2001. Immunostimulation by leaf extract of Ocimum sanctum Linn. in Oreochromis mossambicus (Peters). J. Aquac. Trop. 16, 1–10.
- Wada, T., Arima, T., Nagashima, H., 1987. Natural killer activity in patients with chronic hepatitis treated with OK432, interferon, adenine arabinoside and glycyrrhizin. Gastroenterol Jpn 22(3):312–321.
- Wu, G., Yuan, C., Shen, M., Tang, J., Gong, Y., Li, D., Sun, F., Huang, C., Han, X., 2007. Immunological and biochemical parameters in carp (*Cyprinus carpio*) after Qompsell feed ingredients for long-term administration. Aquac Res 38(3):246–255. doi:10.1111/j.1365-2109.2007.01660.x
- Yilmaz, S., Ergun,S., Yigit,M., 2011. Effects of thyme, rosemary and fenugreek on some hematological and immunological parameters of tilapia, *Oreochromis mossambicus*, Aquaculture Europe, Conferince Rhodes, Greece, October 18- 21. 311-312.

- Yogeeswaran, A., 2007. Protection of *Penaeus monodon* against white spot syndrome virus by inactivated vaccine and herbal immunostimulants, M.Phil Dissertation submitted to Manonmaniam Sundaranar University, Tirunalveli.
- Zhang, Y.H., Yoshida, T., Isobe, K., Rahman, M.J., Nagase, F., Ding, L., Nakashima, I., 1990Modulation by glycyrrhizin of the cellsurface expression of H-2 class I antigens on murine tumour cell lines and normal cell populations. Immunology, 70(3), 405.



# ORAL PRESENTATION

## Trend Analysis for Annual Streamflow of Araç Stream (Turkey)

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Abstract: The aim of this study is to determine the tendency of mean monthly, seasonally and annual streamflow of Araç Stream (Turkey). Streamflow data obtained from the gauging station were between 1985 and 2007. Change-point analysis was performed to determine the change year of the time series of the data. Mann-Kendall test, Spearman test and trend analysis were applied to detect trends in the streamflow. Results of the change-point analysis for mean annual streamflow pointed out that the change year was 1993. Trend analysis results showed that there were decreasing trends for mean annual, seasonally (all seasons) and monthly (all months except February and March). However, there was no statistically significant trend for streamflow. In conclusion, decreasing trends for the streamflow of Araç Stream have been predicted for monitoring period. These decreasing trends could be linked with decrease in snowmelt and rainfall, in addition to extremely increase in temperature and evaporation patterns of air or water, and other causes by the climate change. Thus, streamflow trends should be monitored to estimate the amount and availability of water resources in the future.

Keywords: Araç Stream, trend analysis, streamflow, climate change, time series

### **1. INTRODUCTION**

Streamflow patterns are important for management and sustainable use of water resources. Knowledge on regime of the streamflow has a key role in determining the function, composition, and structure of river basin (Pumo et al., 2016). Ensuring the availability of surface water is compulsory for providing sufficient water resources to support the ecosystem services. Streamflow regimes rely on varying climatic factors. Climate change is anticipated to have noteworthy consequence on the streamflow regimes globally. In order to investigate the future patterns of the streamflow in rivers, trend analysis has significant contributions especially in the perspective of the climate change.

Great attention has been paid to investigate the trends in hydrologic and climatic parameters (Kadıoğlu, 1997; Büyükyıldız and Berktay, 2004; Cigizoglu et al., 2005; Yıldırım et al., 2013; Saplıoğlu et al., 2014; Sütgibi, 2015; Yenigün and Ülgen, 2016; Ay and Özyıldırım, 2017; Ercan and Yüce, 2017; Kale, 2017a, b; Tosunoğlu, 2017; Tosunoglu and Kisi, 2017). In addition, trends in water parameters were determined by using various approaches (Sen, 1968; Hirsch et al., 1982; Helsel and Hirsch, 2002; Şen, 2012). These approaches were performed to estimate the trends of streamflow in rivers by several authors in Turkey (Kalayci and Kahya, 1998; Albek, 2002; Doğan Demir et al., 2016; Ejder et al., 2016a, b; Kale et al., 2016a, b; Kale et al., 2018; Kişi et al., 2018; Sönmez and Kale, 2018). Unfortunately, there is no investigation on determining the trends of the streamflow in Araç Stream. Thus, this paper aimed to perform a trend analysis for the streamflow of Araç Stream and to determine the trends in the surface runoff in the river for the first time.

## 2. MATERIALS AND METHODS

### Study Area

Araç Stream has its source from Ilgaz Mountain and passes through broad valleys. It combines with Soğanlı Stream near Karabük province and serves as a water supplier resource for agricultural irrigation purposes. The region has a typical continental climate with warm summers, snowy winters, cold and frost spring and autumn seasons (Kale and Sönmez, 2018b).

The streamflow data were acquired from streamflow gauging station (station number: D13A053) of The General Directorate of State Hydraulic Works (DSI) at Kayaboğazı (Figure 1). Statistical analyses were performed by annual, seasonally, and monthly.



Figure 1. Araç Stream and streamflow gauging station

### **Change-Point Analysis**

Pettitt (1979) proposed a method to detect the change point of the time series data. Then, several scholars used this method to detect the change time of the climatic or hydrologic time series. In this study, change-point analysis was applied to the streamflow data by using R statistical software (R Core Team, 2019). Pettitt's change-point test can be expressed as in Equation 1:

$$U_{t,T} = \sum_{i=1}^{t} \sum_{j=t+1}^{T} sgn(x_i - x_j) \text{ for } t = 2, \dots, T, K_T = max |U_{t,T}|,$$
(1)

#### **Trend Analysis**

Trend analysis is a broadly performed method to determine the trend in a time series. In this study, Box and Jenkins (1976) methodology was used to determine the trends in hydrologic time series. The formula is given in Equation 2.

$$X_{t} = c + \Phi_{I} X_{t-1} + \dots + \Phi_{p} X_{t-p} + \theta_{I} e_{t-1} + \theta_{q} e_{t-q} + e_{t}$$
(2)

### Mann-Kendall and Spearman's Rho Tests

Non-parametric Mann-Kendall and Spearman's rho tests were used to determine the correlation between the parameters. Mann-Kendall test (Mann, 1945; Kendall, 1955) is widely used to determine the trends in time series. Calculation is as follow:

$$S = \sum_{i=1}^{n-1} \sum_{k=i+1}^{n} sgn(x_k - x_i)$$
(3)

Spearman's rho test is widely used to measure the magnitude of monotonic relationship between two variables (Lehmann, 1975; Sneyers, 1990). The formula is given in Equation 4:

$$\rho = 1 - \frac{6(\sum d^2)}{n(n^2 - 1)} \tag{4}$$

### 3. RESULTS AND DISCUSSION

### Results

The basic statistics for streamflow dataset including mean, standard deviation (SD), maximum and minimum values, range, coefficient of variation (CV), coefficient of skewness (CS), and coefficient of kurtosis (CK) were listed in Table 1.

| Streamflow | Mean  | Standard Deviation | <b>Coefficient of Variation</b> | Coefficient of Skewness | Max   | Min   |
|------------|-------|--------------------|---------------------------------|-------------------------|-------|-------|
| Annual     | 21.81 | 6.44               | 0.30                            | 0.14                    | 31.41 | 7.05  |
| Spring     | 47.10 | 14.49              | 0.31                            | -0.44                   | 72.83 | 17.63 |
| Summer     | 12.59 | 8.00               | 0.64                            | 0.17                    | 25.84 | 0.56  |
| Autumn     | 7.00  | 4.44               | 0.63                            | 1.11                    | 17.42 | 1.96  |
| Winter     | 20.57 | 9.37               | 0.46                            | 0.46                    | 39.43 | 8.06  |

Table 1. Descriptive statistics of annual and seasonally streamflow data

The results of change-point analyses were given in Table 2. Change-point was found to be 1993 for mean annual streamflow during the monitoring period. In addition, trend analysis results detected a decreasing trend in the mean annual streamflow of Araç Stream for this period (Figure 2).

Table 2. Change-point analysis results of mean annual, seasonally and monthly streamflow

| Streamflow   |           | Change Point |
|--------------|-----------|--------------|
| Annual       |           | 1993         |
|              | Spring    | 1993         |
| Concernally, | Summer    | 1992         |
| seasonally   | Autumn    | 1991         |
|              | Winter    | 1990         |
|              | January   | 1989         |
|              | February  | 1989         |
|              | March     | 2003         |
|              | April     | 2005         |
|              | May       | 1993         |
| Mandhlu      | June      | 1992         |
| монту        | July      | 1992         |
|              | August    | 2000         |
|              | September | 1991         |
|              | October   | 1991         |
|              | November  | 1991         |
|              | December  | 2000         |



Figure 2. Trend analysis result for mean annual streamflow of Araç Stream

Decreasing trends were also determined in the mean seasonal streamflow of Araç Stream for all seasons (Figure 3). Moreover, there were decreasing trends in the mean monthly streamflow for all months excluding February and March (Figure 4).



Figure 3. Trend analysis results for mean seasonally streamflow of Araç Stream

Water resources are affecting by the effects of the climate change and global warming (Hisar et al., 2015). Hence, investigations of trends in water resources make significant contributions for water resource managers and decision-makers (Kale and Sönmez, 2018b). Trend analysis of streamflow in rivers is crucial to evaluation of the past, present and future of available water resources. In addition, it suggests management strategies of water resources for the future periods.



Figure 4. Trend analysis results for mean monthly streamflow of Araç Stream

### Discussion

Numerous scholars have globally investigated trends in water resources (Douglas et al., 2000; Zhang et al., 2001; Birsan et al., 2005; Kundzewicz et al., 2005; Zhang et al., 2006; Salarijazi et al., 2012; Herawati et al., 2015; Yeh et al., 2015; Zhou et al., 2015; Pumo et al., 2016). Similarly, many researchers evaluated the trends in streamflow of rivers in Turkey. Kahya and Kalaycı (2004) reported that decreasing trends were detected for river basins located in the west of Turkey whereas no trend was found for river basins in the east of Turkey. Ozkul (2009) documented decreasing trends for both streamflow of Gediz and Büyük Menderes rivers. Durdu (2010) found a decreasing trend in the streamflow of Büyük Menderes River. Bahadır (2011) described a downward trend in the streamflow of Kızılırmak River. Türkeş and Acar Deniz (2011) detected downward trend in river streamflow of the southern Marmara region. Koçman and Sütgibi (2012) determined decreasing trend in the streamflow of Gediz River basin. Ejder et al. (2016a) reported a downward trend in streamflow of Sariçay Stream. Ejder et al. (2016b) found a decreasing trend for the streamflow of Kocabaş Stream. Kale et al. (2016a) detected a decreasing trend in the streamflow of Karamenderes River while Kale et al. (2016b) determined a decreasing trend in the streamflow of Bakırçay River. Kale et al. (2018) documented that Gediz, Büyük Menderes, and Tuzla Rivers have tendencies to decrease. Moreover, Sönmez and Kale (2018) found a downward trend for the streamflow of Filyos River. Kale and Sönmez (2018b) indicated that streamflow of Daday Stream presented a decreasing trend similar to the streamflow of Akkaya Stream which reported by Kale and Sönmez (2018a). In this paper, although decreasing trends were determined for almost all mean annual, seasonally and monthly, no statistically significant trend was found for the streamflow of Arac Stream. On the other hand, these annual, seasonally and monthly decreasing trends in the streamflow could be affected by several reasons such as climate change, excessive utilization of water resources for domestic or agricultural purposes. Variations in temperature and evaporation could affect the streamflow. In addition, changes in rainfall regimes could also have significant effects on the amounts of the streamflow. Similarly, Kale and Sönmez (2018b) claimed that significant downward trends in annual, seasonally and monthly streamflow of Daday Stream could be associated with the climatic changes particularly temperature and rainfall. Sönmez and Kale (2018) indicated that streamflow significantly correlated with precipitation. Furthermore, authors notified that increasing temperature and decreasing precipitation caused to decrease in the streamflow of Filyos River. Likewise, Kale and Sönmez (2018a) stated that decreasing trend in the streamflow of Akkaya Stream could be related to decline in precipitation and snowmelt besides increasing temperature due to the climate change. Therefore, evaluating possible effects of the climate change on the river streamflow should be continuously monitored. The past, present and future of the water resources can be monitored by

applying trend analysis on hydrologic time series data. The outputs of trend analysis will benefit for the water resource managers and decision-makers to make the most appropriate management policy.

#### 4. CONCLUSION

Decreasing trends for the streamflow of Araç Stream have been predicted for the monitoring period. These decreasing trends could be linked with decrease in snowmelt and rainfall, in addition to extremely increase in temperature and evaporation patterns of air or water, and other causes by the climate change. Thus, streamflow trends should be monitored to estimate the amount and availability of water resources in the future.

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#### REFERENCES

- Albek E (2002) Statistical Analysis of Water Quality Trends: An Application to the Porsuk Stream. Anadolu University Journal of Science and Technology, 3 (2): 281–292.
- Ay M & Özyıldırım S (2017) Trend Analysis of Monthly Total Rainfall and Monthly Mean Air Temperature Variables of Yozgat in Turkey. Çukurova University Journal of the Faculty of Engineering and Architecture, 32 (2): 65-75.
- Bahadir M (2011) A Statistical Analysis of the Flow Changes of Kızılırmak River. Turkish Studies International Periodical for the Languages, Literature and History of Turkish or Turkic, 6: 1339-1356.
- Birsan M V, Molnar P, Burlando P & Pfaundler M (2005) Streamflow Trends in Switzerland. Journal of Hydrology, 314: 312-329.
- Box G E P & Jenkins G (1976) Time Series Analysis: Forecasting and Control. Holden Day, San Francisco.
- Büyükyıldız M & Berktay A (2004) Parametrik Olmayan Testler Kullanilarak Sakarya Havzasi Yağişlarinin Trend Analizi. Journal of Faculty of Engineering and Architechture of Selcuk University, 19 (2): 23-38.
- Cigizoglu H K, Bayazit M & Onoz B (2005) Trends in the Maximum, Mean and Low Flows of Turkish Rivers. Journal of Hydrometeorology, 6 (3): 280-290.
- Doğan Demir A, Şahin Ü & Demir Y (2016) Trend Analysis and Agricultural Perspective Availability of Water Quality Parameters at Murat River. Yuzuncu Yıl University Journal of Agricultural Sciences, 26 (3): 414-420.
- Douglas E M, Vogel R M & Kroll C N (2000) Trends in Floods and Low Flows in the United States: Impact of Spatial Correlation. Journal of Hydrology, 240 (1–2): 90-105.
- Durdu Ö F (2010) Effects of Climate Change on Water Resources of the Büyük Menderes River Basin, Western Turkey. Turkish Journal of Agriculture and Forestry, 34: 319-332.
- Ejder T, Kale S, Acar S, Hisar O & Mutlu F (2016a) Restricted Effects of Climate Change on Annual Streamflow of Sarıçay Stream (Çanakkale, Turkey). Marine Science and Technology Bulletin, 5 (1): 7-11.
- Ejder T, Kale S, Acar S, Hisar O & Mutlu F (2016b) Effects of Climate Change on Annual Streamflow of Kocabaş Stream (Çanakkale, Turkey). Journal of Scientific Research and Reports, 11 (4): 1-11.
- Ercan B & Yüce M İ (2017) Trend Analysis of Hydro-Meteorological Variables of Kızılırmak Basin. Nevşehir Bilim ve Teknoloji Dergisi, 6 (ICOCEE 2017 Özel Sayı): 333-340.
- Helsel D R & Hirsch R M (2002) Statistical Methods in Water Resources Techniques of Water Resources Investigations, Book 4, Chapter A3. U.S. Geological Survey. 522 p.
- Herawati H, Suripin & Suharyanto (2015) Impact of Climate Change on Streamflow in the Tropical Lowland of Kapuas River, West Borneo, Indonesia. Procedia Engineering, 125: 185-192.
- Hirsch R M, Slack J R & Smith R A (1982) Techniques of Trend Analysis for Monthly Water Quality Analysis. Water Resources Research, 18 (1): 107–121.
- Hisar O, Kale S & Özen Ö (2015) Sustainability of Effective Use of Water Sources in Turkey. In: Leal Filho, W., Sümer, V. (Eds.), Sustainable Water Use and Management: Examples of New Approaches and Perspectives. Springer International Publishing, Switzerland, pp. 205-227.
- Kadıoğlu M (1997) Trends in Surface Air Temperature Data over Turkey. International Journal of Climatology, 15: 511-520.
- Kahya E & Kalaycı S (2004) Trend Analysis of Streamflow in Turkey. Journal of Hydrology, 289: 128-144.
- Kalayci S & Kahya E (1998) Detection of Water Quality Trends in the Rivers of the Susurluk Basin. Turkish Journal of Engineering and Environmental Science, 22: 503-514.
- Kale S (2017a) Climatic Trends in the Temperature of Çanakkale City, Turkey. Natural and Engineering Sciences, 2 (3): 14-27.

Kale S (2017b) Analysis of Climatic Trends in Evaporation for Çanakkale (Turkey). Middle East Journal of Science, 3 (2): 69-82.

- Kale S & Sönmez A Y (2018a) Trend Analysis of Streamflow of Akkaya Stream (Turkey). In: Proceedings of the 1st International Conference on Food, Agriculture and Animal Sciences, Antalya, Turkey. pp. 33-45.
- Kale S & Sönmez A Y (2018b) Trend Analysis of Mean Monthly, Seasonally and Annual Streamflow of Daday Stream in Kastamonu, Turkey. Marine Science and Technology Bulletin, 7 (2): 60-67.
- Kale S, Ejder T, Hisar O & Mutlu F (2016a) Climate Change Impacts On Streamflow of Karamenderes River (Çanakkale, Turkey). Marine Science and Technology Bulletin, 5 (2): 1-6.
- Kale S, Ejder T, Hisar O & Mutlu F (2016b) Effect of Climate Change on Annual Streamflow of Bakırçay River. Adıyaman University Journal of Science, 6 (2): 156-176.
- Kale S, Hisar O, Sönmez A Y, Mutlu F & Filho W L (2018) An Assessment of the Effects of Climate Change on Annual Streamflow in Rivers in Western Turkey. International Journal of Global Warming, 15 (2): 190-211.
- Kendall M G (1955) Rank Correlation Methods. 2nd ed. Hafner Publishing Co., New York. 196 p.
- Kişi Ö, Guimarães Santos C A, Marques da Silva R & Zounemat-Kermani M (2018) Trend Analysis of Monthly Streamflows using Şen's Innovative Trend Method. Geofizika, 35 (1): 53-68.
- Koçman A & Sütgibi S (2012) Hydrograpic/Hydrologic Characteristics of Gediz River Basin in the Context of the Environmental Components Problems and Suggestions. Eastern Geographical Review, 28: 155-174.
- Kundzewicz Z W, Graczyk D, Maurer T, Pińskwar I, Radziejewski M, Svensson C & Szwed M (2005) Trend Detection in River Flow Series: 1. Annual Maximum Flow. Hydrological Sciences–Journal–des Sciences Hydrologiques, 50 (5): 797-810.
- Lehmann E L (1975) Nonparametrics: Statistical Methods Based on Ranks. Holden-Day, San Francisco, CA, USA. 457 p.
- Mann H B (1945) Nonparametric Tests against Trend. Econometrica, 13: 245-259.
- Ozkul S (2009) Assessment of Climate Change Effects in Aegean River Basins: The Case of Gediz and Buyuk Menderes Basins. Climatic Change, 97: 253-283.
- Pettitt A N (1979) A Non-Parametric Approach to the Change-Point Problem. Journal of the Royal Statistical Society. Series C (Applied Statistics), 28: 126-135.
- Pumo D, Caracciolo D, Viola F & Noto L V (2016) Climate Change Effects on the Hydrological Regime of Small Non-Perennial River Basins. Science of The Total Environment, 542 (Part A): 76-92.
- R Core Team (2019) R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria [online] https://www.R-project.org/ version (01/2019).
- Saplıoğlu K, Kilit M & Yavuz B K (2014) Trend Analysis of Streams in the Western Mediterranean Basin of Turkey. Fresenius Environmental Bulletin, 23 (1): 1-12.
- Sen P K (1968) Estimates of the Regression Coefficient Based on Kendall's Tau. Journal of the American Statistical Association, 63 (324): 1379–1389.
- Sneyers R (1990) On the Statistical Analysis of Series of Observations. World Meteorological Organization, Technical Note no. 143, WMO no. 415. 192 p.
- Sütgibi S (2015) Büyük Menderes Havzasının Sıcaklık, Yağış ve Akım Değerlerindeki Değişimler ve Eğilimler. Marmara Coğrafya Dergisi, 31: 398-414
- Salarijazi M, Akhond-Ali A-M, Adib A & Daneshkhah A (2012) Trend and Change-Point Detection for the Annual Stream-Flow Series of the Karun River at the Ahvaz Hydrometric Station. African Journal of Agricultural Research, 7 (32): 4540-4552.
- Sönmez A Y & Kale S (2018) Climate Change Effects on Annual Streamflow of Filyos River (Turkey). Journal of Water and Climate Change (In press). DOI: 10.2166/wcc.2018.060.
- Şen Z (2012) Innovative Trend Analysis Methodology. Journal of Hydrologic Engineering, 17 (9): 1042-1046.
- Tosunoglu F & Kisi O (2017) Trend Analysis of Maximum Hydrologic Drought Variables Using Mann–Kendall And Şen's Innovative Trend Method. River Research and Applications, 33: 597-610.
- Tosunoğlu F (2017) Trend Analysis of Daily Maximum Rainfall Series in Çoruh Basin, Turkey. Iğdır University Journal of the Institute of Science and Technology, 7 (1): 195-205.
- Türkeş M & Acar Deniz Z (2011) Climatology of South Marmara Division (North West Anatolia) and Observed Variations and Trends. International Journal of Human Sciences, 8: 1579-1600.
- Yeh C F, Wang J, Yeh H F & Lee C H (2015) Spatial and Temporal Streamflow Trends in Northern Taiwan. Water, 7 (2): 634-651.
- Yenigün K & Ülgen M U (2016) Trend Analysis of Maximum Flows under Climate Change Evaluation and Its Impact on Spillway. Disaster Science and Engineering, 2 (1): 25-28.
- Yıldırım U, Yılmaz İ Ö & Akınoğlu B G (2013) Trend Analysis of 41 Years of Sunshine Duration Data for Turkey. Turkish Journal of Engineering & Environmental Sciences, 37: 286-305.

- Zhang Q, Liu C, Xu C, Xu Y & Jiang T (2006). Observed Trends of Annual Maximum Water Level and Streamflow During Past 130 Years in the Yangtze River Basin, China. Journal of Hydrology, 324 (1-4): 255–265.
- Zhang X, Harvey K D, Hoggy W D, Yuzyk T R (2001) Trends in Canadian Streamflow. Water Resources Research, 37 (4): 987-998.
- Zhou Y, Shi C, Fan X & Shao W (2015) The Influence of Climate Change and Anthropogenic Activities on Annual Runoff of Huangfuchuan Basin in Northwest China. Theoretical and Applied Climatology, 120: 137-146.



## Distribuion and Clinical Manifestation of Toxoplasma Invasion in Domestic Carnivores

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Abstract: In the modern world, the problem of toxoplasmosis is becoming increasingly urgent. It is assumed that the population of the entire planet is invased up to 65% by parasites of Toxoplasma gondii. According to official statistics, the number of people infected with toxoplasma in the world is about 1.5 billion people. But according to various authors, the percentage of infection differs significantly from country to country, from 22% in the United Kingdom to more than 88% in France, while in South Korea the infection is very low, only 4.3%, and in Brazil it is over 66.9%. In Ukraine, about a third of the population under the age of 25 has antibodies to toxoplasma. As age increases - infection of percentage of population is increasing, too. Today toxoplasmosis has been registered in more than 300 species of mammals and 60 species of poultry. Up to 90% of cats in different countries of the world are affected by pathogens of toxoplasmosis. In sinanthropic focus, the source of invasion is cattle, sheep, goats, camels, horses, donkeys, pigs, carnivores, ducks, geese, turkeys, guinea fowls, parrots and other animals. Given these data, it should be noted that the issue of diagnosis, differentiation and treatment of toxoplasmosis is extremely relevant at present. For the study, the serum of homemade omnivores (cats and dogs) was used. Their owners for various reasons turned to the veterinary clinic for medical assistance. In total, 703 blood serums were examined (including 378 from cats (53.8%) and 325 from dogs (46.2%)). Blood was removed from the elbow and centrifuged at 1800 rpm. Further research on the titre of specific antibodies against toxoplasma was performed by the ELISA method using the "Hema" test system. Studies show a high degree of seropositive results in domestic cats and dogs in relation to toxoplasma invasion. Taking into account the results of these studies, the incidence of toxoplasma invasion in dogs and cats from the seasons of year was analyzed. An increase in the number of seropositive tests for toxoplasmosis in cats in summer was found. This is probably due to the increase of the population of mice and rats in the warm season. According to Dubey J. P. (2008), while eating them cats are infected with toxoplasmosis. An additional favorable factor of infection is the ambient temperature, since the temperature optimum for sporulation of the oocysts is 20-24°C. Analyzing the indicators, it is necessary to note the differences in clinical manifestations in seropositive for toxoplasmosis dogs and cats. So dogs are dominated by neurological problems (38%) which appear in the form of paresis of limbs, epileptic seizures, spontaneous aggression, lesion of the visual system. In cats, recorded lesions more often (49.5%) are not related to neurology and dermatitis, and hyperglycemia is a significant part of such manifestations. It was found that 24.9% of blood serum from domestic animals responded positively to ELISA for toxoplasma invasion. There is a difference in the clinical manifestation of toxoplasmosis in dogs and cats. At the same time, dogs often register pathology from the nervous system.

Keywords: Toxoplasmosis, immunity, leukocytes, clinical signs, antibodies



## Financial Effects of HPAI H5N1 Cases on Backyard Poultry in the Outbreak Area

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**Abstract:** Influenza viruses in poultry and mammals such as humans, pigs, horses, cats and dogs have caused major economic losses. Then this study was carried out to investigate the risk factors relating Highly Pathogen Avian Influenza H5N1 and to evaluate consumer demand and related economic losses for poultry raised in an outbreak area. Data were obtained from 361 householders in the outbreak area and surrounding rural areas where a Highly Pathogen Avian Influenza outbreak occurred in 2008. The data set obtained from the surveys was analysed with the SPSS. Evaluating of the data descriptive statistical methods (number, percentage, mean, standard deviation, maximum and minimum) were used. Kolmogorov-Smirnov test was applied to the normal distribution of the study variables. The total cost of the disease outbreak in the area was estimated as TL 501.768 for the 3.116 enterprises. Based on market prices for 2011, the compensation cost for the disease was estimated at TL 276 per enterprise. The time of the Highly Pathogen Avian Influenza outbreaks had happenned, correlation between the consumption of eggs and poultry families were significant at 0.05 level. In conclusion, the management of epidemic diseases, particularly avian influenza, the re-establishment of poultry farming and Broiler sector after disease outbreaks is of great importance. The present study contributed to the determination of production losses due to the disease Highly Pathogen Avian Influenza, disease-related control and protection measures, estimated payments and direct economic effects. Correctly estimating the number of affected poultry is essential for the determination of identifying the possible loss amounts.

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Keywords: HPAI, poultry, production loss, risk assessment, outbreak



## Effect of Season on Footpad Dermatitis of Broiler Chickens

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**Abstract:** This research is a survey study in which slaughterhouse records are used to examine the effect of season on footpad dermatitis (FPD) in broiler chickens. In this research, 24 295 573 Ross 308 line chicken poultry feet records which were transferred to slaughterhouse between October 2016 and September 2018 in Samsun province. FPD is a welfare concern in broiler chickens as the dermatitis cause pain, reduced weight gain and decreases in feed intake. Several factor associated with FPD such as litter type and thickness, season, lighting schedule, breed, stocking density, content of diet and drinker system. Prominent factor causing FPD is wet litter. The litter condition was affected by season as FPD related to temperature and humidity. Foot lesions were scored according to presence or absence of lesions between Score 0 and Score 2 after slaughter. The ratio of chickens' according to the scores and the ratio of chickens' according to the season were determined. The highest score ratio was 36.12% with Score 2 and the lowest score ratio was 16.09% with score 0. The effect of season on FPD was significant (P<0.001). Score 3 was the highest in autumn (29.56%), while Score 2 was the highest in spring (38.51%). The ratio of chicken feet with lesion-free or little lesion (Score 0 and Score 1) was found to be 38.83%, while the middle or extensive score lesion (Score 2 and 3) was 61.17%. As a result, the effect of season on FPD was significant. In order to reduce the economic loss caused by FPD, it is necessary to increase the rate of foot with lesion-free or little lesion foot and to decrease the ratio of foot middle or extensive lesion foot. Type and thickness of litter, ventilation of pen, type of feed is important for FPD especially season of spring and autumn.

Keywords: Broiler chicken, season, foot pad dermatitis, animal welfare, Ross 308

### **1. INTRODUCTION**

It is necessary to take 80 g of protein daily and at least half of it should be animal protein in order to have enough and balanced nutrition and to be resistant to diseases. The demand for animal products is constantly increasing, and in the shortest period of time, the growing of animals in which animal proteins can be obtained is getting more and more valuable. Poultry especially broiler chickens maintains its first place in this respect and attracts more attention especially in countries with animal protein deficits (Altınel, 1999).

Poultry meat production in Turkey is 1 894 669 tons annually. This production level having a proportion 2% in the world and Turkey provided 60% of the total meat production from poultry meat (FAO, 2014).

At the same time, important income is obtained from the foot of the broiler meat which is an important source of animal protein and source of income. East and Southeast Asia has become a key value store for Turkey in recent years.

There is increasing demand for good quality broiler foot in Turkey. The importance of FPD is increasing as it causes economic loss or destruction of lowering the quality (Shepherd et al., 2010). The severity and incidence of dermatitis in the foot base, tarsal region and chest area is an important criterion for evaluating the welfare of broilers. The most common type in this dermatitis is FPD (Hester, 1994; Berg, 2004). Pain, difficulty in walking, difficulty in accessing to feed and water and a decrease in body weight can be observed depending on the severity of dermatitis (Skrbic et al., 2012).

The main factors that cause FPD are the type and thickness of the litter, temperature and humidity values of the indoor and outdoor environment, lighting program and light distribution (Jong et al., 2013), race, age, feed content, stocking density in the house and irrigation system (Ekstrand et al., 1998). Among these factors, the seasonal effect of temperature and humidity values of the external environment of the house has an important role on FPD. The FPD ratios and intensity seen in broiler chickens vary due to the change in the moisture content of the litter material used in the poultry.

In the study conducted by Ekstrand et al. (1998), the increase in the lesions on the base and fingers of broiler chickens has been reported in the winter months when the relative humidity is increased. This increase was reported by Bruce et al. (1990) reported that it was 28% and higher in winter than that broiler chickens raised in summer.

In addition, some researchers have reported an increase in the incidence of FPD in the spring season (Musilova et al., 2013) and in the fall season (Costa et al., 2014) by some researchers.

This research was conducted on a large population reflecting the commercial conditions, while previous studies were conducted on an experimental and limited sample. This survey study, which was carried out using the slaughterhouse records, was conducted to determine the effect of season on the formation of FPD in broiler chickens.

### 2. MATERIAL AND METHOD

The records were surveyed about between October 2016 and September 2018 for a period of two years transferred broilers chickens of 24 295 573 Ross 308 line in Samsun province. In this study, the feet were evaluated macroscopically by the expert personnel according to the presence or absence of the lesion on the soles of the feet and / or fingers, following the cutting of broiler chickens brought to the slaughterhouse. Lesion scoring was performed according to Martrenchar et al. (2002) by three expert persons. If there is no lesion Score 0; if the lesion has spread to 25% of the foot sole Score 1; if lesion is between 25% and 50% of the foot base Score 2; if the lesion was more than 50% and / or spread to the toes Score 3. The numbers of foot was divided into two and the number of chickens was obtained. Chi square test was applied to determine the effect of the season on the FPD (SPSS 2013).

### 3. RESULTS AND DISCUSSION

The number of chickens according to the scores is given in Table 1. According to this, the highest score was in Score 2 with a score of 36.12% and the lowest score was in Score 0 with a score of 16.09%. Score 3, the most common lesion score, was determined as 25.05% in all chicken feet. It was found that the ratio of chicken feet with the middle or common lesion (Score 2 and 3) was 61.17%, while the ratio of chicken feet without lesion or with little lesion (Score 0 and Score 1) was 38.83%.

| Season      | Score 0 (n)        | Score 1 (n)         | Score 2 (n)        | Score 3 (n)         | Overall (n)        |
|-------------|--------------------|---------------------|--------------------|---------------------|--------------------|
| Spring      | 1 009 296          | 1 288 593           | 2 386 796          | 1 513 648           | 6 198 333          |
| Summer      | 1 311 852          | 2 172 333           | 2 849 833          | 2 037 037           | 8 371 055          |
| Autumn      | 696 944            | 913 500             | 1 847 870          | 1 451 352           | 4 909 666          |
| Winter      | 892 519            | 1 149 444           | 1 690 704          | 1 083 852           | 4 816 519          |
| Overall (%) | 3 910 611 (%16.09) | 5 523 870 (% 22.74) | 8 775 203 (%36.12) | 6 085 889 (% 25.05) | 24 295 573 (% 100) |

Table 1. The number of broilers and rates with FPD and by season

The rates and significance of broiler chickens scored according to the seasons are given in Table 2. The effect of season on FPD in broiler chickens was significant (P<0.001). According to this, the Score 2 was the highest in spring (38.51%), second in autumn (37.64%), while score 3 was the highest in autumn (29.56%).

Table 2. The rates of FPD by season and scores

| Score 0 (%) | Score 1 (%)                                     | Score 2 (%)  | Score 3 (%)   | <b>Overall %</b>   | $\chi^2$  | Significance  |
|-------------|---|--|---|--|---|---|
| 16.28       | 20.79   | 38.51  | 24.42   | 100  |   |   |
| 15.67       | 25.96   | 34.04  | 24.33   | 100  |   |   |
| 14.19       | 18.61   | 37.64  | 29.56   | 100  | 197064.358  | ***   |
| 18.53       | 23.87   | 35.10  | 22.50   | 100  |   |   |
|             | Score 0 (%)<br>16.28<br>15.67<br>14.19<br>18.53 | Score 0 (%)Score 1 (%)16.2820.7915.6725.9614.1918.6118.5323.87 | Score 0 (%)Score 1 (%)Score 2 (%)16.2820.7938.5115.6725.9634.0414.1918.6137.6418.5323.8735.10 | Score 0 (%)Score 1 (%)Score 2 (%)Score 3 (%)16.2820.7938.5124.4215.6725.9634.0424.3314.1918.6137.6429.5618.5323.8735.1022.50 | Score 0 (%)Score 1 (%)Score 2 (%)Score 3 (%)Overall %16.2820.7938.5124.4210015.6725.9634.0424.3310014.1918.6137.6429.5610018.5323.8735.1022.50100 | Score 0 (%)Score 1 (%)Score 2 (%)Score 3 (%)Overall %χ²16.2820.7938.5124.4210015.6725.9634.0424.3310014.1918.6137.6429.56100197064.35818.5323.8735.1022.50100 |

\*\*\* P<0.001

Hashimoto et al. (2011) studied on the numbers of 8985 broilers and concluded that broilers of 13.1% was Score 0; 33.3% was score 1; 33.4% was score 2 and 20.2% was score 3. Jong et al. (2012) studied on the numbers of 386 broiler chickens for two years, and they reported that 35.5% of broiler chickens had no lesion on their feet, 26% had moderate and 38.4%

had severe lesion. Berg (1998) reported that 5-10% of broiler chickens had severe lesions and 10-35% of broilers had moderate lesions in Swiss.

In this study, Scores 0, 1, 2 and 3 was 16.09%; 22.74%; 36.12% and 25.05%, respectively. These results were similar to the results of Jong et al. (2012). In addition, Berg (1998) and Haşimoto et al. (2011) found the rate of the foot with severe and middle lesion (Score 2 and 3) were lower than that the present study. It is thought that this is due to differences in maintenance and management conditions transported broiler to the slaughterhouse.

In this study, Score 3 was first as 29.56% in autumn season and was second as 24.42% in spring season. In addition, Score 2 was first as 38.51% in spring and was second as 37.64% in autumn. The results of the present study were similar to the results of Musilova et al. (2013) and Costa et al. (2014). In other words, it has been reported by these researchers that the rate of chicken feet with moderate and widespread lesions was higher in autumn and spring season than that in other seasons.

On the other hand, in the studies conducted by other researchers (Haslam et al., 2007; Meluzzi et al., 2008; Haşimoto et al. 2011; Jong et al. 2012), it has been reported that the Score 2 and Score 3 rates of broiler chickens transported to the slaughterhouse were higher in winter than that in other seasons. The reason for this difference may be due to the different types of litter used in the house and humidity. However, in most of the studies about FPD, it was found that the rate of middle and widespread chicken feet was the lowest in summer. This finding is consistent with the results of the present study. This reason may be the relative humidity is low although the temperature is high, thereby low humidity may have resulted from low humidity in the farm.

### 4. CONCLUSION

In conclusion, the season has an important impact on FPD, which is an important criterion for animal welfare and causes significant economic damage. In order to increase the efficiency of poultry welfare and to reduce the economic loss due to FPD, it is necessary to increase the rate of foot without lesions and to decrease the rate of foot middle or widespread lesion. For this purpose, it is necessary to pay attention to the type and thickness of the litter, the ventilation of the farm, stocking density in the farm, the feed characteristics.

### REFERENCES

- Allain, V., Mirabito, L., Arnould, C., Colas, M., Le, Bouquin, S., Lupo, C., Michel, V., 2009. Skin lesions in broiler chickens measured at the slaughterhouse: relationships between lesions and between their prevalence and rearing factors. British Poultry Science 50: 407-417.
- Altınel, A., 1999. Özel Zootekni (Tavuk Yetiştirme), İstanbul. İstanbul Üniversitesi Veteriner Fakültesi Yayını, Ders Notu No: 103, 1-38.
- Berg, C. C., 1998. .Foot-Pad Dermatitis in Broilers and Turkeys Prevalance, risk factors and prevention. Doctoral thesis, Swedish University of Agricultural Sciences, Uppsala,
- Berg, C., 2004. Pododermatitis and hock burn in broiler chickens. Page 37 in Measuring and Auditing Broiler Welfare. C. A. Weeks and A. Butterworth, ed. CABI Publishing, Wallingford, UK.
- Bruce, D. W., Mcilroy, S. G., Goodall, E. A., 1990. Epidemiology of a Contact-Dermatitis of Broilers. Avian Pathology 19: 523-537.
- Costa, M. J. D., Grimes, J. L., Oviedo-Rondon, E. O., Barasch, I., Evans, C., Dalmagro, M., Nixon, J., 2014. Footpad dermatitis severity on turkey flocks and correlations with locomotion, litter conditions, and body weight at market age. The Journal Applied Poultry Research 23: 268-279.
- Ekstrand, C., Carpenter, T. E., Andersson, I., Algers, B., 1998. Prevalence and control of footpad dermatitis in broilers in Sweden. British Poultry Science 39: 318-324.
- FAO, 2014. Chicken meat in Turkey. http://www.fao.org/faostat/en/#data/QL
- Hashimoto, S., Yamazaki, K., Obi, T., Takase, K., 2011. Footpad Dermatitis in Broiler chickens in Japan. The Journal of Veterinary Medical Science 73: 293-297.

- Haslam, S. M., Knowles, T. G., Brown, S. N., Wilkins, L. J., Kestin, S. C., Warriss, P. D., Nicol, C. J., 2007. Factors affecting the prevalence of foot pad dermatitis, hock burn and breast burn in broiler chicken. British Poultry Science 48: 264-275.
- Hester, P. Y., 1994. The role of environment and management on leg abnormalities in meat-type fowl. Poultry Science 73: 904-915.
- Jong, I. C., Veldkamp, T., Van Harn, J., 2013. Management tools to reduce footpad dermatitis in broiler chickens. Pages 78–83 in Proc. of the 19th Eur. Symp. On Poult. Nutr. Working Group 2: Nutrition of the European Federation of Branches of the World Poultry Science Association, Potsdam, Germany.
- Jong, L. C., Harn, J. V., Gunnink, H., Hindle, V. A., Lourens, A., 2012. Footpad dermatitis in Dutch broiler flocks: Prevalance and factors of influence. Poultry Science 91: 1569-1574.
- Martrenchar, A., Boilletot, E., Huonnic, D., Po, IF., 2002. Risk factors for foot-pad dermatitis in chicken and turkey broiler in France. Preventive Veterinary Medicine 52; 213-226.
- Meluzzi, A., Fabbri, C., Folegatti, E., Sirri, F., 2008. Survey of chicken rearing conditions in Italy: effects of litter quality and stocking density on productivity, foot dermatitis and carcase injuries. British Poultry Science 49: 257-264.
- Musilova, A., Lichovnikova, M., Hampel, D., Przywarova, A., 2013. The Effect of the season on icidence of footpad dermatitis and its effect on broilers performance. Acta Universitatis agriculturae et silviculturae mendellianae Brunensis. 6: 1793-1798.
- Shepherd, E. M, and Fairchild, B. D., 2010. Footpad dermatitis in poultry. Poultry Science 89: 2043-2051.
- Skrbic, Z., Pavlovski, Z., Lukić, M., Petričević, V., Miljković, B., Marinkov, G., 2012. The effect of the diet on incidence of footpad lesions and productivity of broilers. Biotechnology. Animal Husbandry 28: 353-360.
- SPSS: Statistical package for the social sciences, Release 2013..22.0, SPSS Inc, Chicago, II, USA,



## The Relationship of Animal Orginated Food Demand and Income

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**Abstract:** In this study, the relationship between changes in population and income occurring with variable amounts of animal product consumption in Turkey between 2003 and 2017 were investigated. Secondary data sources were used in the study to evaluate the extent to which expectations for increase of animal product consumption were realized. The data of national income per capita, demografic, consumption of major livestock product (cattle and buffaloes meat, mutton and goat meat, poultry meat, cow, sheep, goat and buffaloes milk) were evaluated in the period of between 2003 and 2017. The situation analysis method was used in the study and consumption of animal-orginated food products were determined. Quantities on animal-originated food product per capita is calculated by dividing the major animal-originated food product (beef, cow milk, chicken meat and mutton-goat meat) amounts to the total population of Turkey.As a result of correlation analysis, a high level of significant correlation was found between all animal product consumption and income except the consumption. In this study, the total meat consumption in Turkey is 18 kg per person in 2003, it was determined that in 2017 reached 32 kg. In conculusion, when people's income increases, people consumption of animal-orginated food consumption increases. This trend goes on up to a certain level so it is expected that investments in animal sector is positively affect on animal sector and have a positive return.

Keywords: Consumption, demand, income, meat, situation analysis

## **1. INTRODUCTION**

Increasing the demand of livestock products driven by urbanization rate, income growth and human population growth (Delgado, 2005). The demand change country livestock farmer profile, social and cultural structure, expectations of society, regulations of the value chain, relegious beleives, macro economic development level of countries effects livestock production systems and will continue at a later time.

Positive macro economic trends and increasing the national income per capita will increase the demand of animal-source food products.

In this study, the per capita income and the change in consumption of animal products were investigated and the change in the data of 2003-2017 was investigated. In an approach that takes into account the changes occurring in the population in 15 years, the effects of the animal production demand on the changes in income were investigated.

## 2. MATERIAL AND METHOD

Material of the study was obtained statistical data of Food and Agricultural Organization of the United Nations (Faostat, 2019), Turkish Statistical Institute (Turkstat, 2019). Situation analysis is used in the study (Clarke et all, 2018).

The key drivers of the study are gross national product per capita, demographic data on total population, major animal products amount and per capita consumption of animal- originated food products were examined by the years of 2003-2017 data.

Quantities on animal-originated food product per capita is calculated by dividing the major animal-originated food product (beef, cow milk, chicken meat and mutton-goat meat) amounts to the total population of Turkey.

National income increase (including ten years15 period) whether affect the demand of animal source food or not was searched in the study. According to the technical acceptance, there is no export and import of animal products in the searched term. Descriptive statistics (Means and St. deviation) per capita on national income, consumption of beef, cow milk, chicken meat and mutton-goat meat and correlation national income and consumption of beef, cow milk, chicken meat and mutton-goat meat are searched in the study with using SPSS 20 program (SPSS, 2013).

### 3. RESULTS AND DISCUSSION

Changes in income and population variables in years are given in Table 1. The year 2003 was determined as the index year and the changes in the parameters examined in the process until 2017 were calculated. The share of national income is given annually per capita.

| Year | National income (US\$) | Index | Population | Index |
|------|------------------------|-------|------------|-------|
| 2003 | 4,59                   | 100   | 66.873.000 | 100   |
| 2004 | 5,76                   | 126   | 67.734.000 | 101   |
| 2005 | 7,02                   | 154   | 68.582.000 | 103   |
| 2006 | 7,59                   | 166   | 69.421.000 | 104   |
| 2007 | 9,24                   | 203   | 70.586.000 | 106   |
| 2008 | 10,38                  | 229   | 71.517.100 | 107   |
| 2009 | 8,56                   | 188   | 72.561.312 | 109   |
| 2010 | 10,02                  | 220   | 73.722.988 | 110   |
| 2011 | 10,47                  | 230   | 74.724.269 | 112   |
| 2012 | 10,5                   | 230   | 75.627.384 | 113   |
| 2013 | 12,8                   | 279   | 76.667.864 | 115   |
| 2014 | 12,12                  | 264   | 77.695.904 | 116   |
| 2015 | 11,19                  | 244   | 78.741.053 | 118   |
| 2016 | 10,83                  | 236   | 79.814.871 | 119   |
| 2017 | 10,02                  | 218   | 80.810.525 | 121   |

Table 1. Population and gross domestic product data of Turkey

Index:2003=100

The per capita annual consumption amounts of some selected animal products and the changes in years are given in Table 2.

Table 2. Annual consumption of animal orginated food product of Turkish people (kg/per capita/year)

| Year | Beef and veal meat | Index | Cow milk | Index | Sheep and Goat meat | Index | Chicken meat | Index |
|------|--------------------|-------|----------|-------|---------------------|-------|--------------|-------|
| 2003 | 4,4                | 100   | 101      | 100   | 4,7                 | 100   | 13,0         | 100   |
| 2004 | 5,4                | 123   | 101      | 100   | 4,7                 | 100   | 12,9         | 99    |
| 2005 | 4,7                | 107   | 105      | 104   | 4,6                 | 98    | 13,3         | 102   |
| 2006 | 4,9                | 111   | 112      | 111   | 4,6                 | 98    | 14,7         | 113   |
| 2007 | 6,1                | 139   | 113      | 112   | 4,6                 | 98    | 14,4         | 111   |
| 2008 | 5,2                | 118   | 157      | 155   | 4,5                 | 95    | 15,2         | 117   |
| 2009 | 4,5                | 102   | 160      | 158   | 4,1                 | 88    | 17,8         | 137   |
| 2010 | 8,4                | 191   | 168      | 166   | 3,7                 | 79    | 19,6         | 151   |
| 2011 | 8,6                | 196   | 185      | 183   | 3,9                 | 84    | 21,6         | 166   |
| 2012 | 10,6               | 240   | 211      | 209   | 4,2                 | 90    | 22,8         | 175   |
| 2013 | 11,3               | 258   | 217      | 215   | 4,6                 | 97    | 22,9         | 176   |
| 2014 | 11,4               | 258   | 219      | 217   | 4,7                 | 103   | 24,4         | 188   |
| 2015 | 12,9               | 293   | 215      | 213   | 5,1                 | 109   | 24,2         | 186   |
| 2016 | 13,3               | 302   | 210      | 208   | 5,1                 | 108   | 23,5         | 181   |
| 2017 | 12,2               | 278   | 232      | 230   | 5,0                 | 105   | 26,4         | 203   |

Index: 2003=100

Descriptive statistics for the parameters examined in the study are given in Table 3.

| Parametres                        | Ν  | Minimum | Maximum | Mean   | Std. Deviation |
|-----------------------------------|----|---------|---------|--------|----------------|
| Income                            | 15 | 5       | 13      | 9,41   | 2,30           |
| Income index                      | 15 | 100     | 279     | 205,80 | 50,24          |
| Population index                  | 15 | 100     | 121     | 110,27 | 6,71           |
| Beef consumption                  | 15 | 4       | 13      | 8,25   | 3,40           |
| Beef consumption index            | 15 | 100     | 302     | 187,73 | 77,39          |
| Cow milk                          | 15 | 101     | 232     | 167,10 | 49,67          |
| Cow milk index                    | 15 | 100     | 230     | 165,40 | 49,19          |
| Sheep-goat meat consumption       | 15 | 4       | 5       | 4,55   | ,41            |
| Sheep-goat meat consumption index | 15 | 79      | 109     | 96,80  | 8,50           |
| Chicken consumption               | 15 | 13      | 26      | 19,12  | 4,86           |
| Chicken consumption index         | 15 | 99      | 203     | 147,00 | 37,31          |

Table 3. Descriptive statistics of the study parameters

In the study, the correlations between income and animal product consumption and population and animal product consumption are given in Table 4.

**Table 4.** Correlation of the study parametres

| Parametres                           | Ν  | R      |
|--------------------------------------|----|--------|
| Population-Income                    | 15 | ,825** |
| Income-Beef consumption              | 15 | ,813** |
| Income -Cow milk consumption         | 15 | ,855** |
| Income -Sheep and goat meat          | 15 | ,018   |
| Income – Chicken meat consumption    | 15 | ,805** |
| Population-Beef consumption          | 15 | ,940** |
| Population-Cow milk consumption      | 15 | ,963** |
| Population -Sheep and goat meat      | 15 | ,282   |
| Population -Chicken meat consumption | 15 | ,975** |

\*\* Correlation is significant at the 0.01 level

The data of the study shows that Turkey's annual population growth rate at 1.3 per cent between the 2003-2012, but this rate is increased in 2003-2017 up to level 1.4. The average per capita Gross Domestic Product is 9,410 U.S. \$ in 15 years period. The same period people's animal orginated food consumption

Average beef and veal meat consumption; 8,25 (kg/percapita)

Average sheep and goat meat consumption; 4,55 (kg/percapita)

Average chicken meat consumption; 19,12 (kg/percapita)

Average cow milk consumption; 167,10 (kg/percapita)

In developing countries, annual per capita meat consumption amount (beef and beef, pork, chicken meat, mutton and goat meat) was reported as 28 kg in 2002 and 44 kg in milk consumption (Thornton, 2010).

In developed countries, annual meat consumption was reported as 78 kg in 2002 and 202 kg in milk consumption (Thornton, 2010).

In 2003, Turkey's annual per person consumption of meat (beef and veal, poultry, mutton and goat meat) is 22.10kg. Milk consumption is calculated as 101 kg. In 2017, the total meat consumption increased to 32 kg and the milk consumption

increased to 232 kg. Turkey's total meat consumption of 18 kg in 2003. In 2017 it reached 32 kg. The gap between the world average is closed (OECD, 2019).

While the average growth rate of world milk production has increased by about 2.1% in the last decade, the demand for milk and dairy products per capita in the world has increased and reached 113 kg / year in 2017 (IFD, 2018).

Production fell in the first half of 2017, with production in major milk exporters such as Europe, New Zealand, Australia and Argentina. There has been a limited increase in milk production in the world in recent years, but OECD and FAO estimates for 2027 increase milk production by 22%.

It is reported that the increase in milk production (80%) is due to developing countries such as India and Pakistan (OECD-FAO, 2018).

Due to Turkey's national income growth, consumption of animal products has increased in the period examined in the study. Turkey's demand for animal products consumption of urban population growth and urban population is expected to increase in the coming years. In a study conducted in 2012, the relationship between family income growth and food expenditures was investigated and a positive correlation was found between the variables (Ghafoor et al., 2012). In the study, very high positive correlations were found between the national income and the animal food demand except for the sheep and goat meat.

In this study, the higher number of positive correlations in the relationship between the population and the food consumption of animal origin may be attributed to the following reasons.

- Expectation of population increase leads to increase in animal production amount,
- Indicates that changes in population will be affected significantly by consumption,
- It shows that high population increases will affect high consumption values.

• Policies for population growth, as well as social and political policies, should be carried out with approaches that take into account population increases.

### 4. CONCLUSION

As a result, when people's income increases, it increases the consumption of animal-orginated food. As this trend continues towards a certain level, it is hoped that investments in livestock sector will positively affect this sector and achieve a positive return. Policies for population growth, as well as social and political policies, should be carried out with approaches that take into account population increases.

### REFERENCES

- Clarke AE, Friese C, Washburn R, 2018: Sitiational Analysis, Grounded Theory After Interpretive Turn, Second Edition, SAGE Publications, 2018.
- Delgado, C. 2005. Rising demand for meat and milk in developing countries: implications for grasslands-based livestock production. In Grassland: a global resource (ed.D. A. McGilloway), pp. 29–39. The Netherlands: Wageningen Academic Publishers.
- FAOSTAT, 2019. The Statistics Division of the Food and Agricultural Organization, Retrived in March 2019. http://faostat.fao.org.
- Food and Agricultural Organization (FAO), 2011 Mapping supply and demand for animal-source foods to 2030, FAO Animal Production and Health Working Paper No.2, E-ISBN 978-92-5-107041-3 (PDF).
- Ghafoor A., Arshad IA., Sheikh AW., 2012. Variation in consumption pattern and calories among different income groups. Sci.Int.(Lahore)., 24(3),317-321. ISSN 1013-5316.
- IDF (2018). Dairy Sustainability Outlook, Issue N°, Page 74, https://www.fil-idf.org/wpcontent/uploads/2018/12/IDF\_Dairy\_Sustainability\_Outlook\_2018\_web.pdf.

IDF (2016). The World Dairy Situation, Bulletin of the IDF No +85/2016;

- OECD Agriculture Statistics: OECD-FAO Agricultural Outlook (Edition 2018), OECD (2019), Meat consumption (indicator). doi: 10.1787/fa290fd0-en (Accessed on 06 April 2019), https://data.oecd.org/agroutput/meat-consumption.htm, OECD (2019).
- OECD-FAO (2018). Annual growth rates of per capita consumption for dairy products, Agricultural outlook 2018-2027, file:///C:/Users/HP/Desktop/i9166e\_Chapter7\_Dairy.pdf. Version 1 Last updated: 20-Jun-2018, http://oe.cd/disclaimer.

TURKSTAT (2019) Agricultural Structura (Production, Price, Value), Retrived in April 8, 2019, www.tuik.gov.tr

SPSS, 2013., SPSS for Windows, Version 22. SPSS Inc. New York.

Thornton PK., 2010. Livestock production: recent trends, future prospects, Phil. Trans. R. Soc. B., 365, 2853–2867, doi:10.1098/rstb.2010.0134.



## The Warburg Effect and its Importance in Cancer Treatment

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**Abstract:** Cancer is one of the most important causes of mortality, regardless of the level of development of countries around the world. According to data from the International Agency for Research on Cancer (IARC), there were expected 18.1 million new cases of cancer and 9.6 million deaths caused by cancer in 2018. In the early 1920's Otto Warburg has shown that cancer cells have a special metabolism, namely an increased uptake of glucose and secretion of lactic acid by cancer cells, even in the presence of oxygen. This aerobic fermentation is specific for cancer cells and it causes the cancer microenvironment to be acidic. The extracellular pH of normal tissues is 7.4 while cancer tissues have average 6.8 pH value. The local acidic microenvironment of a cancer cell plays an important role in cancer development. Communication between cancer cell and microenvironment has a critical role for its progression and metastasis. Basically, tumor cell acts like a seed and its microenvironment acts like a soil for growing and development period of cancer. This metabolic difference between cancer cells and normal cells is called "Warburg Effect". Changing the pH value of the microenvironment in cancer chemotherapy is essential for drug uptake into the cell and trap within endosomal and lysosomal compartments. Therefore, studies on pH sensitive nano-systems and drug molecules that changing the pH of microenvironment that can affect the absorption, release and distribution of the drug have been increased. In this article, Warburg Effect and its importance in cancer treatment reviewed.

Keywords: Warburg Effect, microenvironment, cancer treatment, pH, drug



## Solid State Fermentation Can Improve the Nutritional Composition of Apple Pomace

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**Abstract:** This study was conducted to investigate the effect of *Aspergillus niger* on the nutritional composition of apple pomace with solid state fermentation (SSF). Before fermentation, apple pomace was dried, milled and sterilized at 121 °C for 15 minutes. Then, *A. niger* strain (ATCC 200345) were inoculated apple pomace at 10<sup>4</sup> inoculation level. Before and after fermentation, dry matter (DM), ash, crude protein (CP), ether extract (EE), crude fiber (CF), neutral-detergent fiber (NDF), acid-detergent fiber (ADF) and acid-detergent lignin (ADL) content of apple pomace were determined before and after fermentation to evaluate the effect of the fermentation. SSF increased CP (P < 0.001) and ash (P < 0.05) content although decreased DM (P < 0.001), EE (P < 0.05) and nitrogen-free extract (P < 0.001), CF (P < 0.01), NDF (P < 0.001) and ADF (P < 0.001) in apple pomace. There was no effect (P > 0.05) of fermentation on ADL content. Obtained results showed that *A. niger* can be used to enrich the nutritional composition of apple pomace.

Keywords: Malus domestica, by-product, Aspergillus niger, nutritional enrichment

### **1. INTRODUCTION**

In recent years, agricultural by-products have received increased attention because of economic and environmental considerations. Apple pomace is a by-product in the apple juice industry. Although apple pomace is rich in mineral and carbohydrate, it has low crude protein (CP) and high crude fiber (CF) content (Beigh et al., 2015). This limits the use of apple pomace in animal nutrition. Ayhan et al. (2009) stated that apple pomace can be used in broiler chickens at 5% without detrimental effect on performance parameters but higher levels deteriorate feed conversion ratio. Similarly, Beigh et al. (2015) reported that apple pomace can be used at limited levels because of its low CP content.

Solid state fermentation (SSF) can improve nutritional composition and eliminate antinutritional factors in agricultural residues (Jazi et al., 2017; Altop et al., 2018). Generally, apple pomace was fermented by *Aspergillus niger* for organic acid (Shojaosadati and Babaeipour, 2002) and enzyme production (Dhillon et al., 2012). However, to our knowledge, there is not any study investigating the effect of *A. niger* on main nutritional components of apple pomace. Therefore, effect of *A. niger* SSF on the nutritional composition of apple pomace was investigated in this study.

## 2. MATERIAL AND METHOD

*A. niger* strain (ATCC 200345) obtained from American Type Culture Collection (ATCC) was cultured in Potato-Dextrose-Agar (PDA). Apple pomace was dried at 75 °C for 48 hours and milled to a size of 2 mm. The nutritional salt (glucose:urea:(NH4)2SO4:peptone: KH2PO4:MgSO4.7H2O=4:2:6:1:4:1) were added dried apple pomace after autoclave sterilization at 121 °C for 15 min. It was inoculated by 10<sup>4</sup> *A. niger* spores and incubated at 30 °C for 7 days. Samples were dried at room temperature approximately 30-35 °C for 6 days until samples reached 90% dry matter (DM). Fermentation was conducted with 3 replicates.

DM, ash, CP, ether extract (EE) and CF analyses were analyzed according to AOAC (2000). Neutral detergent fiber (NDF), acid detergent fiber (ADF) and acid detergent lignin (ADL) analyses were conducted according to Van Soest et al. (1991).

### **Statistical Analysis**

Experiments were conducted in triplicate and the results were given as means and pooled standard error of mean (SEM). Data were analyzed with Student t test (SPSS 21.0 Statistics). Results were considered significantly different at P < 0.05.

### 3. RESULTS AND DISCUSSION

Nutritional changes in apple pomace by *A. niger* SSF are given in Table 1. DM, EE and nitrogen-free extract (NFE) of apple pomace were decreased (P < 0.001, P < 0.05 and P < 0.05, respectively) by SSF. Fermented apple pomace (FAP) had higher CP (P < 0.001) and ash (P < 0.05) content than unfermented apple pomace (UAP). Moreover, CF, NDF and ADF of FAP were diminished (P < 0.05, P < 0.001 and P < 0.001, respectively) compared with UAP. However, fermentation did not affect (P > 0.05) the ADL content.

| Composition (%) | UAP   | FAP   | SEM   | Р   |
|-----------------|-------|-------|-------|-----|
| Dry Matter      | 90.12 | 82.53 | 1.699 | *** |
| Crude Protein   | 5.39  | 17.55 | 2.721 | *** |
| Ether Extract   | 2.78  | 2.39  | 0.100 | *   |
| Crude Ash       | 3.34  | 7.62  | 1.163 | *   |
| NFE             | 62.31 | 48.33 | 3.142 | *** |
| Crude Fiber     | 26.14 | 22.77 | 0.792 | **  |
| NDF             | 51.78 | 43.99 | 1.746 | *** |
| ADF             | 39.00 | 36.99 | 0.454 | *** |
| ADL             | 21.52 | 19.94 | 0.685 | NS  |

Table 1. Nutritional changes in apple pomace by Aspergillus niger solid-state fermentation

\* = P < 0.05, \*\* = P < 0.01, \*\*\* = P < 0.001, NS = not significant, UAP = unfermented apple pomace, FAP = fermented apple pomace, SEM: standard error of mean, NFE: nitrogen-free extract, NDF: neutral-detergent fiber; ADF: acid-detergent fiber; ADL: acid-detergent lignin.

Similar to the results of the present study, CP was increased in grape seed (Altop et al., 2018) and cherry kernel (Güngör et al., 2017) by fermentation. Mycelia or enzymes produced by *A. niger* may cause the increase in CP (Raimbault, 1998).

Fungi use firstly carbohydrates in the substrates as a carbon source (Papagianni, 2007). This decrease in NFE content can be attributed to using carbohydrates as a carbon source by *A. niger*. Similar results were reported by Güngör et al. (2017) and Altop et al. (2018).

Dhillon et al. (2012) showed that *A. niger* produces cellulase enzyme in apple pomace with SSF. In this study, CF decreased by fermentation may because of degradation in structural carbohydrates by cellulase enzymes of *A. niger*. Similar results were obtained in various studies (Jazi et al., 2017; Altop et al., 2018).

#### 4. CONCLUSION

*A. niger* can improve the nutritional composition of apple pomace, which makes it a feed source for animals. However, there is a lack of information on the effect of fermented apple pomace on ruminant and non-ruminant animals. Detailed studies should be conducted in future to recommend this product for animal production.

### REFERENCES

- Altop, A., Erener, G., Duru, M., and Isik, K., 2018. Effects of essential oils from *Liquidambar orientalis* Mill. leaves on growth performance, carcass and some organ traits, some blood metabolites and intestinal microbiota in broilers. British Poultry Science 59:121-127.
- AOAC. 2000. Official Methods of Analysis of AOAC International (17th Edition). AOAC International, USA.
- Ayhan, V., Duru, A. A., and Özkaya, S., 2009. Possibilities of using dried apple pomace in broiler chicken diets. Kafkas Univ Vet Fak Derg 15:669-672.
- Beigh, Y. A., Ganai, A., and Ahmad, H., 2015. Utilisation of Apple pomace as livestock feed: A review. Indian Journal of Small Ruminants (The) 21:165-179.
- Dhillon, G. S., Kaur, S., Brar, S. K., and Verma, M., 2012. Potential of apple pomace as a solid substrate for fungal cellulase and hemicellulase bioproduction through solid-state fermentation. Industrial Crops and Products 38:6-13.
- Güngör, E., Altop, A., Öztürk, E., and Erener, G., 2017. Nutritional changes of sour cherry (*Prunus cerasus*) kernel subjected to *Aspergillus niger* solid-state fermentation. Journal of Tekirdag Agricultural Faculty The Special Issue of 2 nd International Balkan Agriculture Congress:99-103.

- Jazi, V., Boldaji, F., Dastar, B., Hashemi, S., and Ashayerizadeh, A., 2017. Effects of fermented cottonseed meal on the growth performance, gastrointestinal microflora population and small intestinal morphology in broiler chickens. British Poultry Science 58:402-408.
- Papagianni, M., 2007. Advances in citric acid fermentation by Aspergillus niger: biochemical aspects, membrane transport and modeling. Biotechnology advances 25:244-263.
- Raimbault, M., 1998. General and microbiological aspects of solid substrate fermentation. Electronic Journal of Biotechnology 1:26-27.
- Shojaosadati, S., and Babaeipour, V., 2002. Citric acid production from apple pomace in multi-layer packed bed solid-state bioreactor. Process Biochemistry 37:909-914.
- Van Soest, P. v., Robertson, J., and Lewis, B., 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. Journal of Dairy Science 74:3583-3597.



## Compare of Lactation Milk Yield Characteristics and Somatic Cell Counts in Morkaraman and Tuj Sheep

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**Abstract:** In this study was investigated of lactation milk yield, lactation length, average daily milk yield and somatic cell counts (SCC) in Morkaraman and Tuj sheep. It focuses on the effects of some environmental factors like breed and age that are affecting these characters. It was determined that breed was not significant on lactation milk yield statistically but significant differences were observed between age groups on lactation milk, lactation length and daily milk yield (p<0.05), and had influence on lactation length very significantly (p<0.01). According to the SCC, breed and age had no effect on SCC.

Keywords: Morkaraman, Tuj, milk yield, somatic cell count

### **1. INTRODUCTION**

In terms of human nutrition, milk has the high nutritional value among animal products. (Teker 2016). Consumption of milk and the products obtained from the milk is very significant for healthy life. Milk consumption preferences depend on socioeconomic and sociocultural conditions (Besler and Ünal 2006; Yerlikaya ve Karagözlü 2008).

Hygiene conditions must be performed in order to keep the quality of milk. Depending on the developing technology and the increasing level of prosperity, the quality of milk and dairy products has a significant effect on consumption preferences. One of the evaluation criteria used to determine whether milk is healthy and has high quality is the SCC that milk contains (Manlongat et al. 1998; Yöney 1998). The objective of this study was focused on milk yield characteristics and SCC of Morkaraman and Tuj ewes.

## 2. MATERIAL AND METHOD

This study was carried out on 31 Morkaraman and 36 Tuj ewes at Ataturk University Faculty of Agriculture Research and Application Farm. The birth season, which started in April, continued until May. About 2 months old lambs were weaned and all of flock started to pasture by June. In order to determine lactation milk yield characteristics, control milking was started after weaning lambs and continued until the end of pasture in 15-day intervals. The lactation length was determined by drying off the sheep which were below 50 g milk yield per day. Lactation milk yield was calculated using Trapeze II (Dutch Method) used by the International Registry Commission (ICAR) (Yakan 2012). Milk was analyzed by DeLaval DCC for the measurement of SCC. The GLM procedure of the SPSS (2011) 20.0.0 package program was used for the analysis of variance of the milk yield characteristics and SCC. Differences between groups were determined by the Duncan Multiple Comparison test.

### **3. RESULTS AND DISCUSSION**

Lactation milk yield, lactation length, average daily milk yield and SCC of Morkaraman and Tuj breed are given in Table 1. It has been determined that breed is not statistically significant for lactation milk yield. The age of dam is significantly affected on lactation milk yield, average daily milk yield and lactation length. The similar results were reported by Küçük et al. (2000) and Abd Allah et al. (2011). It was observed that the values of lactation milk yield characteristics were below the average values reported in the literature. It was determined that lactation milk yield and average daily milk yield were the highest in Morkaraman; the length of lactation was highest in Tuj. The values for the lactation characteristics in the Morkaraman were found lower than the results Küçük et al. (2000), Özbey and Akcan (2000) and Kırmızıbayrak et al

(2005). In this study, lactation milk yield found for Tuj breed is lower than that of Kırmızıbayrak et al. (2005); is higher than Karaoglu (1997); the length of lactation is similar to Kırmızıbayrak et al. (2005) and Karaoglu (1997).

| Sources    | Lactation milk yield (kg) | Lactation length (day)     | Average daily milk yield (kg) | SCC<br>(cell*1000/ml) |
|------------|---------------------------|----------------------------|-------------------------------|-----------------------|
| Breed      | ns                        | ns                         | ns                            | ns                    |
| Morkaraman | $41.4\pm5.0$              | $128.2\pm4.9$              | $0.32\pm0.2$                  | $285.1\pm74.9$        |
| Tuj        | $40.2\pm3.6$              | $131.6\pm4.6$              | $0.31\pm0.2$                  | $327.8\pm89.2$        |
| Age        | **                        | **                         | *                             | ns                    |
| 2          | $30.9\pm3.9^{\rm c}$      | $119.8\pm5.9^{b}$          | $0.26\pm0.2^{b}$              | $385.7\pm158.5$       |
| 3          | $42.3\pm5.8^{abc}$        | $137.1\pm7.2^{\rm a}$      | $0.31\pm0.3^{ab}$             | $209.5\pm100.5$       |
| 4          | $38.5\pm5.2^{bc}$         | $126.9\pm7.4^{b}$          | $0.30\pm0.3^{ab}$             | $899.8\pm309.5$       |
| 5          | $53.0\pm5.2^{\rm a}$      | $146.6\pm6.9^{\mathrm{a}}$ | $0.36\pm0.3^{\rm a}$          | $440.1\pm239.4$       |
| ≥6         | $50.1\pm5.4^{ab}$         | $149.7\pm7.2^{\mathrm{a}}$ | $0.34\pm0.3^{ab}$             | $411.8\pm196.6$       |

Table 1. The least squares means and standard errors of lactation yield characteristics and SCC

\*: Significant (p < 0.05); \*\*: Very significant (p < 0.01); ns: Insignificant

SCC is an important marker for udder health. Tuj sheep had higher SCC than Morkaraman sheep, but the influence of breed on SCC was not significant. SCC increased with age. However, SCC was not significantly affected by age. The mean SCC obtained in the study was determined lower than the SCC values in milk reported by Konar et al. (1994) and Huntley et al. (2012), less than by Yağcı (2005), Othmane et al. (2002) and Baro et al. (1994).

### 4. CONCLUSION

There are limited researches of SCC in sheep and the studies concentrate on cow milk, so the quality standards have defined by limiting the SCC in the sheep milk. This case creates a gap in terms of quality standards for milk and dairy products obtained from sheep. This study has taken a step to fill this gap and it is suggested that new studies must be done to resolve this shortcoming.

#### REFERENCES

- Abd Allah M, Abass SF & Allam FM (2011). Factors affecting the milk yield and composition of Rahmani and Chios sheep. International Journal of Livestock Production, 1-2 (3): 024-030.
- Baro JA, Carriedo JA & San Primitivo F (1994). Genetic parameters of test day measures for somatic cell count, milk yield, and protein percentage of milking ewes. Journal of Dairy Sci. 77: 2658-2662.
- Besler H & Ünal S (2006). Ankara'da satılan sokak sütlerinin bazı vitaminler açısından değerlendirilmesi ve ev koşullarında uygulanan kaynatmanın süreye bağlı olarak vitaminlere olan etkisi. IV International Nutrition and Dietetic Congress, Ankara.
- Huntley SJ, Cooper S, Bradley AJ & Green LE (2012). A cohort study of the associations between udder conformation, milk somatic cell count, and lamb weight in suckler ewes. J. Dairy Sci. 95: 5001–5010.
- Karaoğlu M (1997). Atatürk Üniversitesi Tarım İşletmesi'nde yarı entansif şartlarda yetiştirilen Tuj koyunlarının bazı verim özellikleri bakımından gösterdikleri performans. PhD Thesis Ata. Uni., Graduate School of Natural and Applied Sciences, Erzurum.
- Kırmızıbayrak T, Aksoy AR, Saatçi M & Tilki M (2005). Tuj ve Morkaraman koyunların süt verimi ve meme özellikleri arasındaki ilişkiler. Journal of the Faculty of Veterinary Medicine, Kafkas University 11 (1): 11-15.
- Konar A, Güven M & Erginkaya Z (1994). Somatic cell counts and mastitis studies in the milks of goats and ewes of Türkiye. Proceeding of the symposium on somatic cells and milk of small ruminants, Bella Italy, 25-27 September, EAAP Publication 77: 327-334.
- Küçük M, Öztürk Y & Bayram D (2000). Yarı Entansif Sartlarda Hamdani, Karagül, ve Morkaraman Koyunlarının Süt Verimi Özelliklerinin Karsılastırılması. Journal of the Faculty of Veterinary Medicine, Yuzuncuyil University. 11 (1): 44-48.
- Manlongat N, Yang TJ, Hinckley LS, Bendel RB & Krider HM (1998). Physiologic-Chemoattractant-induced migration of polymorphonuclear leukocytes in milk. Abstract- Medline. 375-381.

- Othmane MH, De La Fuente LF, Carriedo JA & San Primitivo F (2002). Heritability and genetic correlations of test day milk yield and composition, individual laboratory cheese yield and somatic cell count for dairy ewes.
- Özbey O & Akcan A (2000). Akkaraman, Morkaraman ve İvesi Koyunlarının yarı-entansif şartlardaki verim performansı I. Döl Ve Süt Verimi Özellikleri. Journal of Veterinary Sciences. 16 (1): 109-120.
- SPSS (2011). SPSS for Windows release 11.5.0. SPSS Inc. Chicago, IL.
- Teker E (2016). Protein Oranı Standardize Edilmiş Yağsız İnek, Koyun ve Keçi Sütünün Asit Koagülasyon Özelliklerinin Karşılaştırılması. MS Thesis Ata. Uni., Graduate School of Natural and Applied Sciences, Şanlıurfa.
- Yağcı Pİ (2005). Koyunlarda klinik, Mikrobiyolojik ve Biyokimyasal Metotlar ile Subklinik Mastitislerin Saptanması. PhD Thesis, Health Sciences Institute of Ankara Üniversitesi, Ankara.
- Yakan A (2012). Koyun ve keçilerde süt verim kontrol yöntemleri ve laktasyon süt veriminin hesaplanması. AVKAE Dergisi 2,18-23.

Yerlikaya O & Karagözlü C (2008). Koyun sütünün beslenmedeki önemi ve Teknolojik özellikleri. Journal of Milk World, 14: 58-61.

Yöney Z (1998). Süt kimyası. Publication of Faculty of Agriculture, Ankara Uni. No: 175, Ankara.



## Microbial Activity Problems in Soils of Tea Orchards in Turkey

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Abstract: Soil microorganisms are reported to influence the physical, chemical and biological properties of soil either directly or indirectly. Microbial diversity has also been recommended as a biological indicator of soil quality. Tea orchard ecosystems are very particular compared with other agriculture ecosystems. Soil quality declined and tea growth was inhibited under long-term continuous cropping tea orchard systems. Also, long-term tea cultivation resulted in the accumulation of soil nutrition content. Cultivated tea bushes grow in close proximity to each other and the root exudates may accumulate in the rhizosphere from all sides, causing a marked inhibitory effect. On the other hand, the intensive use of chemical fertilizers creates a highly selective environment and adversely affects microbial diversity. Long-term heavy mineral fertilizer application, root exudates and leaf litter can result in the decline of pH and the accumulation of Al toxicity and antimicrobial substances. Therefore, the objective of this study is to compare the bacterial community in the non-rhizosphere and rhizosphere soils of tea bushes by different production systems and tea stand ages with continuously cropping histories for 8, 15, 30 and 40 years. Only soil adhering very closely to tea roots was collected for rhizosphere soil samples. The microbial population associated with tea soils, in terms of colony-forming units assessed by the platecount method, were carried out. According to the results of the study, the soil was strongly acidified following the research of tea, and soil pH generally continued to decrease with the increase in stand age and tea productivity. In the old tea gardens, microbial diversity and distribution of bacterium were found to be very low according to the mixed tea orchard systems with legumes, natural naturally grown tea plants in forests and forest soils. In young tea plants, the population of bacteria in the rhizosphere soil were found to be more than the non-rhizosphere area, due to age. Also, the number of bacteria in the tea rhizosphere soil reduced in long-term continuous cropping tea orchard systems. The intensive use of chemical fertilizers created a highly selective environment, reduced pH, and adversely affected microbial diversity and richness.

**Keywords:** Tea plant, rhizosphere soil, non-rhizosphere, bacterial population, tea orchard soil, microbial diversity, soil pH, stand age

### **1. INTRODUCTION**

Tea is an important plant for Turkey, and is grown in the eastern Black Sea acidic soils. Tea is grown as a monoculture, and also nitrogen, root exudates, root and leaf residues are the main determinants of tea rhizosphere. Soil acidity in tea bushes increases day by day. In particular, excessive nitrogen use increases costs and soil acidity, affects the rate of nitrification, causes water and environmental pollution in washing. The main source of high nitrogen pollution in the region is tea production areas and more than 40% of the applied nitrogen is washed and significantly increases nitrate pollution. On the other hand, the amount of nitrogen released increases more in rainy and sloping areas, which leads to eutrophication and reduces the quality of drinking water and biodiversity. Nitrogen fertilizers used are poor in terms of organic matter from day to day, decrease biological activity with increasing acidity and increase heavy metal uptake of tea, and also excess nitrogenous fertilizer leads to higher fiber yields in black tea. The application of chemical fertilizer can increase the crop yield but changes the soil properties and bacterial community structure and results in N loss from agricultural systems.

Soil organic matter may be a prime determinant of the reduction in microbial community diversity. The soil microbial community plays an important role in the decomposition of soil organic matter and elemental cycling (Wang et al., 2014). Microbial diversity has also been recommended as a biological indicator of soil quality. The fact that microbes are essential for the entire tea ecosystem since they perform numerous important functions like maintenance of biogeochemical cycles, promotion of plant growth; and inhibition of destructive tea pest and pathogens. Growth promoting substances are produced in large quantities by the beneficial microorganisms that influence indirectly on the overall morphology of the plants (Bhattacharyya et al 2015). Soil microbial community diversity is an important measure of sustainable land use and

sensitive to changes in the soil chemical properties. Also, soil microbial communities is extremely important in maintaining soil quality, function and the ecosystem sustainability (Nannipieri et al., 2003)

Soil microorganisms are reported to influence the physical, chemical and biological properties of soil either directly or indirectly (Saharan et al., 2011). Soil quality declined and tea growth was inhibited under long-term continuous cropping tea orchard systems (Li et al., 2016). Although the optimum pH range for tea growth is 4.5–6.0, the soil of tea gardens is often strongly acidified due to the high nitrogen fertilizer level applied, and the acidity in tea orchard soil gradually increases with the tea-orchard age (Liao, 1998; Shi et al., 1999; Yu et al., 2003, 2004; Xue et al., 2010). High levels of nitrogen fertilizer are applied, altering N cycling and microbial community structure (Tachibana et al., 1995). Soil becomes strongly acidified following the planting of tea and soil pH generally continues to decrease with the increase in stand age and tea productivity (Song and Liu 1990; Han et al. 2007; Wang et al. 2010).

Cultivated tea bushes grow in close proximity to each other and the root exudates may accumulate in the rhizosphere from all sides, causing a marked inhibitory effect (Pandey and Palni, 1996). Leaf litter may also contain antimicrobial substances which are slowly released upon decomposition. On the other hand, the intensive use of chemical fertilizers creates a highly selective environment and adversely affects microbial diversity (Gulati et al., 2011). Despite the increase in input costs in existing agriculture, additional fertilizer does not increase the yield, because of soil productivity and biological activity decrease and acidity increases. Tea orchard ecosystems are very particular compared with other agriculture ecosystems. Long-term heavy N application, root exudates and leaf litter can result in the decline of pH and the accumulation of Al toxicity and antimicrobial substances (Pandey and Palni 1996). So the consecutive monoculture problems are widespread in tea agriculture. This phenomenon is even more pronounced in the monocultural conditions of tea plantations, where the perennial nature of bushes also exerts a strong rhizosphere effect on the soil microflora (Pandey and Palni, 1996; Han et al., 2007). The aim of this study was to compare rhizospheric and non-rhizospheric bacterial populations and soil pH at different tea stand ages and land use.

### 2. MATERIALS AND METHODS

The soil samples were collected from four soil samples plots that were randomly chosen within an 8-year-old tea orchard, a 15-year-old tea orchard, a 30-year-old tea orchard, a 40-year-old tea orchard, a neighbouring non-rhizosphere soil, and an wild-growing tea in the forest (approximately 40-year-old), in Rize, Turkey. These tea orchards had similar environmental characteristics, such as altitude, slope position and slope aspect, and similar agronomic management. The annually applied compound fertilizer (25% N, 5% P and 10% K) was applied in April at the rate of 800 kg ha<sup>-1</sup> for each time.

Only soil adhering very closely to tea roots was collected for rhizosphere studies. Non-rhizosphere soil samples were collected well away from tea bushes. Soil samples under young tea bushes were collected only from the upper most depth (0-20 cm). Also, non-rhizosphere soil samples were collected only from the upper depth (0-20 cm) well away from tea bushes. Root adhering soil was collected from nursery plants, and soil from a depth of 5 cm to 25 cm was collected from adult plants. Since the rhizosphere soil of tea plants represents a special ecological niche, interactions between tea roots and surrounding soil result in a specific microbial community that plays an important role in the growth of tea plants. So in this study, only soil adhering very closely to tea roots (rhizosphere soil) was collected from the four tea orchards, and the Non-rhizosphere soil sample was collected near each tea orchard. In each tea bushes and its near non-rhizospheric zone, soil sample consisted of twelve randomly taken subsamples of rhizosphere soils of tea plants. From each sampling plot, 20 cores (5 cm in diameter  $\times$  20 cm in length) were taken and mixed. In total, 12 bulked soil samples were collected and transported to the laboratory.

Soil pH was measured by a digital pH meter after extracting with water (Wsoil:Vwater = 1:2.5). Investigations of the microbial population associated with tea soils, in terms of colony-forming units assessed by the plate-count method, were carried out at the active growth period. Rhizosphere and non-rhizosphere soil microbial counts were calculated on the basis of serial 10-fold dilutions according to the procedures described already (Çakmakçı et al., 2007). Soil samples were analysed to enumerate the colony forming units (CFU) of total bacteria following serial dilution and the pour plate method (Salle, 1973). From each sample 10 g of soil was aseptically weighed and transferred to an Erlenmeyer flask containing 100 ml of sterilised water, and shaken for 30 min at about 150 rpm. Immediately after shaking, each suspension was diluted 10-fold by pipetting 1-ml aliquots into 9 ml of sterilised water. A  $10^8$ -fold final dilution 3 replicate dishes were made.

The agar plates were incubated at 30 °C for 7 days. Data from each replicate were averaged for a soil sample and expressed as CFU per gram of oven-dried soil. entire year.

## 3. RESULTS AND DISCUSSION

### Results

The rhizospheric soil samples were collected from nursery (8-year-old trees), adult (15-year-old trees) and old (30 and 40-year-old trees) tea plants. Also, non-rhizosphere soil sample was collected near each tea orchards. Non-rhizosphere soil samples were collected well away from tea bushes. Rhizosphere microbial community diversity increased in 8-year-old tea orchards and then it starts to fall, in 30 to 40 years of tea, there were serious declines (Figure. The pH of rhizosphere soil was significantly increased from 1-year to 8-year tea orchards and then dropped in 15, 30 and 40-year-old tea orchards. Similar patterns of the bacterial community were observed between 1-year and 8-year tea orchards, which significantly differed from those of 30-40-year tea orchard (Figure 1-2). With the increasing year of tea planting, the microbial community structure in rhizosphere soils changed greatly, reflecting in the decline of microbial diversity. In young tea plants, the population of bacteria in the rhizosphere soil is greater than the non-rhizosphere area, due to age. Hovewer, the number of bacteria in the tea rhizosphere soil is reduced in long-term continuous cropping tea orchard systems.



Figure 1. Bacterial populations in the rhizosphere and non-rhizosphere soils in tea bushes of different stand ages.

Soil pH in tea orchards generally continues to decrease with the increase in cultivation age (Figure 2). In the old tea gardens, microbial populations and the rhizospheric soil pH were very low according to the young tea bushes and or 40-year-old wild-growing tea in the forest. Soil pH of rhizosphere soil samples ranged between 4.1 and 5.7 and ranged between 5.4 and 6.0 for non-rhizospheric soil samples. The pH of the tea orchard soils decreased gradually with increasing age after the establishment of an orchard. The pH in the 40-year-old tea orchard soil was even lower than that in the wild-growing tea in the forest (about 35-40-year-old self-growing tea). The population of bacteria in the rhizosphere soil in young tea bushes was greater than the non-rhizosphere area (Figure 2).



Figure 2. Rhizosphere and non-rhizosphere soil pH in tea bushes of different stand ages

### Discussion

In the absence of any change in agricultural practices in the region, excessive chemical fertilization can cause serious environmental and health problems in the region. The results revealed that soil pH and bacterial populations and quality declined and tea growth was inhibited under long-term continuous cropping tea orchard systems. While the abundance and activity of total bacterial populations in tea bushes of 30-year-old and 40-year-old decreased as the pH of the rhizosphere soil decreased, the bacterial community were found to be higher in the young tea bushes or 40-year-old wild-growing tea in the forest. As a matter of fact, in the 30-year-old and 40-year-old tea orchard, bacterial populations and soil pH in the rhizosphere was also lower than the non-rhizosphere zone. Xue et al. (2006) found that microbial community diversity increased from 5-year-old to 8-year-old tea orchards and then decreased from 50-year-old tea orchards. Li et al. (2016) pointed out that the continuous cropping of tea led to significant declines in soil pH, organic matter and soil nutrient contents. A key factor in determining soil microbial community structure is soil pH (Xue et al., 2010). Some researches demonstrated that soil pH had a strong influence on the microbial biomass and the growth activity of soil microbes (Han et al. 2007).

The rhizosphere of established tea bushes has several specific characteristics such as negative rhizospheric affect, lowering of soil pH, synergistic and antagonistic activities among microbial communities and dominance of certain species (Saharan and Nehra, 2011), which are strongly associated with this perennial tea plant. Also, tea roots, after attaining a certain age, may start secreting exudates that contain antimicrobial metabolites to which bacteria are the most susceptible group. Soil pH in tea orchards generally continues to decrease with the increase in cultivation age (Wang et al. 2010). In this study, continuous tea cropping and monoculture was found to result in low the rhizosphere soil pH as expected. Soil microorganisms have a profound effect on the conversion of other biogeochemical cycles such as N, P and S as well as various micronutrients and heavy metals (Morton and Edwards, 2005). This change in PH always affects the availability of plant nutrients and microbial processes in the soil (Plante, 2007; Pietri and Brookes, 2008) as well as the decomposition rate of organic compounds in soil (Leifeld et al., 2008; Yao et al., 2009). Some studies have shown that long-term tea cultivation resulted in the accumulation of soil nutrition content or in so-called nutrient sequestration (Xue et al. 2006; Zhao et al. 2012).

According to the results of the research, soil was strongly acidified following the planting of tea and the pH generally continued to decrease with the increase in stand age and tea productivity. However, in the old tea (40-year-old tea wild-growing tea in the forest) which grows spontaneously in the forest, the pH decrease was not very high. This is important in that the soil is acidified by the tea plant itself and by fertilization, in particular with ammonium sulfate and urea. Tea soils, on the other hand, also receive large amounts of tea litter and prunings, which contain high concentrations of aluminum and polyphenols. Al is beneficial for the growth and development of tea plants but may have a toxic effect on most soil microorganisms. Because of this, it has been reported that the intensity and duration of tea cultivation can have a strong impact on microbial biomass and community structure and functioning (Han et al. 2007). The 40-year-old or

even 30-year old tea orchard had the lowest pH, which may be due to a combination of high and unbalanced N fertilizer application and acidification by nitrification, plant growth, and acidic phytochemical inputs. Indiscriminate use of chemical fertilisers for long period of time reduced the soil health and tea productivity (Chakraborty et al., 2013).

Ammonia-oxidizing bacteria abundance decreased with decreasing pH, while pH was shown to be an important factor controlling the abundance of bacteria in the soil (Yao et al., 2011). In addition, long-term experiments have shown that ammonium fertilizer significantly reduces the abundance of denitrifiers (Hallin et al., 2009). Previous studies showed that N fertilizer application stimulates soil nitrification and ammonia oxidizer abundance (Di et al., 2009). Even the differences in litter quality and quantity may explain differences in nitrification rate and abundance of ammonia oxidizer (Xue et al., 2006, 2008). Tea gardens are usually grown in Turkey as a monoculture and receive considerable amounts of fertilization, root exudates, and leaf litter (Çakmakçı et al., 2010). Land use involving different plant species and soil characteristics may affect the soil bacterial community structure. Plant species have been reported to have a large effect on microbial community structure (O'Donnell et al., 200; Marschner et al., 2001). Because a number of organic compounds are secreted by the root of tea plants and continuously supplied by the litter, the amount and species of soil organic matters may be changed and therefore have a significant influence on the microbial community.

In order to tolerate low pH and microbial activity in tea bushes, it is necessary to use biological and organic manures. Previous studies have also demonstrated that microbial organic fertilizer has a significant effect on variation in the amount and composition of the microorganisms in the soil and that 3.0 tons of applied organic fertilizer per hectare significantly increases the number of bacteria and actinomycetes and diversity of soil microorganisms (Tu and Toan, 2017). Previous research has shown that organic manures improve soil health by increasing the organic carbon, organic acids, enzymatic activity, microbial populations and microbial biomass carbon content in the soil, and also adding organic manure to the soil enhances the biomass yields and chlorophyll content in case of tea leaf (Rajkonwar et al., 2016). On the other hand, three factors have been considered to induce the consecutive monoculture problems, including an imbalance of soil nutrients, the autotoxicity of root exudates and the shifts in the microbial community (Lin et al. 2012).

#### 4. CONCLUSION

In young tea plants, the population of bacteria and pH in the rhizosphere soil is greater than the non-rhizosphere area, due to age. Hovewer, the number of bacteria and pH in the tea rhizosphere soil is reduced in long-term continuous cropping tea orchard systems. Long-term tea cultivation resulted in the reduction of rhizosferic soil pH and microbial population.

### REFERENCES

- Bhattacharyya PN, Sarmah SR, Dutta P &,Tanti AJ (2015). Emergence in mapping microbial diversity in tea (*Camellia sinensis* (L.) O. Kuntze) soil of Assam, North-East India: A novel approach. European Journal of Biotechnology and Bioscience, 3 (12): 20-25.
- Chakraborty U, Chakraborty B, Chakraborty A, Sunar K & Dey P (2013). Plant growth promoting rhizobacteria mediated improvement of health status of tea plants. Indian J Biotechnol 2013; 12: 20-31.
- Çakmakçı R, Dönmez MF & Erdoğan Ü (2007). The effect of plant growth promoting rhizobacteria on barley seedling growth, nutrient uptake, some soil properties, and bacterial counts. Turkish Journal of Agriculture and Forestry, 31 (3):189-199.
- Çakmakçı R, Dönmez MF, Ertürk Y, Erat M, Haznedar A & Sekban R (2010). Diversity and metabolic potential of culturable bacteria from the rhizosphere of Turkish tea grown in acidic soils. Plant and Soil, 332 (1-2): 299-318.
- Di HJ, Cameron KC, Shen JP, Winefield CS, O'Callaghan M, Bowatte S & He JZ (2009). Nitrification driven by bacteria and not archaea in nitrogen-rich grassland soils. Natura Geoscience, 2:621–624.
- Gulati A, Swati Sood S, Rahi P, Thakur R, Chauhan S, & nee Chadha IC (2011). Diversity analysis of diazotrophic bacteria associated with the roots of tea (*Camellia sinensis* (L.) O. Kuntze). Journal of Microbiology and Biotechnology, 21(6): 545–555.
- Hallin, S., Jones, C.M., Schloter, M., *et al.*, 2009. Relationship between N-cycling communities and ecosystem functioning in a 50year-old fertilization experiment. The ISME Journal, 3(5):597-605.
- Han W, Kemmitt SJ & Brookes PC (2007) Soil microbial biomass and activity in Chinese tea gardens of varying stand age and productivity. Soil Biology and Biochemistry, 39: 1468–1478.

Leifeld J, Zimmermann M & Fuhere J (2008). Simulating decomposition of labile soil organic carbon: Effect of pH. Soil Biology and

Biochemistry, 40:2948-2951.

- Li YC, Li, Z, Li ZW, Jiang YH, Weng BQ & Lin WX (2016). Variations of rhizosphere bacterial communities in tea (*Camellia sinensis* L.) continuous cropping soil by high-throughput pyrosequencing approach. Journal of Applied Microbiology 121: 787–799.
- Liao W Y (1998). Acidification and prevention of tea garden soils in Chinese. Agro-environmental Protection, 17(4): 178–180.
- Lin WX, Chen T & Zhou MM (2012). New dimensions in agroecology. Chin J Eco-Agriculture 20, 253-264.
- Marschner P, Yang CH, Lieberei R & Crowley DE (2001). Soil and plant specific effects on bacterial community composition in the rhizosphere. Soil Biology and Biochemistry, 33:1437–1445.
- Morton SC & Edwards M (2005). Reduced phosphorus compounds in the environment. Critical Reviews in Environmental Science and Technology, 35:333-364.
- Nannipieri P, Ascher J, Ceccherini MT, Landi L, Pietramellara G & Renella G (2003) Microbial diversity and soil functions. European Journal of Soil Science, 54: 655- 670.
- O'Donnell AG, Seasman M, Macrae A, Waite I & Davies JT (2001). Plants and fertilizers as drivers of change in microbial community structure and function in soils. Plant and Soil, 232: 135–145.
- Pandey A & Palni LMS (1996). The rhizosphere effect of tea on soil microbes in a Himalayan monsoonal location. Biology and Fertility of Soils, 21: 131-137.
- Pietri JCA & Brookes PC (2008). Relationships between soil pH and microbial properties in a UK arable soil. Soil Biology and Biochemistry, 40:1856-1861.
- Rajkonwar U, Gogoi B & Ahmed, S (2016). Soil organic carbon, biological properties and plant biomass as affected by application of organic manures in tea. International Journal of Science and Nature, 7 (2): 444-449.
- Saharan BS & Nehra V (2011). Plant growth promoting rhizobacteria: a critical review. Life Sciences and Medicine Research, 21: 1-30.
- Salle AJ (1973). Laboratory Manual on Fundamental Principles of Bacteriology. McGraw-Hill Book Company, New York, USA.
- Shi J Q, Ding R X, Liu Y Z &, Sun Y H, 1999. Acidification of soil by urea and fallen tea leaves. Journal of Tea Science, 19(1): 7–12.
- Song M & Liu Y 1990: Effect of biogeochemical cycle in tea garden on the soil acidification. Journal of Tea Science, 10(2): 19-26.
- Tachibana N, Yoshikawa S & Ikeda K (1995). Influences of heavy application of nitrogen on soil acidification and root growth in tea fields. Jpn. J. Crop Sci. 64:516–522.
- Tu VN & Toan NV (2017). Effects of Microbial Organic Fertilizer and Mulch to Population and Bioactivity of Beneficial Microorganisms in Tea Soil in Phu Tho, Viet Nam. International Journal of Agricultural Technology 2017 Vol. 13(4): 469-484.
- Wang H, Xu RK, Wang N & Li XH (2010). Soil acidification of Alfisols as influenced by tea cultivation in eastern China. Pedosphere 20: 799–806.
- Wang H, Yang J-P, Yang, S-H, Yang, Z-C & Lv Y-M (2014). Effect of a 10 °C-elevated temperature under different water contents on the microbial community in a tea orchard soil. European Journal of Soil Biology, 62: 113-120.
- Xue D, Yao H Y & Huang C Y (2006). Microbial biomass, N mineralization and nitrification, enzyme activities, and microbial community diversity in tea orchard soils. Plant and Soil, 288: 319–331.
- Xue D, Yao HY, Ge DY & Huang CY (2008). Soil microbial community structure in diverse land use systems: a comparative study using BIOLOG, DGGE and PLFA analyses. Pedosphere 18:653–663.
- Xue, D, Huang, XD, Yao HY & Huang CY (2010). Effect of lime application on microbial community in acidic tea orchard soils in comparison with those in wasteland and forest soils. Journal of Environmental Sciences, 22(8):1253–1260.
- Yao H, Bowman D, Rufty T & Shi W (2009). Interaction between N fertilization, grass clipping addition and pH in turf ecosystems: Implications for soil enzyme activities and organic matter decomposition. Soil Biology and Biochemistry 41:1425-1432.

Yao, HY, Gao YM, Nicol GW, Campbell CD, Prosser JI, Zhang L, Han WY & Singh BK (2011). Links between ammonia oxidizer

community structure, abundance, and nitrification potential in acidic soils. Applied and Environmental Microbiology, 71:4618–4625.

- Yu S, He Z L, Chen G C & Huang C Y (2003). Soil chemical characteristics and their impacts on soil microflora in the root layer of tea plants with different cultivating ages. Acta Pedologica Sinica, 40(3): 433–439.
- Yu S, He Z L, Huang C Y, Chen G C & Zhu B L (2004). Soil acidification under tea bushes and its influence on the biological characteristics of a red soil. In: The Red Soils of China (Wilson M J, He Z L, eds.). Kluwer Academic Publishers, the Netherlands. 331–345.
- Zhao J, Wu X, Nie C, Wu T, Dai W, Liu H & Yang R (2012). Analysis of unculturable bacterial communities in tea orchard soils based on nested PCR-DGGE. World Journal of Microbiology and Biotechnology, 28, 1967–1979.


# Cold Hardiness Assessment of Four Grapevine Cultivars by using Tissue Browning Method in Erzincan

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Abstract: In this study was to compare bud death of four different grapevine cultivars ('Adesa' (*Vitis labrusca L.*), 'Hamburg Misketi', 'Sultani seedless' and 'Narince' (*Vitis vinifera L.*) after the winter cold of 2018-2019, and was to investigate the relationship between bud death ratio and soluble carbohydrates and water content of winter buds. Winter buds were sectioned through the tip with a razor blade and examined under a binocular microscope, checking for tissue discoloration after winter temperature (-19.8°C). Significant differences ( $p \le 0.01$ ) were determined at the low temperature tolerance of winter buds of four grape cultivars. Grape cultivars were classified as hardy ('Adesa'), moderately hardy ('Sultani seedless' and 'Narince') and least hardy ('Hamburg Misketi') based on the bud death ratio. The water content and soluble carbohydrates of winter buds were found to be highly consistent with the bud death ratio. There was a positive correlation between bud water cortent and bud death ratio for grape cultivars examined. However, for grape cultivars examined, there was a negative correlation between bud carbohydrate content and bud death ratio. Study findings showed that grape cultivars should be selected based on region conditions. In cold regions planting of sensitive cultivars such as 'Hamburg Misketi' should be avoided. The best method to increase frost tolerance is to use resistant grape cultivars such as 'Adesa', 'Sultani seedless' and 'Narince' in grapevine.

Keywords: Grapevine, frost tolerance, soluble carbohydrate content, water content

#### **1. INTRODUCTION**

For the past few years there has been mounting interest in the growing of different grape cultivars in Erzincan. However, cold hardiness is an essential characteristic that determines the suitability of grape cultivars for growing in a region where the air temperature is characterized by frosty winters (Kaya and Köse 2017; Köse end Kaya 2017; Buztepe et al., 2017; Kalkan et al., 2017; Rende et al., 2018; Kaya and Köse 2018; Köse et al., 2018; Kaya and Köse 2019). Despite the existence of a few cultivars of grapevine in Erzincan, only a single grape cultivar ('Karaerik') account for the commercial production. The majority of the vineyards are also located in cold climate areas where the winter minimal temperatures may plunge down -22°C, and such a situation at times could be causing serious damages to vineyards (Kalkan et al., 2017). The cold damage in the region is usually caused by low temperatures occurring in mid-winter. Indeed, when the temperature reached -21.7°C in 2016, a severe winter frost led to significant product loss, as compared to the preceding year. Therefore, it would be useful to evaluate the cold hardiness of a range of Turkey grape cultivars to assess their potential for this region.

Harsh winter temperatures followed by accurate viability assays provide useful information for growing of new grape cultivars. Accurate predictions help also in identifying main cultural and ecological influences on established vines (Barka and Audran 1997; Jones et al. 1999; Fennell 2004). The accuracy of cold hardiness estimation can be improved by combining results of different a few parameters frost tolerance evaluation ways applied concurrently. Among the ways of cold hardiness measurement, bud brown discoloration or straw colored and it's the relationship between winter bud water content and soluble carbohydrate analyses are the most common (Ershadi et al., 2016; Rende et al., 2018) and cold hardiness estimated by these analyses correlate well (Kaya and Köse 2017).

The reduction in winter bud water content is effectively in association with increases in cold hardiness and is thought to contribute to an increased ability to supercooling, a mechanism that allows tissue water to remain in the liquid phase below sub-freezing temperatures (Salzman et al., 1996; Bourne and Moore 1991; Grant and Dami 2015). For this reason, it is feasible to use alterations in these coherent parameters in winter buds exposed to the harsh min-winter temperatures as indicators to determine cold hardiness in different grape cultivars. Besides winter bud water content and soluble

carbohydrate metabolism has been determined to influence the susceptibility of grape winter buds to low temperatures (Grant and Dami 2015; Rende et al., 2018). Published studies have reported the close relationship between carbohydrate metabolisms and cold hardiness in grapevine winter buds (Wample and Bary 1992; Hamman et al., 1996), but the exact nature of the changes in soluble carbohydrate metabolism in response to cold injury has not been fully understood for 'Adesa', 'Hamburg Misketi' 'Sultani seedless' and 'Narince' grape cultivars in this region. Therefore, the aim of this work was (1) to determine the bud tolerance of four grape cultivars thought to be of value to the Turkey and Erzincan viticulture industry, using a visual expression of winter bud tissue necrosis as described by Stergios and Howell (1977), (2) to evaluate the relationship between primer bud death ratio and soluble carbohydrates and water content of winter buds.

#### 2. MATERIAL AND METHOD

#### **Plant Material**

The experiment was performed on winter buds from six-year-old 'Adesa' (*Vitis labrusca L.*), 'Hamburg Misketi', 'Sultani seedless' and 'Narince' (*Vitis vinifera* L.) vines grown at the Erzincan. One year old shoot segments containing nodes (nearest to the base) at positions 1 through 4 (except for 'Sultani seedless' - 9 through 14 nodes) were collected from unpruned vines on 10 March 2019 for the frost damage and other analysis experiments. These canes were divided into three groups and each group was placed in a sealed plastic bag. Then, samples were brought to the laboratory immediately and stored at room temperature (22°C) one day (24h).

#### Evaluation of Winter Bud Damage: (Bud or Tissue Browning Assay)

Following the one day anticipation, cross sectioning were made on winter buds with a single-edged razor blade to confirm cold injury manifested by tissue necrosis or browning. These winter buds were determined under a stereomicroscope (Model BX50F4; Olympus, Japan), checking for necrosis. The winter buds that appeared green and bright was considered alive and that appearing straw colored and brown discoloration was considered dead (Stergio and Howell 1977). The proportion of injured winter buds for each grape cultivars were calculated based on the number of deaths of winter buds (Odneal 1984).

#### **Evaluation of Bud Soluble Carbohydrates**

Soluble carbohydrates of winter buds were measured according to the anthrone method (Yemm and Willis 1954). Winter buds taken from nodes (the first 4 for three cultivars, except for 'Sultani seedless' - 9 through 14 nodes) of one year old shoot segments were oven-dried at 80°C for 72h. Then winter bud samples were ground in a grinder and were kept in lightless condition until soluble carbohydrate analysis. Soluble carbohydrates of winter buds were extracted three times from 0.2g of milled dry bud tissues with 5 mL of 80% ethanol and centrifuged for 20 min at 4000 gn. Two mL of 0.2% anthrone reagent was added to  $50\mu$ L of the ethanolic extract. The mixture in glass tubes was incubated in a water bath at 90°C for 15 min and then the tubes were rapidly cooled. Absorbance of the extract was read at 620 nm using a Thermo Fisher Multiskan Sky (model-51119700DP) Microplate Spectrophotometer (Olympus, Japan). The concentration of soluble carbohydrates of winter buds was eventually calculated by using a standard glucose curve and expressed as mg g<sup>-1</sup> dry weight (DW).

#### **Evaluation of Bud Water Content**

Winter buds taken from the first 4 nodes (except for 'Sultani seedless' - 9 through 14) of an old shoots were excised and were weighed immediately with precision scales and after placing in an oven for 3 days at 70°C. Winter bud water content was determined as percent of fresh tissue weight by using the following formula:

Bud water content = [(fresh weight - dry weight) / fresh weight] x 100

#### **Statistical Analysis**

All statistical analysis was performed using SPSS, version 25 (SPSS Inc., Chicago, IL, USA). The relationship between bud death ratio and soluble carbohydrates, bud water content was also tested using correlation analysis. Data for the measurement parameters and cultivar comparison were tested using one-way analysis of variance. Duncan's new multiple range test was analyzed for post-hoc comparison of mean values.

#### 3. RESULTS AND DISCUSSION

The daily minimum air temperatures (°C) recorded in the vineyard are shown from Oct. 2018 through Mar. 2019 in Figure 1, and the lowest air temperature recorded in the region was -19.8°C on Jan. 11. In the present study, when buds of four grape cultivars were tested on 10 Mar., significant differences ( $p \le 0.01$ ) were found between frost tolerances of grape cultivars. According to method Odneal, (1984), between these four cultivars, 'Hamburg Misketi' was sensitive, 'Sultani seedless', 'Narince' were semi-sensitive and 'Adesa' was hardy (Table 1). The bud browning or necrosis test seems to be an effective method to estimate cold injury of grapevine winter buds. Indeed, researchers working on cold injury of dormant buds have noted that discoloration or necrosis of bud structure following a low temperature injury is a good indication of bud death, as a result, there will not be a successful produce for grapevine damaged by exposure to low winter temperatures (Odneal 1984; Köse and Güleryüz, 2009; Ershadi et al. 2016; Kalkan et al. 2017; Köse and Kaya 2017). In addition, many investigators confirmed that the *Vitis labrusca* cultivars, which belongs to the *Vitis* species, are more tolerant to low temperatures than the *Vitis vinifera* cultivars (Fennell, 2004; Kaya and Köse, 2018). The fact that 'Adesa' grape cultivar (*Vitis labrusca* L.) is more tolerant to low temperatures compared to other grape cultivars (*Vitis vinifera* L.) confirms these results.



Figure 1. Minimum daily air temperatures recorded in Erzincan Horticultural Research Institute Weather Station (Oct. 2018- Marc. 2019).

In grapevine dormant buds it has been determined that soluble carbohydrates rise in the fall to winter when dormant buds cold acclimate and reduce in the spring when deacclimation occurs (Hamman et al., 1996; Jones et al., 1999; Grant and Dami 2015; Kaya and Köse 2017; Rende et al., 2018). In this study, significant interactions were found indicating bud deaths were dependent on soluble carbohydrate concentration and grape cultivars. The buds of the 'Adesa' had generally more soluble carbohydrates compared to the other grape cultivars, and 'Adesa' had more cold hardy than other grape cultivars, which had the least cold hardiness (Table 1). Additionally, the relationship between soluble carbohydrate and bud death. Most correlation coefficients between bud death and the soluble carbohydrates were different from zero at  $p \le 0.01$  and so considered biologically significant (Table 2). The correlation between soluble carbohydrates and bud death obtained in the present study are, similar to those reported in the studies (Kaya and Köse, 2017; Rende at al., 2018).

The water content of winter buds was higher by 25.51% for the 'Hamburg Misketi' grape cultivar, and by 19.03, 19.63, 14.94% for 'Adesa', 'Sultani seedless' and 'Narince' grape cultivars, respectively (Table 1). During late summer cold acclimation starts and water content of dormant buds decrease with induction of frost tolerance (Salzman et al., 1996; Kaya and Köse, 2017). If there is much water in the dormant bud cells when the temperature gets low it will form ice crystals in the cells which destroys the cell membranes.

**Table 1.** Comparison of Adesa (*Vitis labrusca* L.), Hamburg Misketi, Sultani seedless and Narince (*Vitis vinifera* L.) grape cultivars according to water (%), soluble carbohydrate contents (mg.g<sup>-1</sup> DW) and bud death rate (%)

| Grape cultivars  | Bud death ratio          | Soluble carbohydrates    | Water                   |
|------------------|--------------------------|--------------------------|-------------------------|
| Adesa            | 35.00±1.03 <sup>a*</sup> | 16.12±0.47 <sup>a*</sup> | $14.94{\pm}0.37^{a^*}$  |
| Hamburg Misketi  | 54.00±1.12°              | 12.26±0.72°              | 25.51±0.12°             |
| Sultani seedless | 43.00±1.18 <sup>b</sup>  | 13.36±0.41 <sup>b</sup>  | 19.03±0.42 <sup>b</sup> |
| Narince          | 45.00±1.23 <sup>b</sup>  | 13.85±0.08 <sup>b</sup>  | 19.63±0.74 <sup>b</sup> |

\*Means within columns followed by the same letter do differ significantly at  $p \le 0.01$  by Duncan's new multiple range test.

The water of dormant buds should therefore decrease to a minimum level during the mid-winter period (Zabadal et al., 2007). Indeed, the 'Adesa' grape cultivar, which has less bud water content, showed more tolerances to low temperatures compared to other grape cultivars (Table 1). On the other hand, dormant buds showed a positive relationship between bud death ratio and bud tissue water content during mid-winter temperatures (Wolpert and Howell, 1986). According to the results of the analysis, a positive relationship was seen among grape cultivars, with 'Adesa' grape cultivar showing greater tolerance and less dormant bud water (Table 1, 2). Indeed, a close relationship between bud water content and cold tolerance has been determined in sweet cherry (*Prunus avium* L.) (Kadir and Proebsting 1994), peach (*Prunus persica* L.) (Quamme 1983), apple (*Malus domestica* Borkh.) (Kang et al. 1998), rhododendron (*Rhododendron* spp.) (Ishikawa and Sakai 1981) and blueberry (*Vaccinium australe* Small) (Bittenbender and Howell 1975).

**Table 2.** Correlation of Adesa (*Vitis labrusca* L.), Hamburg Misketi, Sultani seedless and Narince (*Vitis vinifera* L.) grape cultivars according to water, soluble carbohydrate contents and bud death rate

| Grape cultivars        | Bud death rate           | Soluble carbohydrates | Water  |
|------------------------|--------------------------|-----------------------|--------|
| Adesa                  | 1.000                    | -0.904*               | 0.940* |
| Hamburg Miskeni        | 1.000                    | -0.829*               | 0.996* |
| Sultani seedless       | 1.000                    | -0.882*               | 0.787* |
| Narince                | 1.000                    | -0.619*               | 0.990* |
| *Completion is signifi | agent at the 0.01 larval |                       |        |

\*Correlation is significant at the 0.01 level

#### 4. CONCLUSION

In the research presented in this paper, the winter bud death ratios were evaluated according to the tissue browning method, and the pattern of cold injury changes in four different grapevine cultivars was consistent with winter bud tissue water content and soluble carbohydrates. 'Adesa' grape cultivar was found to have the highest frost tolerance among the four studied grapevine cultivars. Although not as much as 'Adesa', 'Sultani seedless' and 'Narince' were also very tolerant of winter low temperature. Therefore, these grapevine cultivars could be considered for breeding programs in the future and are perfect to be used in regions under high risk of winter cold.

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#### REFERENCES

- Barka EA, Audran JC (1997). Response of 'Champenoise' grapevine to low temperature: changes of shoot and bud proline concentrations in response to low temperatures and correlations with freezing tolerance. Hort Sci 72:557–582
- Bittenbender HC, Howell GS (1975). Predictive environmental and phonological components of flower bud hardiness in high bush blueberry. Hort Sci 10: 409-411

Bourne TF, Moore JN (1991). Cold hardiness in grape cultivar development. Fruit Var J 45:26-28

Buztepe A, Kose C, Kaya O (2017). Evaluation of cold tolerance of dormant buds according to position using thermal analysis in Karaerik (*V. vinifera* L.) grape. International Journal of Research and Review 4(10):38-45

- Ershadi A, Karimi R, Mahdeí KN (2016). Freezing tolerance and its relationship with soluble carbohydrates, proline and water content in 12 grapevine cultivars. Acta Physiol Plant 38(1):1–10
- Fennell A (2004). Freezing tolerance and injury in grapevines. J Crop Improv, 10(1-2), 201-235
- Grant TN, Dami IE (2015). Physiological and biochemical seasonal changes in Vitis genotypes with contrasting freezing tolerance. Am J Enol Vitic. 66:195-203
- Hamman RA, Dami IE, Walsh TM, Stushnoff C (1996). Seasonal carbohydrate changes and cold hardiness of Chardonnay and Riesling grapevines. *Am J Enol Vitic*, 47(1), 31-36
- Ishikawa M, Sakai A, (1981). Freezing avoidance mechanisms by supercooling in some Rhododendron flower buds with reference to water relations. *Plant Cell Physiol.* 22: 953-967.
- Jones KS, Paroschy J, McKersie BD, Bowley SR (1999). Carbohydrate composition and freezing tolerance of canes and buds in *Vitis vinifera*. J Plant Physiol 155:101–106
- Kadir SA, Proebsting EL (1994). Screening sweet cherry selections for dormant floral bud hardiness. Hort. Sci. 29: 104-106.
- Kalkan NN, Kaya Ö, Karadoğan B, Köse C (2017). Determination of cold damage and lipid peroxidation levels of Karaerik (*Vitis vinifera* L.) grape cultivar having different trunk height in winter buds. *Alinteri J of Agric Sci* (in Turkey) 32(1):11-17
- Kang SK, Motosugi H, Yonemori K, Sugiura A (1998). Supercooling characteristics of some deciduous fruit trees as related to water movement within the bud. J. Hort. Sci. & Biotechnol. 73: 165-172.
- Kaya Ö, Köse C (2017). Determination of resistance to low temperatures of winter buds on lateral shoot present in Karaerik (*Vitis vinifera* L.) grape cultivar. *Acta Physiol Plant* 39(9), 209
- Kaya Ö, Köse C (2018). Effects of Cold Damage on Grapevine. Yyu. J. Agr. Sci. (in Turkey) 28(2):241-253
- Kaya Ö, Köse C (2019). Detection of death point in grapevine dormant buds by comparing thermal analysis and differential thermal analysis methods. *Turkish Journal of Agricultural and Natural Sciences*. 6(1): 118-124
- Köse C, Güleryüz M (2009). Frost damage in dormant buds of Karaerik grapevine grown at Uzumlu province of Erzincan during the winter of 2007–2008. Atatürk Univ J Fac Agric 40(1):55–60
- Köse C, Kaya Ö (2017). Determination of resistance to low temperatures of winter buds according to position in Karaerik (*V. vinifera* L.) grape cultivar. *Int J Sci Res Publ* 7(4):4-5
- Köse, C., Güneş, A., Kaya, Ö., Kıtır, N., Turan, M., & Şahin, F. (2018). Freeze Injure and Antioxidant Enzyme Activity of Grapevine (Vitis Vinifera) Under Bio-Boron Fertilizer Applications. *Erwerbs-Obstbau*, 60(1), 3-10.
- Odneal MB (1984). Cold hardiness of grapes. State fruit experiment station, College of Health and Applied Sciences, Missouri State University. Bulletin, (41)
- Quamme HA (1983) Relationship of air temperature to water content and supercooling of overwintering peach flower buds. J. Amer. Soc. Hort. Sci. 108: 697-701.
- Rende M, Kose C, Kaya O (2018). An assessment of the relation between cold-hardiness and biochemical contents of winter buds of grapevine cv.'Karaerik' in acclimation-hardening-deacclimation phases. *Mitteilungen Klosterneuburg*, Rebe und Wein, Obstbau und Früchteverwertung, 68(2):67-81
- Salzman RA, Bressan RA, Hasegawa PM, Ashworth EN, Bordelon BP (1996). Programmed accumulation of LEA-like proteins during desiccation and cold acclimation of overwintering grape buds. *Plant Cell Environ* 19:713–720
- Stergios BG, Howell GS (1977) Effect of site on cold acclimation and deacclimation of Concord grapevines. Amer J Enol Vitict 28:43–48
- Wample RL, Bary A (1992). Harvest date as a factor in carbohydrate storage and cold hardiness of Cabernet Sauvignon grapevines. J. Amer. Soc. Hort. Sci. 117: 32-36.
- Wolpert JA, Howell GS (1986). Cold acclimation of Concord grapevines III. Relationship between cold hardiness, tissue water content. Vitis, 25(1), 151-159
- Yemm EW, Willis AJ (1954). The estimation of carbohydrates in plant extracts by anthrone. Biochem J 57:508-514

Zabadal TJ, Dami IE, Goffinet MC, Martinson TE, Chien ML (2007) Winter injury to grapevines and methods of protection. Michigan State University Publications on Grape Production, Michigan, pp 36–37



# Seasonal Varation of Stable Isotope Ratios in Cow Milk in Lithuania

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Abstract: Dairy products are of particular interest as a group of foods that play an important role in feeding the population. The composition of milk is fundamentally dependent on the feeding of the cows, and thereby on a particular environment. To better understand the amount of variation in  $\delta^{18}$ O,  $\delta^{13}$ C, and  $\delta^{15}$ N values in the milk from the same area, we measured stable isotope ratios in cow milk water, artesian water, and precipitation ( $\delta^{18}$ O) as well as in bulk milk samples ( $\delta^{13}$ C and  $\delta^{15}$ N) collected in 2014–2016. Different water and food sources were available during the winter (artesian water only and dry grass) and summer (artesian water and fresh grass), and spring and autumn seasons reflected transitional periods. Oxygen stable isotope ratios in milk water were relatively lower in winter and transitional seasons and higher in summer, showing the dependence on the main water source.  $\delta^{13}$ C values reflected particular food sources. This study shows the applicability of the stable isotope ratios in the same area. These results could be valuable for other studies on geographical origin determination of dairy products.

Keywords: Dairy products, Stable isotope ratio, Carbon, Nitrogen, Oxygen, Isotope ratio mass spectrometry.



# The Effect of Body Weight on Laying Performance of Laying Hens

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Abstract: This study was conducted to investigate the effects of body weight on performance parameters of Lohmann white laying hens. At the beginning of the experiment, hens were weighed up individually and groups were classified as light (<1500 gr), medium (1500-1750 gr) and heavy (>1750gr) as to standard deviation of mean. The study was conducted over a period of 36 wks. Feed and water were offered ad libitum in the experiment. Considering the findings of performance traits differences among the groups were found significant. This study emphasized that body weight affected the laying performance except for FCR and cracked egg rate. Egg weight was determined to be lower in the group with light body weight than those of medium and heavy groups. However, egg production in the light group was higher (P<0.01) than that of heavy group. Feed intake was found to be higher in medium and heavy groups than that of light group.

Keywords: Laying hen, body weight, performance parameters

#### **1. INTRODUCTION**

Human nutrition is a key factor in maintaining a healthy life. Egg is one of the most important food with its high protein value, rich in vitamins and minerals and low in calorie. In poultry farming, egg production, feed consumption and feed conversion ratio are the most important criteria for economic breeding (Akbay, 1980; Nazlıgül et al., 2001). In the flock, the heavier weight hens produce larger eggs than the other hens. However, the body weight of hens is not desirable to rise above a certain limit. Selection studies for increasing body weight decreased egg production (Özen 1986). For the development of egg production, it is necessary to determine the nature of the relationship between body weight and egg production (Oke et al, 2004; Lacin et al., 2008). This study was conducted to investigate the effects of body weight on performance parameters of Lohmann white laying hens.

#### 2. MATERIAL AND METHOD

Total of 288 Lohmann white layers, 44 wks of age, were allocated randomly three groups, each formed 24 replicate cages as subgroups, comprising of four hens. At the beginning of experiment, hens were weighed up individually and groups were classified to be light (<1500 gr), medium (1500-1750 gr) and heavy (>1750gr) as to standard deviation of mean. The study was conducted over a period of 36 wks. Feed and water were offered ad libitum in the experiment. Feed intake and egg production were recorded daily; egg weight was measured at 2-weeks intervals and body weight was measured monthly. Before determination of egg weight, a sample of 12 eggs from each experimental group was stored for 24 hrs in room temperature. Feed conversion ratio (FCR) was expressed as kilogram of feed consumed per kilogram of egg produced. The data were analyzed using a General Linear Model procedure (SPSS, 1996) for a completely randomized experimental design. Differences between means were determined by Duncan's multiple range test.

#### 3. RESULTS AND DISCUSSION

Tablo 1 shows laying performance parameters of hens in the various body weight groups (light, medium and heavy). During the experimental period, feed consumption was significantly different among the three groups. Feed consumption of hens also increased linearly as the body weight increased. Similarly to present study, some authors reported that there was a significant relationship between body weight and feed consumption (Lacin et al., 2008; Balcioğlu et al., 2005).

|           | BW (kg) | FC (g)  | EP (%) | FCR  | EW (g) | CE (%) |
|-----------|---------|---------|--------|------|--------|--------|
| Light     | 1572.5c | 115.2 b | 82.8 a | 1.73 | 66.9c  | 0.95   |
| Medium    | 1710.5b | 116.6ab | 79.8 b | 1.73 | 67.5b  | 0.96   |
| Heavy     | 1846.5a | 118.1a  | 76.7 c | 1.72 | 68.8a  | 1.15   |
| SEM       | 6.72    | 0.67    | 0.54   | 0.01 | 0.19   | 0.10   |
| Anova     |         |         |        |      |        |        |
| Group (G) | **      | **      | **     | ns   | **     | ns     |
| Time (T)  | **      | **      | **     | **   | **     | **     |
| G x T     | ns      | ns      | ns     | ns   | ns     | ns     |

Table 1. The Effects of Different Body Weight on Laying Performance Parameters of Laying Hens

BW=body weight ; FC= feed consumption (g/d); EP= egg production FCR= feed conversion ratio (kg feed consumed per kg egg produced) ; EW=egg weight ; CE=cracked egg (%)

Egg production was affected by body weight (P<0.01). The highest egg production was obtained from the light group. Egg production decreases as the body weight increases in the groups. Although heavy body weight group had higher egg weight than the other groups, egg production decreased (P<0.01) in this group. The effect of body weight on egg weight was found to be significant in light, medium and heavy body weight groups (P<0.01). It was found that the heavy group had a higher egg weight than the other groups (P<0.01). Lacin et al., (2008) and Summers and Leason (1983) reported similar results. The FCR and cracked egg rate were not affected by body weight. These results were similar to Harms ve ark., 1982; Leeson ve Summers, 1987).

#### 4. CONCLUSION

As a result, it was found that there was a positive relationship between the egg weight and the body weight of the hens and the heavier eggs were obtained from heavier chickens. The relationship between egg production and egg weight was negative and egg weight decreased as the egg production increased.

#### REFERENCES

- Akbay R (1980). Leghorn tavuklarında düşük canlı ağırlık yönünde yapılan seleksiyonun diğer özellikler üzerindeki etkileri VII. Bilim Kongresi, 29 Eylül-3 Ekim 1980, İstanbul, 193-209.
- Balcıoğlu MS, Karabağ K, Yolcu HI & Şahin E (2005). Japon bıldırcınlarında canlı ağırlığa göre iki yönlü seleksiyonun eşeysel olgunluk yaşı ve bazı verim özellikleri üzerine etkisi. GAP. IV. Tarım Kongresi, 21–23 Eylül, Şanlıurfa.
- Harms RH, Costa PT & Miles RD (1982). Daily feed intake and performance of laying hens grouped according to their body weight. Poultry Sci., 61: 1021-1024.
- Lacin E, Yildiz A, Esenbuga N & Macit M (2008). Effects of differences in the initial body weight of groups on laying performance and egg quality parameters of Lohmann laying hens. Czech J. Anim. Sci. 53:466-471.
- Leeson S & Summers JD (1987). Effect of immature body weight on laying performance. Poultry Sci., 66: 1924-1928.
- Nazlıgül A, Türkyılmaz K & Bardakçıoğlu EH (2001). Japon bıldırcınlarında (coturnix coturnix japonica) bazı verim ve yumurta kalite özellikleri üzerine bir araştırma. Türk J. Vet. Anim. Sci., 25: 1007-1013.
- Özen N (1986). Tavukçuluk, Yetiştirme, Islah, Besleme, Hastalıklar, Et ve Yumurta Teknolojisi. Ondokuz Mayıs Üniversitesi Ziraat Fakültesi, Samsun.
- Oke UK, Herbert U & Wachukwu ENN (2004). Association between body weight and some egg production traits in the guinea fowl (*Numida meleagris galeata*. Pallas). Livestock Research for Rural Development 16 (9).
- SPSS (1996). IBM SPSS Statistics Premium N.C., U.S.A

Summer JD & Leeson S (1983). Factors influencing egg size. Poultry Science 62: 1155–1159.







# **ORAL PRESENTATION**

# Potential Uses of Paddy By-Products

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Abstract: When paddy cultivation area in Turkey is analysed in terms of production and productivity values, it can be stated that 35 thousand tons production and 6.04 tons productivity were gained in 50 thousand ha cultivation area in 2000 while in 2018 940 thousand tons production and 7.82 ton/ha productivity were gained in 120 thousand ha cultivation area. Generally, the grain of paddy are commonly considered as white rice in the world. However, the straws remaining from the harvesting of pieces of broken rice, bran, photocell, raw piece and glumes generated during processing of paddy into rice are by products of paddy. After the paddy is harvested, alternatives methods are found to mix the botanic components remaining on the surface into the farm and to utilise the stems somewhere else except for farm. The alternatives can be using these by products in manufacturing collecting as bales, using as structure materials, in drainage and pipeline constructions, as backfill material in plastic pattern dust and plywood glue, for the purpose of security and safe, for mulching, in compost, in animal breeding, as base plate in poultry husbandry, in seedling cultivation environment, erosion, benches, forest control and soil stabilization, in hand crafts, for manufacturing packaging material, paddy stem ash, hydrolysis and synthesis gas, alcohol, methane and fibre wood. Rice flour, rice starch, rice wine and beer are produced from broken ice. Raw piece and photocell remaining which are other by products of paddy are used as additive in forages. Paddy husk are used as construction material, in wall construction and manufacture of fire-resistant brick, animal breeding and as base material in poultry husbandry, in production of compost, electricity, as absorbing material in watery solutions in order to separate ions, as potential filter in order to separate arsenic in water and in order to absorb oil on hard surfaces. The ash of paddy husk is used for the production of glass, cement, ceramic and other products. Finally, rice bran is used as forage and the oil generated from the bran is used in sectors of food, cosmetology and pharmacy.

Keywords: Paddy, straw, husk, broken, bran

#### **1. INTRODUCTION**

Paddy has the largest production quantity worldwide and has the second largest cultivated lands after wheat. About onethird of world population is feed with rice. In Turkey, paddy has the forth place for production quantity and cultivated lands (TÜİK, 2019). Paddy is originated from Southeastern Asia. Paddy farming started about 300 B.C. in Europe. Paddy was introduced into Turkish agriculture 500 years ago. Paddy farming has started later in American continent.

In Turkey, paddy farming was practiced over 59 thousand decares land area in 1961 with an annual production of 233 thousand tons and unit area yield of 396 kg/da. In 2018, paddy farming was practiced over 120 thousand hectares land area with an annual production of 940 thousand tons and a unit area yield of 782 kg/da. Paddy farming is practiced almost in all regions of Turkey. Marmora region has the first place in paddy farming with respect to cultivated lands and production quantities. In Turkey, about 68% of paddy farming lands are located in Thrace-Marmora region, 36.5% is located in Black Sea region and 6.5% is located in the other regions (TÜİK, 2019).

A rice grain is composed of about 20% hull (husk), 1-2% embryo, 8-9% bran and around 70% amylaceous endosperm. With the process of 100 kg paddy, about 55-60 kg full grain, 7-8 kg broken grain, 8-10 kg bran, 2-3 kg photocell (sorter) return (red-lined, defected, chalky grains), 2 kg rough grain and 15-20 kg husk are obtained (Sürek, 2002). Paddy grain is composed of a caryopsis and an inner husk tightly surrounding caryopsis, pericarp and two interlocked external husks. These husks are not detached from the grain after threshing and the grain is then called as paddy. The non-husked and non-pearled rice in caryopsis form is called as cargo.

The grain product obtained through partial or fully removal of embryo, pericarp and aleurone with different milling operations following appropriate husking of the fruit of cultural *Oryza sativa* L. Species is called as rice. With the process

of paddy into rice, about 70% blanched rice is obtained. Of this quantity, 50-60% is composed of unbroken full grains and 10-20% is composed of broken grains. Rice grains contain 77-79% starch, 7-8% protein, 12-14% moisture and 1% ash and oil (Sürek, 2002).

#### 2. PADDY BY-PRODUCTS

Paddy grains are largely used as white rice grains worldwide. However, there are several by-products of paddy processing into rice including broken grains, bran, photocell-returns, rough grain and husks, postharvest and threshing straws.

#### Straws

Straws are the plant residues left over the field after harvest and threshing of paddy. Quantity of paddy straw vary with the cutting height and paddy cultivar and generally vary between 200 - 400 kg/da. Considering 120 thousand ha paddy farming lands of Turkey in the year 2013, it is quite easy to estimate that there will be high quantities of paddy straw left over the field after the harvest. Such high quantities significantly hinder soil preparation activities. Since there are quite low demands in markets for paddy straws and it is quite costly to incorporate these residues into the soils, majority of the growers tend to eliminate paddy straws through burning. However, burning may result in nitrogen, phosphorus ( $\approx 25\%$ ), potassium ( $\approx 20\%$ ) and sulphur (5-60%) losses in soils. There are various alternative methods for elimination of paddy straws instead of burning (Sürek, 2007; Sezer and Mut, 2004).

**Baling:** Bales are made through compressing paddy straws without panicles with a baling machine. In case of finding a proper market, baling may offer a more economical means of use instead of burning paddy straws (Sonia, 2004).

**Construction materials:** It is possible to use paddy straws in masonry construction and manufacture of fire-resistant bricks. Paddy straws are quite resistant to fire and can be used as a light-weight and well-insulative construction material. The buildings constructed with paddy straw bales are resistant to earthquakes and they have quite-well heat insulation (Figure 1).

**Construction of drainage and pipe lines:** Since paddy straws are resistant to rotting, light-weight and durable, they are used in construction of drainage and pipe lines (Figure 1).

For safety and security purposes: Paddy straw bales are used in safety barriers in motor and horse races.



Figure 1. Potential use of paddy straws as construction material, construction of drainage and pile lines and for erosion control

**Erosion prevention, slope protection, forest management and soil stabilization:** Paddy straws are used in erosion prevention over the fields and paddy lands. They are also chopped into small pieces, placed into cloth sacks and used for stabilization of berms along the road sides. They can also be used for mulching over forest lands of cool climates to control the trees (Figure 1).

**Mulching;** Mulching is to cover especially around the trees and bushlands with dead organic materials. It may provide significant contributions for soil moisture preservation and erosion control. Mulch is then incorporated into the soil as a composting material. (Figure 2) (Işık et al., 2011).

**Composting:** Decomposition of paddy straws to preserve nutrient and organic material content is defined as composting. Composting can be performed in open fields or in controlled closed facilities. Livestock manure and similar materials can be supplemented into paddy straws to improve rate of decomposition (Figure 2) (Khan et al., 1981).

**Packing material:** Paddy straws with their resistance to squeezing and stretching can be used as a well-packaging material. However, increasing use of synthetic materials reduced the use of paddy straw-like natural materials as packaging material. But increasing prices of petroleum products tended people towards back to natural materials (Figure 2) (Işık et al., 2011).



Figure 2. Potential use of paddy straws as mulching, composting and packaging material

**Livestock bedding and poultry litter material:** In livestock operations of several countries, paddy straws are used as bedding material since they can easily absorb manure and act as a well conveyor. In poultry industry, paddy straws are used as litter material and then turned into material with a quite high fertilizer value (Çam and Sarıca, 1996).

**Seedling growth media:** Paddy straw bales can be used in preservation of cucumber, tomato and flower seedlings. Bales are wetted and powder nitrogen impregnated or enriched with various fertilizer and then used as seedling growth media (Şeniz, 1998).

**Mushroom growth medium:** Paddy straws are used as an important compost material in mushroom production activities of the countries with well-developed mushroom industry. In mushroom production, compost made of paddy straw bales together with livestock urea and manure to supply nitrogen requirement is used (Erkel, 2000).

**Plastic molding powder and filling materials in plywood adhesives:** Paddy straws were smashed and passed through 80 mesh sieve and then can be used as plastic molding powder. This material has equivalent characteristics with the other materials. Very fine straw powder can also be used as filling material in resin adhesives of plywood.

**Alcohol and fiber wood manufacture:** It is possible to produce alcohol and high-density wood with paddy straw. From 4000 tons of paddy straw, it is possible to produce 3000 tons wood and 80000 gallons alcohol. Success of this process can be improved through well-marketing and widespread of domestic use (Houston, 1972).

**Handcrafts:** Especially in western countries, paddy straws are commonly used in manufacture of string, sack, basket and hat-like materials, decorative and textile items (Figure 3).



Figure 3. Potential use of paddy straw in handcrafts

**Paddy straw ash:** In some western countries, paddy straw ash is used for medical purposes, in hair washing, fertilizations because of high silica content and to improve concrete compressive strength (Figure 4) (Kıvrak and Başyiğit, 2012).



#### Figure 4. Potential use of paddy straw ash

**Hydrolysis and synthesized gas:** With the hydrolysis of paddy straw, pentose, glucose and lignin is obtained. Lignin is used in paper manufacture. It is also possible to convert paddy straw into synthesis gases though couple pyrolytic processes. Cellulose and lignin in paddy straw can also be converted into sugar through hydrolysis. It is possible to obtain glucose as much as half of cellulose. Following the acquisition of glucose, ethyl alcohol can be obtained through sugar fermentation and alcohol distillation. Ethyl alcohol is used as biodiesel (Figure 5) (Houston, 1972).



Figure 5. Paper manufacture from paddy straws

**Methane production through anaerobic digestion:** Methane is produced from paddy straws through anaerobic (without oxygen) digestion. Paddy straws are converted into fatty acids and low-molecular weight alcohols with acid forming bacteria. Resultant product is then converted into methane, carbon dioxide and other gases with the methane bacteria (Huang, 2000).

#### **Broken Grains**

The rice grains smaller than three-fourth of a full grain are called as broken rice. These broken grains are used in manufacture of rice flour, starch, wine, vinegar, beer and pet food (Sezer et al., 2007; Figure 6).

**Rice flour:** Rice grains are dry-milled to produce rice flour. This process is also performed as wet-milling. Wet-milled flour has finer texture and generally used in cakes and desserts. But in wet-milling, starch may be degraded and thus water-soluble vitamins, minerals, free amino acids, sugar and some oils may be lost (Ylimaki et al., 1988).

Rice flour does not contain gluten, thus can reliably be used in celiac patient diets. Rice flour is thus commonly used in manufacture of gluten-free products. Rice flour is also used in infant formulas, breads for celiac patients, puddings and facial masks.

**Rice starch:** Rice starch is produced from broken rice grains through wet-milling in 0.3-0.5% solution to separate protein. For this purpose, broken grains are kept in alkaline solution for 24 hours and wet-milled later on (Juliano and Hicks, 1990). Rice starch is especially used in diets of celiac patients.

**Rice wine:** Broken rice grains are fermented with yeast and fungi. Initially a sweet product is obtained, then this product is converted into alcohol after fermentation. Rice wines are marketed under different brands in different countries (Figure 6; Juliano and Hicks, 1990).

**Rice vinegar:** Rice vinegar is a traditional product of China and Japan. It is produced through fermentation of rice starch. Acetic acid formation is obtained through mixture of seed vinegar and rice vinegar. Fermentation may take about 1-3 months. Rice vinegar has about 4-5% total acidity composed of acetic acid, lactic acid and succinic acid (Figure 6; Shimoji et al., 2004).

**Rice beer:** Broken grains are used in beer production in the USA and Japan through mixing with corn meal. Rice is preferred instead of corn because of low oil content. For this purpose, broken grains are obtained through regular rice processing. However, in Japan, broken grains are obtained from processing of cargo rice. Broken grains should not contain bran. In this process, the rice with low gelatinization and low amylose content is used since the starch of rice with medium gelatinization and medium amylose content is quite resistant to liquification (Juliano and Hicks, 1990) (Figure 6).



Figure 6. Manufacture of wine, vinegar and beer from broken grains

#### **Unripe Grain**

Unripe grains indicate immature green and greenish color rice grains. They are generally used in livestock feed additives.

#### Photocell (sorter) Return

**Chalky rice:** Chalky rice indicates the rice grains with pale milky white color for various reasons and the rice grains with a chalky appearance.

**Red-lined rice:** It indicates the rice grains with a thin or thick red line from end to end which is not specific to species. These grains are generally used in livestock feed additives.

#### **Rice Husk**

The external cover or coating of the rice grain is called as husk (hull). Rice husk is produced as the by-product of rice processing industry of Asia and Pacific countries with intensive paddy farming. Husk constitutes about 20% of production quantity (Doberman and Fairhurst, 2002).

**Construction material:** Rice husk is used in masonry construction and manufacture of fire-resistant bricks. It can provide a light-weight construction material with quite well insulation. Rice husk is also mixed with mud and used in manufacture of masonry bricks. It is also used as filling material in limestone manufacture, between wall and ceiling materials in construction industry (Doberman and Fairhurst, 2002; Figure 7).

**Rice husk ash:** Rice husk contains about 20% silica. The slica obtained from from rice husk ash are used in the following industries:

Manufacture of glass, cement, ceramic and other products; as filling material in various polymers especially in plastics; as filling material in cement manufacture to improve firming power; as low-cost bio-absorbent to remove heavy metals and dies from the waste waters; to hold oxidation products in frying oils; as fertilizer.



Figure 7. Potential use of rice husk as construction material

**Livestock bedding and poultry litter material:** In livestock operations of several countries, rice husk is used as bedding material since it can easily absorb manure and act as a well conveyor. In poultry industry, rice husk is used as a litter material and then turned into material with a quite high fertilizer value (Çam and Sarıca, 1996).

**Composting:** Decomposition of rice husk to preserve nutrient and organic material content is defined as composting. Composting can be performed in open fields or in controlled closed facilities. Livestock manure and similar materials can be supplemented into rice husk to improve rate of decomposition (Khan et al., 1981).

**Electric power generation:** There are some countries generating electricity from rice husk. For instance, annually 1,5 million tons rice produced in Australia and some of resultant husk is used in electric power generation. The energy equivalent of world annual rice production is about 1 billion barrels of oil (Ahiduzzaman, 2007).

**Other uses:** Rice husk is used as absorbent to remove metal ions from aqueous solutions, as a filter material to remove arsenic from water and again absorbent to remove oil from hard surfaces (Kalapathy and Proctor, Srivastava et al., 2006) (Işık et al., 2011).

#### **Rice Bran**

Bran is the upper layer of brown rice. It is a by-product of rice processing industry and it includes germ of the grain and majority of aleurone layer. It also includes a little husk and starchy endosperm (Salunkhe et al., 1992).

The primary protein fractions of rice bran are composed of albumin, globulin and free amino acids.

Greater albumin and globulin levels may enrich rice bran in lysine, methionine, phenylalanine and tryptophan-like essential amino acids.

Starch, cellulose and pentosanes are the most important carbohydrates in rice bran.

Rice husk includes high quantity of group B vitamins, but it is quite poor in vitamins A and C (Orthoefer, 1996b). Therefore, it is mostly used as animal feed.

Rice lipids are primarily composed of triglycerides and to less extent phospholipids, glucolipids and vexes (Salunkhe et al., 1992).

Non-saponified substances of raw rice bran oil contain high quantities of tocopherol, tocotrienol and  $\gamma$ -oryzanol-like antioxidant compounds and small quantities of sterols, triterpene alcohols, 4-methyl-sterol and polar compounds (Lee et al., 2004; Cicero and Derose, 2005; Juliano et al., 2005). The crude oil obtained after oil extraction from rice bran is refined and oil-extracted rice bran is then used as animal feed. Total oil content of rice depends on variety, ripening level, growing conditions and oil extraction method (press or solvent extraction) (Table 1).

Oryzanol is non-saponified fraction of rice bran oil and it was named after the first discovery in rice bran oil (Oryza sativa L.). Among the oryzanols,  $\gamma$ -oryzanol is the most appropriate one and extracted crude rice bran oil contains about 1-3% (Cicero and Derose, 2005; Juliano et al., 2005).

| Fatty acids                | Percent (%) |
|----------------------------|-------------|
| C14:0 Myristic Acid        | 0.6         |
| C16:0 Palmitic Acid        | 21.5        |
| C18:0 Stearic Acid         | 2.9         |
| C18:1 Oleic Acid           | 38.4        |
| C18:2 Linoleic Acid        | 34.4        |
| C18:3 Alpha Linolenic Acid | 2.2         |

Table 1. Fatty acid composition of bran oil

Oryzanol was reported to reduce cholesterol absorption, vascular occlusion, prevent thrombocyte accumulation, have antioxidative activity, reduce serum cholesterol, be effective against ageing like tocopherols, inhibit tumor formation, be

effective in menopause disorders, treatment of nerve system disorders, prevent dandruff formation in hairs and inching in body (Tuncel, 2016).

Rice bran oil is used in cosmetics, pharmaceuticals and animal feeding. The oil is especially used as frying oil since the taste and oxidative stability of rice bran oil are better than the other vegetable oils. Winterized rice bran oil is also quite available as salad sauce and for mayonnaise production. Stearin separated through winterization process is used in margarine and shortening production.

#### 3. CONCLUSION AND RECOMMENDATIONS

Turkey is quite lucky for paddy production potential, available lands, water resources, climate, yield, income level and domestic market.

Considering the economic value of paddy plants, the problem is not the main product (rice), but the by-products (straw and husk).

Although there are several potential uses of paddy straws, they are not widely used because of some technical limitations, cost of removal from the field, problems experienced in supply and storage. Therefore, stubble burning is preferred and such burnings generate significant environmental pollution.

Since the markets in which paddy straw biomass could be used commercially are quite limited, incorporation of paddy straws or stubbles into soil through appropriate methods come into prominence. Prospective studies on this issue will enlighten the way of industrial practices and contribute the development of the sector. Idle paddy straws can be squeezed into solid fuels and be used as an alternative of coal. Therefore, potential uses of paddy straws in different sectors should be supported, projects on these issues should be given priority, researches on these issues should be promoted in universities.

Besides using as white rice, paddy by-products (straw, bran, husk, oil and etc.) have also significant uses. Together with increasing paddy farming lands, resultant by-products should urgently be used for different purposes. Such uses will have great potentials for different industries.

The demand for rice bran oil is increasing worldwide. Although acquisition and processing are quite hard as compared to the other vegetable oils, oil bran oil is quite rich in nutrients and thus can reliably be used as cooking oil and then may reduce import of vegetable oils which is the second greatest group of products imported after oil.

The other paddy by-product, rice husk, has also a wide range of use and such a potential should also be turned into commercial products urgently.

#### REFERENCES

- Ahiduzzaman, M (2007). "Rice Husk Energy Technologies in Bangladesh" Agricul- tural Engineering International: the CIGR Ejournal. Invited Overview No. 1. Vol. IX. January, 2007.
- Akbar, M., and Yabuno, T., 1974, Breeding for saline resistant varieties of rice. II. Comparative performance of some rice varieties to salinity during early development stages, Jpn. J. Breed. 25(4):176-181.
- Cicero, A.F.G. Derose, 2005. Rice bran and its main components:potentiao role in the management of coronary risk factors. Current Topics in Nutraceutical Resaarch, 3 (11): 29-46.
- Çam, M.A., ve M. Sarıca, 1996. Broiler Üretiminde Farklı Altlık Materyallerinin Performansa ve Altlık Özelliklerine Etkileri. O.M.Ü. Zir. Fak. Derg., 11(2): 149-161.
- De Datta, S.K., 1981, Principles and Practise of Rice Production, The International Rice Research Institute, Los Banos, The Philippines.

Dobermann A, Fairhurst T.H, 2002. Rice straw managment, Better Crops International, Vol.16, Special Supplement, P. 7-11.

Emeklier, H.Y., 1993, Sıcak İklim Tahılları (Tahıllar II), Ankara Üniv. Ziraat Fak. Ders kitabı, Yayın No: 1296, Y. Ders kitabı:372, 118 s., Ankara.

Erkel İclal, Kültür Mantarı Yetiştiriciliği, Kocaoluk Yayınevi, İstanbul, 2000.

- Glaszmann, J.C., ve Arraudeau, M., 1986, Rice plant type variation: "Japonica" "Javanica" relationships, rice genetics newsletters, Volume 3. IRRI, P.O. Box 933, Manila, Philippines.
- Houston, D. F., "Rice Chemistry and Technology", American Association of Cereal Chemistry, St. Paul, Minnesota, 1972
- Huang, H. T. "Science and civilisation in China. Volume 6. Biology and biological technology. Part V: fermentations and food science." (2000).
- IRRI, 2011, International Trade in Rice: Recent Developments and Prospects. www.İrri.Org/Publications/Wrrc/Wrrcpdf/Session17-01.Pdf, Erişim Tarihi; Kasım 2011
- Işık, D., Mennan, H., Kaya Altop E., Macit, İ., 2011. Çeltik Saplarının Meyve Bahçelerindeki Yabancı Otların Kontrolünde Malç Olarak Kullanılabilirliğinin Araştırılması. Türkiye IV. Bitki Koruma Kongresi 28-30 Haziran 2011, Kahramanmaraş, s164.
- Juliano, B. O., and Hicks, P. A. 1990. Rice functional properties and rice food products (Review). Food Rev. Int. 12:71-103.
- Juliano, B.O. 1993. Rice in human nutrition. FAO Food and Nutrition Series, No: 26. The International Rice Research Institute (IRRI). Laguna, and Food and Agriculture Organization of the United Nations (FAO), Rome.
- Juliano, O.B. (Ed). 1994. Rice: Chemistry and Technology (2nd ed., pp. 17-160). St. Paul, EUA: The American Association og Cereal chemist' Inc., 647-680.
- Kaddah, M.T., 1963, Salinity effects on growth of rice at the seedling and inflorescence stages of development, Soil Sci. 96:105-111.
- Kalapathy, U., A. Proctor, 2000. A new method for free fatty acid reduction in frying oils using silicate flms produced from rice hull ash. J. Am. Oil Chem. Soc.7 (6), 593-598.
- Khan, S.M., A. G. Kausar, and M. A. Ali. 1981. Yield performance of different strains of oyster mushrooms (Pleurotus spp.) on paddy straw in Pakistan. Mushroom Science XI Proceeding of the Eleventh International Scientific Congress on the Cultivation of edible Fungi, 675-678, Australia.
- Kıvrak, S.O. ve Başyiğit C., 2012, Çeltik Sapı Külü Elde Edilmesinde Puzolaniklik Açısından Uygun Yakma Sıcaklığı Araştırılması, Süleyman Demirel Universitesi, International Technologic Science, Vol. 4, No 3, December 2012, pp. 94-101
- Kruining, Van; Hendricus A., "Rice hull ash filter", US Patent 4765545, 1988
- Kün, E., 1997, Sıcak İklim Tahılları. Ank. Üniv. Ziraat Fak. Yay. No: 1452 Ders Kitabı No: 432, Ankara.
- Lee, J-W., S-W. Lee, M-W. Kim, C. Rhee, L-H. Kim, K-W. Lee, 2004. Beneficial effect of the unsaponiflable matter from rice bran and oxidative stress in vitro comparede with a-tocopherol. Journal of the Science of Food and Agriculture. 85 (3): 493-498.
- Orthoefer, F.T. 1996a. Rice Bran Oil, p:393. In Y.H. Hui, ed. Bailey's Industrial Oil and Fat Products. Vol:2; Edible Oil and Fat Products: Oil Seeds. John Wiley and Sons, Inc., New York.4
- Orthoefer, F.T. 1996b. Rice bran oil: Healthy lipid source. Food Technology, 50, 62-64.
- Pearson, G.A., Ayers, A.D., Eberhard, D.L., 1966, Relative salt tolerance of rice during germination and early seedling development, Soil Sci. 102:151-156.
- Sezer, İ. ve Z. Mut, "Samsun İlinde Çeltik Tarımının Durumu ve Üretimi Artırmak İçin Öneriler", Gazi Osman Paşa Üniversitesi, Ziraat Fakültesi Dergisi, Cilt:21 (1) S:57-66 (2004).
- Sezer, İ., H. Akay, Z. Mut, A. Gülümser ve M. Şahin, "Çeltik Yetiştiriciliğinde Fideleme Ekim Yöntemi", Uluslararası Katılımlı I. Ali Numan Kıraç Tarım Kongresi ve Fuarı, 27-30 Nisan, Eskişehir, Cilt III, Bildiriler Kitabı, S:2309- 2316 (2011).
- Sezer, İ., H. Akay, Z. Mut, F. Öner ve M. Şahin, "Karadeniz Bölgesinde Çeltik Tarımı ve Sorunları", Uluslararası Katılımlı I. Ali Numan Kıraç Tarım Kongresi ve Fuarı, 27-30 Nisan, Eskişehir, Cilt III, Bildiriler Kitabı, S:2317-2325 (2011).
- Sezer, İ.,Z. Mut ve F. Öner, "Çeltikte (Oryzasativa L.) Kırıklı Randımana Etkili Faktörler", Türkiye VII. Tarla Bitkileri Kongresi, 25-27 Haziran, Erzurum, Bildiriler Kitabı, S:226-230 (2007).
- Shaikh, N.P., J.A. Brady, 1999. Rice bran oil and the subsequent isolation of other micronutrient. Paper presented at AOCS Annual Meeting and Expo, Orlando, Florida, May 9-12.

Shimoji, Yumi; Kohno, Hiroyuki; Nanda, Kumiko; Nishikawa, Yasushi; Ohigashi, Hajime; Uenakai, Kazuo; Tanaka, Takuji (2004). "Extract of Kurosu, a Vinegar from Unpolished Rice, Inhibits Azoxymethane-Induced Colon Carcinogenesis in Male F344 Rats". Nutrition and Cancer 49 (2): 170–3. doi:10.1207/s15327914nc4902\_8. PMID 15489210.

Sonia Schloemann. 2004. Winter Mulch for Strawberries. New York Berry News, Vol.3 NO.11, November 16, 2004.

- Srivastava, V.C., I.D. Mall, I.M. Mishra, 2006. Characterization of mesoporous rice husk ash (RHA) and adsorption kinetics of metal ions from aqueous solution onto RHA.J. Hazard. Mater. 134 (1-3), 257-267.
- Sürek, H. (2007). Ülkemizde çeltik anızı sorunu ve çözüm önerileri, Hasad Bitkisel Üretim, Yıl:23, Sayı: 270, Sayfa: 58-61.
- Sürek, H., 2002. Çeltik Tarımı. Hasad Yayıncılık, İstanbul.
- Şeniz, V. 1998. Sebzecilikte Fide Yetiştiriciliği ve Sorunları. TAV Tarımsal Araştırmaları Destekleme ve Geliştirme Vakfı Yayın No: 35. Yalova
- TMO, 2011, Hububat Raporu 2010, Toprak mahsulleri ofisi genel müdürlüğü, ANKARA Ağustos 2011 sf: 91-99
- TÜİK 2014. http://www.tuik.gov.tr/ Veri Tabanı internet Adresinden, Erişim Tarihi Aralık 2014.
- Tuncel, N. Y. 2016. Gama-orizanol. Academic Food Journal/Akademik GIDA, 14(4).
- Uygur, F.N., Lanını, W.T., 2006. Organik Tarımda Yabancı Ot Kontrol Yöntemleri ve Yan Etkileri. Türkiye 3. Organik Tarım Sempozyumu, Program ve Bildiri Özetleri. 1-4 Kasım 2006, Yalova
- Vaughan, L.K., Ottis, B.V., Prazak-Havey, A.M., Bormans, C.A., Sneller, C., Chandler, J.M., Park, W.D., 2001, Is all red rice found in commercial rice really (Oryza sativa) Rice Abstracts, Vol. 24, No. 4.
- Vergara, B.S., ve Visperas, R.M., 1988, Low temperature problems in growing in rice, Lecture Paper for GEU Training Program, 1987, IRRI, Manila, Philippines, Sf:12
- Ylimaki, G., Z.J. Hawrysh, R.T. Hardin and B.R. Thomson. 1988. Application of response surface methodology to the development of rice flour yeast breads: objective measurements. Journal of Food Science 53(6): 1800-1805.



# Effects of Different Densities of *Tetranychus urticae* (Acari: Tetranychidae) on Chlorophyll Content of *Phaseolus vulgaris* L. cv. 'barbunia' (Fabaceae) Leaves

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**Abstract:** The two spotted spider mite, *Tetranychus urticae* (Trombidiformes: Tetranychidae) is one of the main pests of agricultural crops including most vegetables. This mite damages its host plants while feeding damaging parenchyma cells and removing chlorophyll and other cell contents causing economic injury. In this experiment, the effects of different *T. urticae* densities on the chlorophyll content of pinto bean leaves were investigated. The experiment was carried out on abaxial surfaces of leaf discs with a diameter of 2 cm. To determine the effects of different mite densities, 5, 10, 15, 20 and 25 *T. urticae* adult females were separately placed on per leaf disc. Each treatment was replicated 6 times. All tests were carried out in the laboratory at  $25^{\circ}C \pm 1$ ,  $65 \pm 5\%$  R.H. and a photoperiod of 16L:8D (Light: Dark). Five days after infestation, the chlorophyll content was measured from 4 different points on each disc using a portable leaf chlorophyll meter (SPAD-502, Konica Minolta, Inc., Japan). The means of chlorophyll contents were 50.725, 48.171, 46.86, 45.10, 39.683 and 39.717 at infestation levels of 0 (control), 5, 10 15, 20 and 25 mites per disc, respectively. The results showed that the leaf chlorophyll content decreased with increasing mite density. However, the mean chlorophyll contents at infestation levels of 20 and 25 mites per disc were statistically lower than the control discs after exposure for 5 days.

Keywords: Chlorophyll, feeding damage, injury assessment, mite density, twospotted spider mite



# Survival Time and Size of *Neoseiulus californicus* (Mesostigmata: Phytoseiidae) Reared on *Polyphagotarsonemus latus* and *Tetranychus urticae*

# (Trombidiformes: Tarsonemidae: Tetranychidae)

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Abstract: Polyphagotarsonemus latus (Banks) (Trombidiformes: Tarsonemidae) and Tetranychus urticae Koch (Trombidiformes: Tetranychidae) are important pest mite species in many agricultural crops worldwide. Neoseiulus californicus (McGregor) (Mesostigmata: Phytoseiidae) is among the most commonly used predatory mites in biological control programs. In this study, survival time of gravid N. californicus females reared on mixed stages of T. urticae only or P. latus only when held without food and water as well as their dorsal-shield size (length and width) were investigated. N. californicus females were obtained by placing eggs individually on bean leaf discs (2 cm in diameter) containing mixed stages of T. urticae only or P. latus only and reared to adults. To ensure mating, two N. californicus males were added individually to each leaf disc when the N. californicus females reached the deutonymphal stage. Twenty-four hours after adults emerged, gravid N. californicus females reared on two diets were transferred individually into a gelatin capsule. The survival times of predators were recorded at 12-hour intervals until death. After females died, they were cleared in lacto-phenol medium. The cleared specimens were then mounted in Hoyer's medium. The length and width of the dorsal shield of the mated females reared on the two different diets were measured. The results indicated that the length of the dorsal shield was not affected by diet type. However, the dorsal shields of mated females reared on T. urticae only were statistically wider than those of females reared on P. latus only. Gravid N. californicus females also exhibited shorter survival times in the absence of food and water if they were reared on P. latus only, while females reared on T. urticae only survived longer.

Keywords: Twospotted spider mite, Yellow tea mite, Neoseiulus californicus, Biological control, Prey-food suitability.



# Effectiveness of *Steinernema carpocapsae* Weiser and *Steinernema feltiae* Filipjev against *Capnodis tenebrionis* L. (Coleoptera: Buprestidae) with Surface Irrigation on a Peach Orchard

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**Abstract:** *Capnodis tenebrionis* L. (Coleoptera: Buprestidae), is one of the most important pest which causes desiccation of stone fruit trees. Entomopathogenic nematodes (EPNs) have a high potential for the control of underground pests. In this study, determination of the effectiveness of *Steinernema carpocapsae* Weiser and *S. feltiae* Filipjev was the aim. Before the experiment, surface of the soil around the trees were cleared of weeds and stones. 8 larvae of *C. tenebrionis* L. per tree were buried in tulle pouches with 2 g artificial diet. *S. carpocapsae* and *S. feltiae* were applied to the soil with water from 101 bins in 25 IJ/cm<sup>2</sup> dose with 3 repetitions by using watering cans. Only water was used in control treatment. After the 1<sup>st</sup>, 3<sup>rd</sup>, 5<sup>th</sup> and 7<sup>th</sup> days of the treatment, number of living and dead larvae were calculated and cadavers were placed on White trap to ensure the cause of death was EPNs. At the end of the study, mortality on the 1<sup>st</sup> day was 29,16% for *S. carpocapsae* and 16,66% for *S. feltiae* with 100% mortality on the 5<sup>th</sup> day for both EPNs. In control treatment, mortality on the 7<sup>th</sup> day was 8,33%. With these results, it is possible to conclude that EPNs are highly effective against *C. tenebrionis* in natural conditions.

Keywords: Capnodis tenebrionis, Steinernema carpocapsae, Steinernema feltiae, Surface irrigation.

#### **1. INTRODUCTION**

Biological control has become a preferred pest control method because of the lack of residue on food and it also does not damage non-target organisms. Also biological control has an advantage for the management of subterranean pests and other pests that live in isolated spaces. Biological control is more effective than other control methods because of the ability of biological control agents to reach their environment. EPNs are especially successful against these type of pests because they naturally live in soil and move in the galleries opened by pests.

EPNs, which lives in soil, kills insects with the help of symbiotic *Xenorhabdus* and *Photorhabdus* bacteria species in their bodies (Akhurst, 1993). Infective juveniles, which is the larval stage of EPNs that can move freely in the soil, enter the insect body through natural orifices and kill their host by releasing their symbiotic bacteria into the insect. Then EPNs reproduce in the suitable environment created by the bacteria and new infective juveniles emerge from the cadaver with diminishing food material to find new hosts (Grewal et al., 1997).

Like other subterranean pest species, the chemical control of C. tenebrionis, whose neonate larvae spend some time in soil until burrowing into the host's roots, is not effective. There are some studies about the usage of EPNs for the control of this pest, whose neonate larva enters into the roots of stone fruit trees and creates galleries in xylem by feeding under the bark, in controlled conditions. But there are no studies about the usage of EPNs against C. tenebrionis in natural conditions in Turkey. Thus, this the first study about the application of EPNs for the biological control of C. tenebrionis with the usage of surface irrigation technique in Turkey.

The purpose of this study was to investigate the effectiveness of *S. carpocapsae* and *S. feltiae* applications with surface irrigation against neonate larvae of *C. tenebrionis*.

#### 2. MATERIAL AND METHODS

#### Materials

The main materials of the study were neonate larvae of *C. tenebrionis* which was collected from Çanakkale in Lapseki Province, native isolates of *S. feltiae* and *S. carpocapsae*, which was isolated from soils of Turkey, surface irrigation system, watering cans and artificial diet for *C. tenebrionis* larvae. The artificial diet is prepared by the method of Garrido et al (1987).

#### Methods

The study was conducted in a 10 decares peach orchard in Çardak town of Çanakkale Province (40°23'39 "N, 26°44'48" E). Before the study, the presence of natural EPN species in the orchard soil was tested by soil sampling. The results of these tests have shown the lack of natural EPN presence in the soil and we decided to use this orchard for our study.

Stones and weeds were cleared from the soil surface around the trees before EPN application. Before the experiment, neonate larvae of *C. tenebrionis* were placed into tulle pouches with 2 g artificial diet and the ability of the larvae to pass through the tulle was tested. Tulle pouches were placed under the soil around the trees homogeneously. Ribbons were tied to tulle pouches for easier detection and the tip of the ribbon was left out of soil.

Surface irrigation was used for EPN treatment into the soil at the bottom of the trees (Figure 1). EPNs were applied with 10 l water/tree with watering cans. EPNs were transferred to the orchard in falcon tubes (10 ml) in an ice box. EPN suspensions were poured into the watering cans and the water in the can was mixed to prevent the sedimentation of EPNs at the bottom. The treatment dose was calculated as 25 IJs/cm<sup>2</sup> and totally 140.000 IJs were applied per tree. The study was conducted in 3 replications and the only water was used in control treatment with the same method.

Tulle pouches were controlled in the first, third, fifth and seventh days after the applications and dead larvae of *C. tenebrionis* were transferred to White trap (White, 1927) to determine the cause of death. At the end of the study, mortalities of *C. tenebrionis* larvae in surface irrigation treatment were calculated.



Figure 1. A view of surface irrigation application from peach orchard

#### **Statistical Analysis**

Data from the study was analyzed with Repeated measures ANOVA on SPSS® 23 software. The Tukey multiple comparison test (P < 0,01) was used to determine the differences between days, doses and species in MSTATC®.

#### 3. RESULTS AND DISCUSSION

Mortalities of *C. tenebrionis* neonate larvae after the application of EPNs into the soil were given in Table 1. As seen in the table, mortalities from *S. feltiae* and *S. carpocapsae* were 16,66% and 29,16% on the 1<sup>st</sup> day, 45,83% and 58,33% on the 3<sup>rd</sup> day, respectively. In both species, 100% mortality was observed on the 5<sup>th</sup> day, while mortality in control treatment was 8,33% on the 7<sup>th</sup> day. Statistical analysis has shown that the difference between the mortalities from *S. feltiae* and *S.* 

*carpocapsae* was not statistically significant, while mortalities from both species were significantly higher than the mortality from the control treatment (P=0,00; F=166,37; df=2).

| Day | S. feltiae       | S. carpocapsae   | Control         |
|-----|------------------|------------------|-----------------|
| 1   | 16,66±7,21       | 29,16±19,09      | $0,00{\pm}0,00$ |
|     | B a              | B a              | A b             |
| 3   | 45,83±26,02      | 58,33±7,21       | $0,00{\pm}0,00$ |
|     | AB a             | AB a             | A b             |
| 5   | $100,00\pm 0,00$ | $100,00\pm 0,00$ | $0,00{\pm}0,00$ |
|     | A a              | A a              | A b             |
| 7   | $100,00\pm 0,00$ | $100,00\pm 0,00$ | 8,33±14,43      |
|     | A a              | A a              | A b             |

Table 1. Mortality of Capnodis tenebrionis larvae after surface irrigation application in peach orchard

Different capital letters in the same column indicate a significant difference between means ( $P \le 0,01$ )

Different small letters in the same row indicate a significant difference between means ( $P \le 0,01$ )

When time related mortalities from both species were evaluated, it was observed that the mortality in the first day was significantly lower than the mortality from the fifth and the seventh days (F=93,50; P=0,00; df=3), while the difference between the mortalities from the third, the fifth and the seventh days was not significant (F=45,32; P=0,72; df=3). Similarly, the difference between the mortalities from the first and the third day was also not significant. In control treatment, there was no significant difference between time related mortalities. Interaction of mortalities related to time and species was found to be statistically significant (F=20,410; P=0,00; df=6), so mortalities of *C. tenebrionis* larvae change with species and the days after the treatment both.

As the result of this study, both EPN species were highly effective (reaching up to 100%) at controlling the neonate larvae of *C. tenebrionis* in natural conditions with surface irrigation. Further studies are need to determine the suitable application dose and EPN persistency in the soil after treatment.

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#### REFERENCES

- Akhurst, R. J., 1993. Bacterial symbionts of entomopathogenic nematodes-the power behind the throne. In R Bedding, R Akhurst & H K Kaya (Eds.), Nematodes and the Biological Control of Insect Pests, CSIRO Publications, East Melbourne, Australia, pp. 127-135
- Garrido, A., Del Busto, T., and Malagón, J., 1987. Me'todo de recogida de huevos de *Capnodis tenebrionis* L. (Coleoptera: Buprestidae) y algunos factores abióticos que pueden condicionar la puesta. Bol. San. Veg. Plagas 13: 303-309.
- Grewal, P. S., Lewis, E. E., and Gaugler, R., 1997. Response of infective stage parasites (Nematoda: Steinernematidae) to volatile cues from infected hosts. Journal of Chemical Ecology, 23(2): 503-515.
- Martinez de Altube, M., Strauch, del, O., Fernandez de Castro, G., and Pena, A. M., 2008. Control of theflat-headedrootborer *Capnodis tenebrionis* (Linnéaus) (Coleoptera: Buprestidae) with the entomopathogenic nematode *Steinernema carpocapsae* (Weiser) (Nematoda: Steinernematidae) in a chitosan formulation in apricot orchards. BioControl, 53(3): 531-539.
- Morton, A., and Del Pino, F. G., 2008. Field efficacy of the entomopathogenic nematode *Steinernema feltiae* against the Mediterranean flat-headed rootborer *Capnodis tenebrionis*. J. Appl. Entomol., 132: 632-637.
- White, G. F., 1927. A method for obtaining infective nematode larvae from cultures. Science, 66: 302-303.



# Evaluation of System Performance of Some Irrigation Projects in the West Black Sea Basin

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**Abstract:** Within the scope of this study, comparative performance indicators of irrigation systems which are located in Kastamonu and were brought into service by DSI (State Hydraulic Works) were evaluated by examining 2018 data of Karadere Irrigation Union, Germeçtepe-Kırcalar Irrigation Union, Karaçomak Irrigation Union, Hasköy Irrigation Union, and Beyler Irrigation Union which are water user organizations that provide operation and maintenance service. While evaluating the performance of irrigation unions affiliated to the DSI 23<sup>rd</sup> Regional Directorate, some of the activity indicators included in the comparative assessment guide developed by the International Program for Technology and Research in Irrigation and Drainage (IPTRID) and World Food and Agriculture Organization (FAO) were used. The activity indicators are used to assess the impact of interventions on irrigation networks, to compare the performance of the system over time and to compare performance between systems.

Keywords: Performance indicators, irrigation union, Western Black Sea Basin irrigation unions



# Efficacy of Entomopathogenic Nematodes against Alfalfa Weevil *Hypera postica* Gyllenhal (Coleoptera: Curculionidae)

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**Abstract:** The alfalfa weevil *Hypera postica* Gyll. (Coleoptera: Curculionidae) is one of the primary insect that causes damages on alfalfa. Both larva and adult of *H. postica* feed on Alfalfa foliage, but the larvae cause the majority of the damage. In this study the efficacy of entomopathogenic nematodes (EPN) against *H. postica* were determined under laboratory conditions. In bioassays *Steinernema feltiae*, *S. carpocapsae* and *Heterorhabditis bacteriophora* were used dose of 500 IJs/larva at two different temperatures 20 and 25 °C. *S. carpocapsae* showed the highest efficacy on the larvae of *H. postica* with 100% at the end of the 4<sup>th</sup> day at 20 °C and followed by *S. feltiae* 100% and *H. bacteriophora* 100% respectively. *S. carpocapsae* was able cause 100% mortality at 25 °C and followed by *H. bacteriophora* 100% and *S. feltiae* 100% respectively. According to the findings of the study, it can be useful to confirm the efficacy in field experiments to investigate the potential of EPNs in the biological control of *H. postica*.

Keywords: Fodder crop, entomopathogenic nematodes, alfalfa weevil, biological control

#### **1. INTRODUCTION**

Fodder crops production is the most important way of continuous and safe feed production and as insurance of plant and animal production. Among the fodder crops; alfalfa, vetch, creeper, wrench and crocus are used primarily by animals, and then turned into products as meat, milk etc. and people can benefit these. In recent years, due to the investments in the animal husbandry sector and feed requirements, alfalfa production is supported and encouraged by goverment.

There are many pests that damage on fodder crops, alfalfa weevil *Hypera Postica* Gyll. (Coleoptera: Curculionidae), is one of these pests. It particularly reduces the growth of plant due to the feeding on buds and shoots in alfalfa production areas. The leaves are started to dry and a silvery appearance is occurred when the population of this pest is high. The economic importance of *H. postica* is great in the areas with limited watering and less harvesting. Chemical control is the most preferred method for this pest. However, due to the intensive and unconscious use of chemicals, many problems can be occurred such as pest resistance, toxic effects on natural enemies and non-target organisms. Furthermore, it can cause problems in the environment, animal and human health because of the residual problems on agricultural crops. Due to these negative aspects, entomopathogenic nematodes (EPNs) can be considered as an alternative method to chemical control. EPNs are soil-inhabiting, lethal insect parasites that belong to the Phylum Nematoda from the families Steinernematidae and Heterorhabditidae, and they have proven to be the most effective as biological control organisms of soil and above-ground pests (Kaya and Gaugler, 1993; Laznik et al. 2010). Potential of EPNs are more efficient against soil-dwelling pests or pests in cryptic habitats such as galleries in plants where IJs find excellent habitat to survive and protect themselves from environmental factors.

*H. postica* larvae feed nearly 3 to 4 weeks depending on the quality of alfalfa and temperature. Larvae molt or shed their skins three times and following the last instar, cocoons are spun on plants or in curled up leaves that have fallen to the soil. Due to this biology of the pest, EPNs can be used successfully in biological control studies. Otherwise, the use of chemical products to control insect pests is costly and can harm the environment. So in this study to determine the efficacy of EPNs native isolates collected from different provinces of Turkey were used against the adults of *H. postica* under laboratory conditions.

#### 2. MATERIALS AND METHODS

#### **Entomopathogenic Nematodes Culture**

Three native species of nematodes; *S. carpocapsae* (Weiser) 1133, *S. feltiae* 113 and *Heterorhabditis bacteriophora* (Poinar) 1144 were evaluated against the alfalfa weevil larvae. Each isolates was reared in the last instar of wax moth larvae *Galleria mellonella* L., which is the most commonly used insect host for in vivo production of EPNs (Bedding and Akhurst, 1975; Lindegren et al., 1993; Kaya and Stock, 1997). *G. mellonella* was preferred because of its high susceptibility to the most nematodes, wide availability, ease in rearing, and high yields (Shapiro-Ilan and Gaugler, 2002; Woodring and Kaya, 1988). Nematode-killed *G. mellonella* larvae were placed on White trap (White, 1927) at 25 °C and IJs that emerged from cadavers were harvested. These IJs were rinsed in distilled water and used within a week for the experiments. Before using the nematodes, their viability was checked under the stereomicroscope.

#### Hypera postica Culture

*H. postica* larvae were collected from alfalfa located in Kumkale district of Çanakkale. The leaves and shoots of the alfalfa were observed and alfalfa weevil infected samples were brought to the laboratory and prepared for efficacy assays.

#### Laboratory Bioassays

Efficacy bioassays were conducted at 20 and 25 °C by 500 IJs/adult in a 12 well plates with two replicates. At the bottom of each well Whatman filter paper was put and one adult of *H. postica* was placed. *S. carpocapsae* 1133, *S. feltiae* 113 and *H. bacteriophora* 1144 were inoculated to the larvae of *H. postica* and larvae were checked 1<sup>st</sup>, 3<sup>rd</sup>, 5<sup>th</sup> and 7<sup>th</sup> days after nematode inoculation. Only distilled water without nematodes was applied to the control plates. Infected *H. postica* adults were transferred to White trap to observe the emergence of IJs.

#### 3. RESULTS AND DISCUSSION

Mortality of *H. postica* adults caused by EPNs in plate bioassays were ranged depending on nematode species and temperature. *S. carpocapsae* 1133, *S. feltiae* 113 and *H. bacteriophora* 1144 caused 83.33, 75 and 50% at 20 °C, respectively and 100, 83.33 and 91.66% at 25 °C, respectively (Table 1).

| Temperature (°C) | Isolate               | Mortality (%) |
|------------------|-----------------------|---------------|
|                  | S. carpocapsae 1133   | 100           |
| 20               | S. feltiae 113        | 100           |
|                  | H. bacteriophora 1144 | 100           |
|                  | S. carpocapsae 1133   | 100           |
| 25               | S. feltiae 113        | 100           |
|                  | H. bacteriophora 1144 | 100           |

Table 1. Mortality of Hypera postica larvae caused by entomopathogenic nematodes in plate bioassays

Infected *H. postica* larvae were dissected under a stereomicroscope, freshly emerged IJs were seen easily and collected in culture flasks.

According to the results of the study it is considered that EPNs have potential in management of *H. postica*. This is the first study focused on the efficacy of native EPNs against *H. postica* that conducted both in Çanakkale and Turkey. The study was ended at the end of 5<sup>th</sup> day, the results were evaluated and the highest mortality was observed on the 5<sup>th</sup> day. All EPN species used in the study, showed that high efficiencies was found as *S. carpocapsae* with 100% mortality at 20 °C, followed by *S. feltiae* with 100% and *H. bacteriophora* with 100%, respectively. As the temperature increases, the mortality rates were not changed.

In similar study Kim et al (2007), determined the efficacy *H. postica* to Korean EPNs, *S. carpocapsae* GSN1 strain (ScG), *S. glaseri* Dongrae strain (SgD), *H. bacteriophora* Hamyang strain (HbH), and *Heterorhabditis* sp. Gyeongsan strain (HeG) at the petri dish assay. The larval mortality of *H. postica* was significantly changed depending on nematode species and doses. SgD and HeG strain were more effective against the larva of *H. postica* than ScG and HbH strain. When SgD and HeG strain were treated with the rate of >20 IJs/larva, mortality was found 77.5-100% at the last instars of *H. postica* in 3 days. The number of established nematode was significantly different based on nematode species, whereas number of progeny was not significantly different. The mean number of established us of SgD strain in a host was the highest at 80 IJs by 30.2. The highest progeny number of HeG strain was 2,671.5 with 80 IJs. All nematode strains were not parasitic

entirely to *H. postica* adults. Their results proved that last instar larva of *H. postica* is highly susceptible to Korean EPNs under laboratory conditions.

Shah et al (2011), *H. indica, S. carpocapsae* and *S. thermophillum* were tested against *H. postica* in laboratory bioassays and micro plot experiment. Laboratory bioassays were conducted utilizing *S. thermophillum* and *H. indica*. They found a positive correlation between nematode concentration, exposure time and *H. postica* mortality. LC 50 values of *S. thermophillum* at 24, 48, 72 and 96 hrs were found to be 108.88, 18.31, 4.96 and 3.49 IJs respectively, while LC 50 values of *H. indica* at 24, 48 and 72 hrs post inoculation were found to be 45.00, 30.41 and 18.59 IJs respectively. Microplot experiment utilizing *S. carpocapsae* and *H. indica* indicated that *H. postica* population was significantly lower in *H. indica* and *S. carpocapsae* application at a dose of 1 billion IJs/acre. A reduction of 72.10% and 49.66% grub population over untreated control were noticed in *H. indica* and *S. carpocapsae* application respectively.

Carrera et al (2013), were conducted a study for potential control, short-term field persistence and the vertical distribution of native EPNs against *Curculio nucum* L. (Coleoptera: Curculionidae) in Muntanyes de Prades, Tarragona during two years. *S. feltiae* strain D114, *Steinernema* sp. strain D122 and *H. bacteriophora* strain DG46 were used in summer and spring applications at a dosage of 5-105 IJs m<sup>2</sup>. All isolates decreased the pest population, ranging from 32 to 88%. Persistence evaluation was carried out during 9 weeks for *S. feltiae* (D114), *Steinernema* sp. (D122) and *H. bacteriophora* (DG46) and showed all species capable of lasting for this period. Nematodes and larval vertical distribution was assessed. Most of the *C. nucum* stayed within the first 25 cm although some were found as deep as 40 cm, EPNs were found along all 40 cm depth. They reported the suitability of EPNs to control *C. nucum*.

#### 4. CONCLUSION

It has been considered that it will be helpful to determine the efficacy of these isolates against *H. postica* by the further study that is going to be conducted on the field. However, to control *H. postica* effectively, it is critical to combine all available control measures including cultural methods, other biological control agents, and the proper and judicious use of registered pesticides.

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#### REFERENCES

- Bedding, R. A., and Akhurst, R. J., 1975. A simple technique for the detection of insect parasitic Rhabditid nematodes in soil. Nematologica, 21: 109-110.
- Carrera-Batalla, L., Morton, A., and Garcia-del-Pino, F., 2013. Field efficacy against the hazelnut weevil, *Curculio nucum* and short-term persistence of entomopathogenic nematodes. Spanish Journal of Agricultural Research, 11(4): 1112-1119.
- Kaya, H. K., and Gaugler, R., 1993. Entomopathogenic nematodes. Annual Review of Entomology, 38: 181-206.
- Kaya, H. K., and Stock, S. P., 1997. Techniques in Insect Nematology. In: L. Lacey (Ed.), Manual of Techniques in Insect Pathology, Academic Press, San Diego, CA, pp. 281-324.
- Kim, H. H., Han, G. Y., Park, C. C., Choo, H. Y., Cho, S. R., Lee, H. S., Lee, D. W., and Park, C. G., 2007. Susceptibility of the alfalfa weevil, *Hypera postica* (Coleoptera: Curculionidae) to Korean entomopathogenic nematodes in laboratory assays. Korean J. Appl. Entomol, 46(1): 147-151.
- Laznik, Ž., Tóth, T., Lakatos, T., Vidrih, M., Trdan, S., 2010. Control of the Colorado potato beetle (*Leptinotarsa decemlineata* [Say]) on potato under field conditions: A comparison of the efficacy of foliar application of two strains of *Steinernema feltiae* (Filipjev) and spraying with thiametoxam. Journal of Plant Disease Protection, 117: 129-135.
- Lindegren, J. E., Valero, K. A., and Mackey, B. E., 1993. Simple in vivo production and storage methods for *Steinernema carpocapsae* infective juveniles. Journal of Nematology, 25: 193-197.
- Shah, N. K., Azmi, M. I., and Tyagi, P. K., 2011. Pathogenicity of Rhabditid nematodes (Nematoda: Heterorhabditidae and Steinernematidae) to the grubs of alfalfa weevil, *Hypera postica* (Coleoptera: Curculionidae). Range Management and Agroforestry, 32(1): 64-67.
- Shapiro-Ilan, D. I., and Gaugler, R., 2002. Production technology for entomopathogenic nematodes and their bacterial symbionts. Journal of Industrial Microbiology and Biotechnology, 28: 137-146.
- White, G. F., 1927. A method for obtaining infective nematode larvae from cultures. Science, 66: 302-303.

Woodring, J. L., and Kaya, H. K., 1988. Steinernematid and Heterorhabditid Nematodes: A Handbook of Biology and Techniques, Southern Cooperative Series Bulletin 331. Arkansas Agricultural Experiment Station, Fayetteville, Arkansas, pp. 1-17.



# Control of *Palpita unionalis* Hübner (Lepidoptera: Pyralidae) by Native Entomopathogenic Nematodes

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**Abstract:** In this study native entomopathogenic nematodes (EPNs); *Steinernema carpocapsae* 1133 and *Heterorhabditis bacteriophora* 1144, were tested as plate and pot experiments to determine their efficacy on the last instar larvae of the olive leaf moth, *Palpita unionalis* Hübner (Lepidoptera: Pyralidae) under laboratory conditions. EPNs were isolated from Sakarya and *P. unionalis* larvae were collected from olive orchards in Çanakkale province. Efficacy assays were done by using two application doses; 25 and 50 IJs/larva at 25 °C. In pot experiments ten *P. unionalis* larvae were used for each pot by dose of 40.000 IJs/pot at 25 °C. *P. unionalis* larvae were checked on the 1<sup>st</sup>, 3<sup>rd</sup>, 5<sup>th</sup>, 7<sup>th</sup> and 9<sup>th</sup> day after nematode application. In plate experiments; mortalities occurred on *P. unionalis* larvae caused by *S. carpocapsae* 1133 and *H. bacteriophora* 1144 were found 91.6% by dose of 25 IJs and 100% by dose of 50 IJs for both species. In pot experiments; mortalities were determined as 60, 100%, respectively. Results of this study show that EPNs caused high mortality both in plate and pot experiments so it will be useful to determine the usage potential of these EPN species as biological control agent of *P. unionalis* by further studies that conducted in the field.

Keywords: Palpita unionalis, olive, entomopathogenic nematodes, biological control

#### **1. INTRODUCTION**

Olive cultivation has been an economic activity for centuries in Anatolia and many researches show that the motherland of the olive is the Mediterranean shores of Anatolia. It is nearly grown at an area of 8.460.619 decares particularly in Aegean, Marmara and Mediterranean coasts of Turkey (Fao, 2017).

Many pests, fungal and bacterial diseases and weeds can cause reduced growth and yield in olive trees. *Palpita unionalis* Hübner (Lepidoptera: Pyralidae) is one of these pests and was reported for the first time by the İyriboz in the Aegean Region of Turkey in 1931 (Nizamlioğlu and Gökmen, 1964). *P. unionalis* was reported as a potential pest in olive orchards in Bursa province.

Broad-spectrum pesticides used in olive orchards can kill natural enemies of the pest and make *P. unionalis* as an important pest. The pest causes serious damages particularly in young olive plantations and nurseries (Kovancı and Kumral, 2004). Olive leaf moth larvae move quickly after hatching and newly hatched larvae usually feed on the parenchyma of the lower surface of the leaves-drying the upper epidermis, which turns brown. As larvae grown, especially after 2<sup>nd</sup> and 3<sup>rd</sup> instar of larva, damages on olive plant also increase. In heavy infestations, larvae attack the olive fruit, especially table varieties, by causing irregular holes in the surface of olive fruit and making them unacceptable for the commercial market (Badawi et al., 1976; Balachowsky, 1972).

In the studies carried out in Aegean and Marmara Region of Turkey it is reported that *P. unionalis* has done serious damages in young saplings (1-1,5 m in height) (İyriboz, 1968; Kovancı et al., 2006). *P. unionalis* usually leave eggs individually or in groups of 2 and 28 to the lower surface of leaves. There are a few information about on the phenology and biology of *P. unionalis* in olive orchards. In Italy, it has four to five generations (Martelli, 1915), in Israel six (Avidov and Harpaz, 1969), in Spain five (Fodale et al., 1988), in France two (Balachowsky, 1972), in Turkey two to three (Kovancı et al., 2006), in Greece three (Mazomenos et al., 2002), and in Egypt, the moth completes ten generations per year (Badawi et al., 1976).

Biological control studies should be increase because of many reasons such as leaving many eggs, being a multivoltin species, being active in the tree all the year of *P. unionalis*. And also the widespread use of pesticides cause many harmful

effects on environment. Alternatively, biological control may be considered using entomopathogenic nematodes (EPNs), due to having potential as biological control agents. EPNs are parasites of soil-borne organisms that infect pests occurring in, close or on top of the soil surface.

EPNs belong to the Steinernematidae and Heterorhabditidae families are identified by carrying symbiotic bacteria of the genus *Xenorhabdus* (Thomas and Poinar, 1979) and *Photorhabdus* (Boemare Akhurst and Mourant, 1993) in their intestine, respectively (Ehlers, 1996; Boemare, 2002). These specific bacteria have an important role in the pathogenicity; they multiply and kill the host insect. EPNs have many advantages such as their high reproductive potential, the ability to kill hosts quickly, high virulence, broad host range, easy mass rearing, and safety to plants, vertebrates, and other nontarget organisms (Kaya and Gaugler, 1993).

Most of the studies conducted in Turkey are about the biology of *P. unionalis* but there has been no study done on the efficacy of EPNs against the pest. In this study it is aimed to determine the efficacy of native EPNs on the larvae of *P. unionalis* under laboratory conditions.

#### 2. MATERIALS AND METHODS

#### Source and Rearing of Entomopathogenic Nematodes

Two native species of nematodes; *S. carpocapsae* (Weiser) (isolate 1133) and *Heterorhabditis bacteriophora* (Poinar) (isolate 1144) were evaluated against the olive leaf moth larvae. Each isolates was reared in the last instar of wax moth larvae *Galleria mellonella* L., which is the most commonly used insect host for in vivo production of EPNs (Bedding and Akhurst, 1975; Lindegren et al., 1993; Kaya and Stock, 1997). *G. mellonella* was preferred because of its high susceptibility to the most nematodes, wide availability, ease in rearing, and high yields (Shapiro-Ilan and Gaugler, 2002; Woodring and Kaya, 1988). Nematode-killed *G. mellonella* larvae were placed on White trap (White, 1927) at 25 °C and IJs that emerged from cadavers were harvested. These IJs were rinsed in distilled water and used within a week for the experiments. Before using the nematodes, their viability was checked under the stereomicroscope.

#### Source of Palpita unionalis

*P. unionalis* larvae were collected from olive orchards located in Gülpınar village of Ayvacık district of Çanakkale. The leaves and shoots of the olive trees were observed and olive leaf moth infected samples were brought to the laboratory and prepared for efficacy assays.

#### Laboratory Bioassays

Efficacy bioassays were conducted at 25 °C by 25 and 50 IJs/larva in a 12 well plates with two replicates. At the bottom of each well Whatman filter paper was put and one last instar larva of *P. unionalis* was placed. *S. carpocapsae* 1133 and *H. bacteriophora* 1144 were inoculated to the larvae of *P. unionalis* and larvae were checked 1<sup>st</sup>, 3<sup>rd</sup>, 5<sup>th</sup> and 7<sup>th</sup> days after nematode inoculation. Only distilled water without nematodes was applied to the control plates. Infected *P. unionalis* larvae were transferred to White trap to observe the emergence of IJs.

#### **Pot Experiments**

Gemlik cultivars were used in pot experiments (1,5 years old), 10 *P. unionalis* larvae were given for each potted olive sapling. Wintering larvae that were fed together were inoculated by 40.000 IJs/pot in a 50 ml of water. *S. carpocapsae* 1133 and *H. bacteriophora* 1144 were inoculated to the larvae of *P. unionalis* and larvae were checked 1<sup>st</sup>, 3<sup>rd</sup>, 5<sup>th</sup> and 7<sup>th</sup> days after nematode inoculation. Only distilled water without nematodes was applied to the control pots. Infected *P. unionalis* larvae were transferred to White trap to observe the emergence of IJs.

#### 3. RESULTS AND DISCUSSION

Mortality of *P. unionalis* larvae caused by EPNs in plate bioassays were ranged depending on nematode species and application dose. *S. carpocapsae* 1133 and *H. bacteriophora* 1144 caused 91,6% for both species by dose of 25 IJs/larva and both 100% for both species by dose 50 IJs/larva (Table 1).

Table 1. Mortality of *Palpita unionalis* larvae caused by entomopathogenic nematodes in plate bioassays

| Dose (IJs) | Isolate               | Mortality (%) |
|------------|-----------------------|---------------|
| 25         | S. carpocapsae 1133   | 91,6          |
| 23         | H. bacteriophora 1144 | 91,6          |
| 50         | S. carpocapsae 1133   | 100           |
| 50         | H. bacteriophora 1144 | 100           |

Mortality of *P. unionalis* larvae caused by EPNs in pot experiment were changed by nematode species. *S. carpocapsae* 1133 and *H. bacteriophora* 1144 caused 60 and 100%, respectively (Table 2).

Table 2. Mortality of Palpita unionalis larvae caused by entomopathogenic nematodes in pot experiments

| Dose (IJs) | Isolate               | Mortality (%) |
|------------|-----------------------|---------------|
| 40.000     | S. carpocapsae 1133   | 60            |
| 40.000     | H. bacteriophora 1144 | 100           |

Infected *P. unionalis* larvae were dissected under a stereomicroscope, freshly emerged IJs were seen easily and collected in culture flasks (Figure 1).



Figure 1. Infective juveniles of Steinernema carpocapsae in the cadaver of Palpita unionalis.

Results of this study proved that EPNs have great potential to infect *P. unionalis* larvae both in plates and pot experiments. This is the first study conducted both in Çanakkale and in Turkey that focused on the efficacy of native EPNs against *P. unionalis* in olive. In a similar study, Mahmoud (2014), evaluated the efficacy of *S. feltiae*, *S. carpocapsae* and *H. bacteriophora* on *P. unionalis* by dose of 0, 5, 10, 20, 40, 80 and 120 IJs/larva. The most efficient species was found as *S. carpocapsae* by 120 IJs/larva with 96% mortality and followed by *S. feltiae* with 88% and *H. bacteriophora* with %84. Our results are consistent with the results of this study.

In another study, Yurt et al (2015), determined the virulence of four native EPN isolates; *S. affine, S. feltiae, S. carpocapsae* and *H. bacteriophora* at 15, 20 and 25 °C) against *Pieris brassicae* L. (Lepidoptera: Pieridae). Mortalities were recorded on  $7^{\text{th}}$  day after nematode inoculation. The highest mortality of *P. brassicae* larvae was occurred on *S. feltiae* 97 with 84.3% at 25 °C.

Odendaal et al (2016), tested three commercial; *S. feltiae, H. bacteriophora* Hb1 and Hb2 and two native EPN strains; *S. jeffreyense* and *S. yirgalemense* against the larvae of *Cydia pomonella* L. (Lepidoptera: Tortricidae). They conducted field studies and found *S. jeffreyense* as the most efficient species with 67% and followed by *H. bacteriophora* Hb1 with 42%, and *S. yirgalamense* with 41% mortality. Our results clearly demonstrate that larvae of *P. unionalis* were highly susceptible to the EPNs tested and these EPNs can be used as efficient biological control agents against *P. unionalis*.

#### 4. CONCLUSION

It has been considered that it will be helpful to determine the efficacy of these isolates against *P. unionalis* by the further study that is going to be conducted on the field. However, to control *P. unionalis* effectively, it is critical to combine all available control measures including cultural methods, other biological control agents, and the proper and judicious use of registered pesticides.

#### REFERENCES

Avidov, Z., and Harpaz, I., 1969. Plant Pests of Israel. Isreal Universities Press, Jerusalem.

- Badawi, A., Awadallah, A. M., and Foda, S. M., 1976. On the biology of the olive leaf moth *Palpita unionalis* Hb. (Lepidoptera: Pyralidae). Zeitschrift für Angewandte Entomologie, 80(1): 103-110.
- Balachowsky, A. S., Guennelon, G., Real, P., and Touzeau, J., 1972. II. Super-famille des Pyraloidea Tome II, Lepidopteres. In: A. S. Balachowsky (Ed.), Entomologie Appliquee à l'Agriculture. (Ed.,), Masson et Cie Eds., Paris, pp. 1071-1247.
- Bedding, R. A., and Akhurst, R. J., 1975. A simple technique for the detection of insect parasitic Rhabditid nematodes in soil. Nematologica, 21: 109-110.
- Boemare, N. E, Akhurst, R. J., and Mourant, R. G., 1993. DNA relatedness between *Xenorhabdus* spp. (Enterobacteriaceae), symbiotic bacteria of entomopathogenic nematodes, and a proposal to transfer *Xenorhabdus luminescens* to a new genus, *Photorhabdus* gen. nov. International Journal of Systematic Bacteriology, 43: 249-255.
- Boemare, N. E., 2002. Biology, taxonomy and systematics of *Photorhabdus* and *Xenorhabdus*. In: R. Gaugler (Ed.), Entomopathogenic Nematology. Wallingford, UK: CABI Publishing, pp. 35-56.
- Ehlers, R. U., 1996. Current and future use of nematodes in biocontrol: Practice and commercial aspects in regard to regulatory policies. Biocontrol Science Technology, 6: 303-316.
- Fao, 2017. http://www.fao.org/faostat/en/#data/QC (Erişim Tarihi: 03.02.2019).
- Fodale, A. S., Mule, R., and Tucci, A., 1988. Bioethological observations on *Margaronia unionalis* Hübner (Lepidoptera: Pyralidae) in Sicily and trials on its control. Annali dell'Istituto Sperimentale per l'Olivicoltura, 10: 31-44.
- Hazır, S., Kaya, H. K., Stock, S. P., and Keskin, N., 2004. Entomopathogenic nematodes (Steinernematidae and Heterorhabditidae) for biological control of soil pests. Turk. J. Biol., Tübitak, 27: 181-202.
- İyriboz, N., 1968. Zeytin Zararlıları ve Hastalıkları. Tarım Bakanlığı Zirai Mücadele ve Karantina Genel Müdürlüğü Yayınları, İzmir.
- Kaçar, G. Ş., 2011. Doğu Akdeniz Bölgesi zeytinlerinde zeytin fidantırtılı *Palpita unionalis* (Hübn.) (Lepidoptera: Pyralidae)'in mücadelesine esas bazı biyolojik özelliklerinin belirlenmesi PhD Thesis, Çukurova University, Turkey.
- Kaya, H. K., and Gaugler, R., 1993. Entomopathogenic nematodes. Annual Review of Entomology, 38: 181-206.
- Kaya, H. K., and Stock, S. P., 1997. Techniques in Insect Nematology. In: L. Lacey (Ed), Manual of Techniques in Insect Pathology, Academic Press, San Diego, CA, pp. 281-324.
- Kovancı, B., and Kumral, N. A., 2004. Insect Pests in Groves of Bursa (Turkey). 5<sup>th</sup> International Symposium on Olive Growing. 27 Sep-2 Oct 2004, İzmir, Turkey, s 67.
- Kovancı, B., Kumral, N.A., and Akbudak, B., 2006. Bursa ili zeytin bahçelerinde zeytin fidantırtılı, *Palpita unionalis* (Hübner) (Lepidoptera: Pyralidae)'in popülasyon dalgalanması üzerinde araştırmalar. Türkiye Entomoloji Dergisi, 30(1): 23-32.
- Lindegren, J. E., Valero, K. A., and Mackey, B. E., 1993. Simple in vivo production and storage methods for *Steinernema carpocapsae* infective juveniles. Journal of Nematology, 25: 193-197.
- Mahmoud, M. F., 2014. Virulence of entomopathogenic nematodes against the Jasmine Moth, *Palpita unionalis* Hb. (Lepidoptera: Pyralidae). Egyptian Journal of Biological Pest Control, 24(2): 393-397.
- Martelli, G., 1915. Intorno a due specie di Lepidotteri dei generi Zelleria e Glyphodes viventi sull'olivo. Agraria Filippo Silvestri Portici Generalis Bollettino di Zoologia, 10: 89-102.
- Mazomenos, B. E., Konstantopoulou, M. A., Raptopoulos, D., Stefanou, D., Scareas, S., and Tzeiranakis, L. C., 2002. Female calling behaviour and male response to the synthetic sex pheromone components of *Palpita unionalis* (Lepidoptera: Pyralidae).

Proceedings 25<sup>th</sup> Anniversary Jubilee Meeting of the IOBC-WPRS Group "Use of pheromones and other semiochemicals in integrated control" (Samos, Greece, 2000), pp. 203-211.

Nizamlıoğlu, K., and Gökmen, N., 1964. Türkiye'de Zeytine Zarar Veren Böcekler. Yenilik Basımevi, İstanbul.

- Odendaal, D., Addison, M. F., and Malan, A. P., 2016. Entomopathogenic nematodes for the control of the codling moth (*Cydia pomonella* L.) in field and laboratory trials. Journal of Helminthology, 90: 615-623.
- Shapiro-Ilan, D. I., and Gaugler, R., 2002. Production technology for entomopathogenic nematodes and their bacterial symbionts. Journal of Industrial Microbiology and Biotechnology, 28: 137-146.
- Thomas, G. M, Poinar, and Jr, G. O., 1979. *Xenorhabdus* gen. nov., a genus of entomopathogenic, nematophilic bacteria of the family Enterobacteriaceae. International Journal of Systematic Bacteriology, 29: 352-360.
- White, G. F., 1927. A method for obtaining infective nematode larvae from cultures. Science, 66: 302-303.
- Woodring, J. L., and Kaya, H. K., 1988. Steinernematid and Heterorhabditid Nematodes: A Handbook of Biology and Techniques, Southern Cooperative Series Bulletin 331. Arkansas Agricultural Experiment Station, Fayetteville, Arkansas, 1-17.
- Yurt, Ç., Gözel, Ç., and Gözel, U., 2015. Bazı entomopatojen nematod türlerinin *Pieris brassicae* (Linnaeus) (Lepidoptera: Pieridae) üzerindeki etkinlikleri. Türkiye Biyolojik Mücadele Dergisi, 6(2): 77-84.



# The Investigation of Adaptation Capabilities of Some Sunflower Genotypes in Erzurum Irrigated Conditions

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**Abstract:** This study was performed as randomized complete blocks with four replications to determine the suitable sunflower genotypes under irrigated conditions of Erzurum in 2016. The developed new sunflower hybrids by sunflower breeding program of TTAE with higher yields in previous researches were used. Determining yield components were, emergence time (day), %50 flowering period (day), maturation period (day), head diameter (cm), plant height (cm), 1000 seed weight (g), seed yield (kg da<sup>-1</sup>), seed oil content (%) and oil yield (kg da<sup>-1</sup>). In this study, the highest seed yield Aga 1301 (231.9 kg da<sup>-1</sup>) and 16Tr61 (202.7 kg da<sup>-1</sup>) agenotypes, the highest oil content, Aga 1301 (%43.4) and 16Tr61 (%202.7) genotypes were determine. The results of the study show that Aga 1301 and 16Tr61 genotypes candidates are promising in terms of yield and quality in Erzurum ekolojik conditions and reveals that continue to work with this lines is appropriate.

Keywords: Sunflower oil, Helianthus annuus, maturation period, oil content.



# Assessment of Nutrient Levels and Physico-chemical Characteristics of Karasu Stream in Sinop Province (Black Sea)

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Abstract: Knowing the various parameters that affect the water quality gives us important information about whether the water is suitable for its intended use. Owing to anthropogenic activities such as industrial and agricultural activities as well as natural events, including erosion and climate change, natural properties of water decreases day by day. Due to negative quality changes in the water resources, the existing balance in the ecosystem has been changed. The quality of surface waters is very important for the natural environment and people, and therefore their specific physical, biological, chemical and microbiological properties are controversial. Nitrogen and phosphorus concentrations play a key role in detecting the ecological conditions of aquatic systems. Therefore this study, Karasu river's average annual values of nutritional elements and some physicochemical parameters were determined. The measurements of pH, temperature, conductivity, ORP, dissolved oxygen (DO), ammonium, and nitrate were measured by the YSI Professional Series Multiparameter device. Nitrite, phosphate and silicon were analyzed by spectrophotometric method. Also salinity, magnesium, calcium, organic matter and alkalinity were calculated by titrimetric analysis method. pH, temperature (°C), conductivity, salinity DO, free CO<sub>2</sub>, alkalinity, organic matter, calcium and magnesium, ammonium, nitrite, nitrate, phosphate and silicon in the river were determined as,  $7.41\pm0.06$ ,  $16.31\pm0.58$  (°C),  $12.29\pm2.38$  (µs/cm),  $0.13\pm0.02$  (g/L), 5.46±0.16 (mg/L), 18.62±0.30 (mg/L), 220.87±0.83 (mg/L), 62.18±4.12 (mg/L), 22.41±2.99 (mg/L), 40.77±2.31(mg/L), 0.173±0.002 (mg/L), 0.091±0.005 (mg/L), 1.462±0.006 (mg/L), 0.080±0.005 (mg/L) and 1.717±0.145 (mg/L), respectively. As a result, when the results of physicochemical parameters of Karasu River were evaluated according to Surface Water Quality Management Regulation, water quality was found in different classes. That is, it was determined that the classification was changed according to the measured parameter. The results of the study will provide a data infrastructure for future studies in terms of assessing possible environmental pollution in the future. In addition, regular observations are required to maintain water quality and ensure the health of aquatic life in this area.

Keywords: Nutritious elements, physico-chemical, stream, water, creak



# Determination of Water Quality Characteristics of Sarıkum Lake (Sinop) and Water Supply Creeks in Spring-Summer Period

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Abstract: Sarıkum Lake, which is chosen as the study area, is a lagoon lake connected to the sea, and is one of the important Nature Conservation Areas. This important wetland is under the influence of many environmental pollutant factors, due to the dense population growth in the surrounding settlements, especially domestic and agricultural wastes and small streams flowing into the lake. In the first comprehensive study on the quality and quantity of these pollutants, it was aimed to determine the water quality characteristics of the spring-summer periods of Sinop Sarıkum Lake and Water Supply Creeks. In the study, 4 stations were identified in the creeks feeding the Lake Sarikum and the lake, and 8 samples were made in the spring-summer seasons. Water samples from lakes and streams were taken in accordance with standard procedure and water quality characteristics were examined. The temperature, pH, dissolved oxygen (DO), turbidity, salinity, conductivity and oxidation reduction potential (ORP) were measured instantaneously on the terrain with the Hanna HI 9829 model multipametr device. Organic matter and suspended solids were analyzed by gravimetric method and total nitrate  $(NO_3 + NO_2)$  was determined by spectrophotometric (With Rayleu UV-72 spectrometer device) method. The data obtained were evaluated according to the Water Pollution Control Regulation, the Quality Criteria According to the Inland Water Resources Classifications. The results of this study were obtained as follows: Temperature 19.820±0.329 °C, pH 8.256±0.102, DO 2.687±0.239 mg/L, Salinity 3.438±0.099 PSU, ORP 63.508±16.965 mV, Organic matter 31.895±1.077 mg/L, Suspended solids 0.020±0.003 mg/L, Chlorophyll-a: 5.217±0.883 mg/L, Total Nitrate 0.018±0.011 mg/L. Water quality analysis results showed that water temperature, pH, parameters were in Class I (very well) compared with Quality Criteria of Water Pollution Control Regulation (according to Classes of Inland Water Resources), while dissolved oxygen class III (poor). As a result, when looking at the streams feeding the lake and lake, total nitrate, suspended solids (in station I) and pH, DO, salinity (in station VI) were measured at the highest value. It was stated that the stream passing through the village (in station I) uses the water quality as a source of irrigation in agriculture by the villagers. Then, it was determined that the pollution load increased with the discharges of polluted water coming from the environment. At the station VI, which is located in the middle of the lake, it can be a threat to aquatic organisms in the lake ecosystem due to high physico-chemical parameter values and especially because of low dissolved oxygen value. In this study where the sources that threaten the Sarıkum Lake ecosystem and the damages of these sources are determined, it should be ensured that the lake should be regularly maintained and supervised for the sustainability of the lake.

Keywords: Sinop, Sarıkum Lake, nature conservation area, water quality


# Effect of Treatment with Silage Effluent of Wheat Straw on *In vitro* Gas Production, Parameters and Methane Production

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**Abstract:** This study was conducted to evaluate *in vitro* total gas (TG) and methane gas (MG) production, gas production parameters (a + b, c), organic matter digestion (OMD), metabolic energy (ME) and net energy lactation (NEL) of wheat straw treated with silage effluent. Only wheat straw (control group, (S)), straw treated with urea + molasses (SÜM) and silage effluent (SS), water, urea and molasses (SÇÜM), water (SÇ), 50% silage effluent +50% water + urea + molasses (SÇSÜM), and then subjected to 45 days anaerobic fermentation.

Crude protein and crude ash contents of the straw groups treated with silage effluent were increased (P<0.01), and cell wall elements such as ADF and NDF of the straw groups were decreased (P<0.01). In addition, OMS, ME and NEL contents and in vitro gas production (IVIG) of straw were increased by treatment with silage effluent (P<0.01). It was determined that the addition of urea and molasses to wheat straw did not affect the production of methane according to the control, and in vitro gas production parameters were improved (P<0.01). As a result, it has been concluded that wheat straw which has very low feed value can be treated with silage effluent which causes environmental pollution and nutrient loss in order to improve feed value and further works should be carried out to illuminate the subject.

Keywords: Wheat straw, silage effluent, in vitro gas produciton, methane



# Usage Bee Pollen into Laying Hens Diet

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**Abstract:** This study was conducted to evaluate the effects of bee pollen supplementation into layer diets on performance, egg quality and some blood parameters. A total of 96 Lohmann layers, 28 weeks of age as six replicate cages each containing four hens, were randomly allocated to one of four dietary treatments. Diets were control (C) or basal diet without bee pollen (BP) and experimental diets containing %0,5 (BP1), %1,0 (BP2) and %1,5 (BP3) bee pollen. Present study lasted for twelve weeks. During the experimental period, hens were feed ad-libitum and water was available all the times.

Layers fed with diets including bee pollen showed linear increases in body weight and body weight changes at the end of the trial. Supplementation of bee pollen into layer diet linearly decreased feed intake, and linearly increased feed conversion ratio. Inclusion of different levels bee pollen into the basal diet increased egg weight (quadratic) and yolk index (linear) but affected Haugh unit and albumen index as cubically. It was determined that adding at different levels of bee pollen to diets of laying hens linearly decreased cholesterol, triglyceride and P values and linearly increased Mg value in the serum parameters. But other serum parameters were not affected by treatment. In conclusion, it can be stated that bee pollen can be used in laying hen rations due to positive effects of linearly improvement of feed conversion ratio and lowering of serum cholesterol and lipid contents.

Keywords: Bee Pollen, egg quality, laying hens, performance, serum parameters



# Stimulatory Effects of Plant Growth Promoting Rhizobacteria on the Growth of Roots of Tea Cutting (*Camellia sinensis* L.) at Nursery Level

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Abstract: Tea plants grown in acidic soils are strongly dependent on rhizospheric bacteria, whereas it is not clear whether soil bacterial populations improve plant growth, root morphology, root-hair growth and root development. This study were conducted in order to investigate the effects of eleven N<sub>2</sub>-fixing, P-solubilizing and phytohormones-producing bacteria (Pantoea agglomerans PA58, Bacillus subtilis BS21, Roseomonas fauriae RF272, Achromobacter xylosoxidans denitrificans AXD47, Paenibacillus polymyxa PP243, Bacillus atrophaeus BA59, Arthrobacter citreus AC484, Rhizobium radiobacter RR112, Bacillus pumilus BP70, Pseudomonas agarici PA71, and Bacillus subtilis BS76) in comparison to control and mineral N (500 mg N per roted cutting) and NPK ((500 mg N + 100 mg P + 200 mg K per rooted cutting) fertilizer application on root systems, seedling growth in two year-old uniform and well-rooted tea plant (Camellia sinensis L.) under natural soil conditions of Rize, in three years. The inoculating plant growth promoting rhizobacteria increased the plant growth parameters such as number of lateral roots, root length, plant height, root fresh and dry weight, trunk diameter, shoot+leaf weight, dry leaf weight, chlorophyll contents and root: shoot dry weight ratio. Good performance of the growth of tea cuttings in PA58, RF272, PP 243, AC484, RR112, and BP70 inoculations may be due to its ability in formation of vigorous root system which could absorb more nutrients from the soil. The elongation or proliferation of root systems has been suggested to be one of the mechanisms by which PGPR stimulate rooting characteristics of tea. The experiment revealed that the PGPR inoculation was effective treatment to improve the root growth and change in root morphology which enhanced the growth performance of nursery tea seedlings.

Keywords: Tea plant, plant growth promoting rhizobacteria, root growth, root morphology, nursery conditions

## **1. INTRODUCTION**

Tea (*Camellia sinensis*) the most important plant of Turkey is used in the traditional preparation of its national food and is planted widely on acidic soils. Tea has a source of many pharmaceutically important compounds, an attractive and characteristic aroma, good taste, health-promoting effects, and these benefits together make it one of the oldest and most popular non-alcoholic beverage in the world. Bacterial association with root growth is an important new field of agricultural research. In general, beneficial free-living N<sub>2</sub>-fixing and P-solubilizing bacteria are usually referred to as plant-growth-promoting rhizobacteria (PGPR) and are becoming increasingly important as an alternative yield-enhancing method that can reduce the use of chemical fertilizers, and also act as supplements to chemical fertilizers. At present, the use of biological approaches has found a popular role in increasing the circulation of plant nutrients in agriculture, developing sustainable systems in crop production and as an additive to chemical fertilizers for improving crop yield.

Nitrogen (N), one of the most important macro nutrients, affects the use of strong environmental resources, and its derivatives are very important for plant growth and physiology (Ruan et al., 2010). As a leaf crop, tea plants require more nitrogen than most other crops and this nutrient, therefore, plays a key role in improving tea yield and quality (Han et al., 2008; Çakmakçı et al., polonya). Phosphorus is the second most plant nutrient available in soil second to nitrogen and the availability of P in soil is largely controlled by biologically mediated processes. Though soils usually contain a high amount of total phosphorous, most of the phosphorous occur in insoluble form as Fe and Al phosphates in acidic soils. Agronomic use efficiency of fertilizers is low in acidic soil under high rainfall conditions and also P utilization efficiency is very low in the soil due to its low solubility and mobility (Çakmakçı et al., 2010; Çakmakçı, 2016). Low availability of P, due to low native content and high P fixation capacity of acidic soil, is one of the limiting factors on the productivity of tea plants, and plants grown in acidic soils are often exposed mineral stress such as Al and Mn toxicity and P, K, Mg, and K deficiency. In Turkey, tea plantations are predominantly located on highly leached, strongly acidic, and low fertility

eastern Black Sea coast soils is a widespread factor limiting tea productivity. The mild climate with high precipitation and luxuriant vegetation is totally different from that of the rest of Turkey, with 300 days a year of steady, gentle rainfall and equable temperatures. Many studies showed that repeated and excess amounts of chemical fertilizers application can contribute in low N use efficiency and cause tea orchard soil acidification, deterioration of soil properties, potential nitrate leaching, gaseous N emissions, persistence of chemicals in plant products, and also serious water and environmental pollution (Tokuda and Hayatsu, 2004; Han et al. 2008; Kamau et al., 2008; Hirono et al. 2009; Liu et al. 2012, Çakmakçı, 2016). Today, agriculture is based on high inputs of agrochemicals and the over-application of chemical fertilizers is one of the main causes of soil and water pollution and depletion of soil quality.

Use of PGPR in turn having a great impact on root biology, influence plant growth, nutrition and development are important for long-term sustainability. Tea plants grown in acidic soils are strongly dependent on rhizospheric bacteria, whereas it is not clear whether soil bacterial populations improve plant growth, root morphology, root-hair growth and root development. Today, the strategy of improving agricultural production should consider the addition of nitrogen, phosphorus and other plant nutrients through microbial processes and improving the physiological processes involved in growth and morphogenesis. While nutrients are mainly supplied through chemical fertilizers in tea plantations, it is widely accepted that a balanced fertilizer application with efficient use of other inputs is the key to achieve higher crop production, soil, and environmental health. It is clear that the application of organic manures and bioinoculants could minimize these problems as they are advantageous and safe than chemical fertilizers in improving soil fertility (Gebrewold, 2018). The use of organics and biofertilizers in the tea crop in Turkey is very much limited. Therefore, there is an urgent need o study the influence of the straight or integrated application of organic manure and biofertilizers on the reduction of inorganic fertilizers and augmentation of tea yield, quality, and root development. So far, few studies have been conducted on the effects of bacterial inoculations on seedling survival, dry matter weight and root growth of tea plants. The objective of this study was to evaluate possible effects of mineral fertilizer N and NPK, and eleven  $N_2$ -fixing, P-solubilizing and phytohormones-producing bacteria on the seedling growth and root systems in two year-old uniform and well-rooted tea plant under natural soil conditions of Rize, in three years.

## 2. MATERIALS AND METHODS

This study was conducted with tea plant (*Camellia sinensis* L.) under natural soil conditions in order to investigated inoculation with eleven IAA-producing, N<sub>2</sub>-fixing, and P-solubilizing bacterial species (*Pantoea agglomerans* PA58, *Bacillus subtilis* BS21, *Roseomonas fauriae* RF272, *Achromobacter xylosoxidans denitrificans* AXD47, *Paenibacillus polymyxa* PP243, *Bacillus atrophaeus* BA59, *Arthrobacter citreus* AC484, *Rhizobium radiobacter* RR112, *Bacillus pumilus* BP70, *Pseudomonas agarici* PA71, and *Bacillus subtilis* BS76) in comparison to mineral N (500 mg N per roted cutting) and NPK (500 mg N + 100 mg P + 200 mg K per rooted cutting) fertilizer application as well as a control treatment without inoculation and any fertilizer application. We selected eleven N<sub>2</sub>-fixing, P-solubilizing and phytohormones-producing bacteria (PGPR potential PGPR from a pool of seven hundred eighty-six rhizobacterial strains were tested on the seedling growth and root systems in two year-old uniform and well-rooted tea plant in pot experiments in three years at the Ataturk Tea and Horticultural Research Institute of Rize (Table 1). The experiments were conducted in a completely randomized design with four replicates (each having five rooted cutting sapling).

For this experiment, pure cultures were grown in 50% strength tryptic soy broth on a rotary shaker (120

rpm; 25 °C) for 3 days. Bacteria were then harvested by centrifugation (ca. 3000 x g for 10 min), washed and re-suspended in 10 mM sterile phosphate buffer (SPB), pH 7 to a density of 10<sup>9</sup> cfu ml<sup>-1</sup> for the bacterial strains. Rooted cutting were surface-sterilized prior to inoculation by soaking in 25% commercial-grade bleach for 5 min, followed by thorough washing under running tap water and air-drying aseptically overnight at room temperature. Uniform height young rooted cutting were inoculated with each of the PGPR strains 1 day prior to planting. The single bacterial inoculation involved dipping the root system of the seedling into a suspension of each PGPR strains for 60 min, prior to planting. Control plants received 5 ml of diluted SPB with no bacteria. Fresh and dry leaf weight, shoot weight, average and total shoot length, shoot and trunk diameter and plant height were collected for all tea rooted cuttings. Dry weight of shoots and roots was determined after drying the materials to a constant weight at 60°C.

Table 1. Biochemical characteristics of the bacterial strains tested

| Bacterial strain                               | Oxidase | Catalase | N2-fixation | IAA-production | <b>P-solubilization</b> |
|--|---------|----------|-------------|----------------|-------------------------|
| Pantoea agglomerans PA58                       | -       | +        | S+          | S++            | S+                      |
| Bacillus subtilis BS21                         | -       | +        | +           | +              | W+                      |
| Roseomonas fauriae RF272                       | W+      | +        | +           | S+             | S+                      |
| Achromobacter xylosoxidans denitrificans AXD47 | -       | +        | W+          | +              | S+                      |
| Paenibacillus polymyxa PP243                   | +       | +        | +           | S+             | +                       |
| Bacillus atrophaeus BA59                       | -       | +        | W+          | +              | S+                      |
| Arthrobacter citreus AC484                     | +       | +        | S+          | S+             | +                       |
| Rhizobium radiobacter RR112                    | -       | +        | S+          | S+             | +                       |
| Bacillus pumilus BP70                          | +       | +        | +           | S+             | S+                      |
| Pseudomonas agarici PA71                       | +       | +        | S+          | +              | +                       |
| Bacillus subtilis BS76                         | W+      | +        | W+          | +              | S+                      |

\*S+": strong *positive reaction*, "+": positive reaction, "W+": weak positive reaction

## 3. RESULTS AND DISCUSSION

#### Results

Screening results of multiple pllant growth promoting traits are depicted in Table 1. All of the strains tested were able to solubilise P, fixe N and positive for the production of IAA. Three years of trials showed that treatments including bacterial inoculations and fertilizer application significantly affected the parameters investigated compared to the control in tea depending on the years, bacterial strains and growth parameter evaluated. Of the 14 treatments, the maximum trunk diameter in tea seedlings was seen in *Rhizobium radiobacter* RR112 inoculation, followed by *Arthrobacter citreus* AC484, *Bacillus pumilus* BP70, *Roseomonas fauriae* RF272, and NPK fertilizer. Under natural conditions, the inoculation of tea with RF272, BP70 and RR112 strains, and application NPK fertilizers showed a significant increase in the plant height in the tea when compared to the control plants. All the treatments, except for AXD47 and BA59, enhanced fresh shoot+leaf weight as compared to the control. Except for PGPR strains *Achromobacter xylosoxidans denitrificans* AXD47, other treatments significantly increased dry leaf weight of per tea saplings. The maximum chlorophyll value (SPAD) in tea saplings was obtained with the inoculation of RR112 and application of NPK fertilizers followed by AC484, BP70, RF272, and PA58 (Table 2).

| Table   | 2.    | Effect   | of      | bacterial   | and     | mineral   | fertilizers    | on       | the | average | trunk | diameter, | plant |
|---------|-------|----------|---------|-------------|---------|-----------|----------------|----------|-----|---------|-------|-----------|-------|
| height, | shoot | +leaf we | ight, c | lry leaf we | ght, ch | lorophyll | contents (SPAE | )) of te | ea  |         |       |           |       |

| Treatmontat  | Trunk diameter  | Plant height  | Fresh shoot+leaf weight    | Dry leaf weight (g      | SPAD chlorophyll |
|--------------|-----------------|---------------|----------------------------|-------------------------|------------------|
| 1 reatments* | (mm)**          | ( <b>cm</b> ) | (g sapling <sup>-1</sup> ) | sapling <sup>-1</sup> ) | value            |
| Control      | 7.27±0.34 c     | 43.0±1.9 d    | 25.9±1.1 f                 | 9.4±0.4 e               | 65.2±1.85 c      |
| NPK          | 8.09±0.45 a     | 49.4±1.2 a-c  | 35.6±1.5 a                 | 13.6±1.1 ab             | 73.6±1.93 a      |
| Ν            | 7.52±0.37 а-с   | 46.1±3.8 a-d  | 30.6±2.3 cd                | 11.9±0.5 d              | 69.7±2.49 a-c    |
| PA58         | 8.04±0,08 f     | 43.4±3.8 d    | 34.4±1.1 ab                | 13.3±0.9 a-c            | 72.9±2.27 a      |
| BS21         | 7.46±0.43 a-c   | 43.9±1.6 cd   | 31.8±0.3 bc                | 12.7±0.3 b-d            | 71.6±3.46 ab     |
| RF272        | 8.14±0,05 a     | 51.2±2.6 a    | 34.6±1.4 ab                | 14.2±0.4 a              | 73.2±2.74 a      |
| AXD47        | 7.28±0.44 c     | 47.1±2.7 a-d  | 25.3±1.2 f                 | 9.5±0.5 e               | 68.6±2.87 a-c    |
| PP 243       | 8.12±0.45 a     | 46.1±2.2 a-d  | 33.5±1.0 a-c               | 13.3±0.8 a-c            | 70.7±1.50 ab     |
| BA59         | 7.68±0.35 a-c   | 45.6±2.0 b-d  | 27.5±1.2 ef                | 11.9±0.6 d              | 67.1±2.74 bc     |
| AC484        | 8.18±0.30 a     | 46.5±1.6 a-d  | 32.8±1.4 a-c               | 12.8±0.7 b-d            | 73.3±2.23 a      |
| RR112        | 8.21±0.33 a     | 49.3±1.6 a-c  | 35.2±1.7 a                 | 14.2±0.4 a              | 73.6±2.39 a      |
| BP70         | 8.18±0.28 a     | 49.7±3.6 ab   | 35.7±1.9 a                 | 14.4±0.8 a              | 73.3±1.90 a      |
| PA71         | 7.51 ±0.41 a-c  | 43.3±2.4 d    | 28.5±0.9 de                | 11.4±0.5 d              | 65.7±1.14 c      |
| BS76         | 7.33±0.31 bc    | 45.0±2.4 b-c  | 30.5±2.1 cd                | 12.1±0.9 cd             | 71.9±0.96 ab     |
| Average      | $7.78 \pm 0.48$ | 46.4±3.4      | 31.6±3.7                   | 12.5±1.7                | 70.73±3.52       |

\*Control: without bacteria inoculation or mineral fertilizers; N fertilizer (500 mg N per roted cutting in the form of ammonium nitrate 33%); NPK fertilizer (500 mg N + 100 mg P + 200 mg K per roted cutting in form of compound NPK 25-5-10 fertilizer); Pantoea agglomerans PA58;, Bacillus subtilis BS21, Roseomonas fauriae RF272; Achromobacter xylosoxidans denitrificans AXD47, Paenibacillus polymyxa PP243, Bacillus atrophaeus BA59, Arthrobacter citreus AC484, Rhizobium radiobacter RR112, Bacillus pumilus BP70, Pseudomonas agarici PA71, Bacillus subtilis BS76.

\*\* Different letters within the same column indicate significant differences according to Duncan's Multiple Range Test (p≤0.01)

Among the treatments tested, except for AXD47, BA59 and PA71, all treatments tested significantly increased rott fresh and root dry weight of tea sapling. Among the various treatments tested, *Rhizobium radiobacter* RR112 and *Arthrobacter citreus* AC484 caused the maximum enhancement in fresh and dry root weight in tea plant, while the *Roseomonas fauriae* RF272 and *Bacillus pumilus* BP70 was the most effective promoter of plant height. Inoculation with the strains PA58, AXD47, PP243, BA59, AC484, RR112, BP70, BS76 and NPK fertilizer increased lateral root number in tea plants, whereas only the isolates PP 243, AC484, RR112, BP70, and BS76 increased the highest root length of tea seedlings. In three years highest root:shoot dry weight and lateral root number were obtained from AC484 inculations followed by PP 243 inoculations while the lowest root:shoot dry weight as recorded for N fertilizer and PA71 inoculation (Table 3).

|             | Doot fresh weight (g     | Doot daw woight (g      | Highest reat | No of lotorol | Destuchest dwg    |
|-------------|--------------------------|-------------------------|--------------|---------------|-------------------|
| Treatments* | Koot fresh weight (g     | Root dry weight (g      | figuest root | No of fateral | Root:shoot ary    |
|             | sapling <sup>-1</sup> )* | sapling <sup>-1</sup> ) | length (cm)  | root          | weight            |
| Control     | 14.23±0.62 f             | 5.70±0.19 f             | 33.3±2.2 de  | 13.8±3.8 f    | 0.478±0.016 bc    |
| NPK         | 18.26±0.25 b-d           | 7.62±0.20 a-d           | 29.0±2.6 f   | 21.3±2.5 bc   | 0.475±0.014 bc    |
| Ν           | 16.26±0,53 e             | 6.57±0.12 e             | 30.3±2.2 ef  | 16.3±1.3 d-f  | 0.431±0.025 d     |
| PA58        | 19.44±0.77 ab            | 7.65±0.54 a-c           | 35.3±1.0 b-d | 20.5±0.6 cd   | 0.506±0.030 ab    |
| BS21        | 16.83±0.15 de            | 6.78±0.21 de            | 29.8±1.0 f   | 16.0±1.8 ef   | 0.472±0.014 b-d   |
| RF272       | 19.42±1.17 ab            | 7.48±0.60 a-d           | 35.8±1.0 a-d | 17.5±1.9 c-f  | 0.483±0.027 a-c   |
| AXD47       | 14.29±0.67 f             | 5.71±0.17 f             | 28.9±1.4 f   | 18.5±2.4 c-e  | 0.502±0.020 ab    |
| PP 243      | 18.94±1.13 a-c           | 7.84±0.71 ab            | 36.9±1.1 a-c | 25.3±1.5 ab   | 0.504±0.032 ab    |
| BA59        | 15.65±0.67 ef            | 5.93±0.19 f             | 33.8±1.5 cd  | 21.0±2.2 c    | 0.470±0.011 b-d   |
| AC484       | 19.90±1.05 ab            | 7.67±0.28 ab            | 38.7±2.0 a   | 26.5±1.3 a    | 0.525±0.010 a     |
| RR112       | 20.33±0.89 a             | 8.32±0.41 a             | 37.7±1.3 ab  | 21.5±2.1 bc   | 0.503±0.036 ab    |
| BP70        | 19.38±0.82 ab            | 7.30±0.41 b-d           | 37.2±1.2 a-c | 18.3±1.9 с-е  | 0.447±0.0 cd      |
| PA71        | 15,62±1.59 ef            | 5.80±0.61 f             | 35.5±1.0 a-d | 15.5±2.1 ef   | 0.492±0.08 ab     |
| BS76        | 17.41±1.18 с-е           | 6.81±0.42 с-е           | 36.9±1.7 a-c | 17.9±1.7 с-е  | 0.495±0.012 ab    |
| Average     | 17.58±2.19               | 6.94±0.94               | 34.3±3.6     | 19.3±4.0      | $0.484{\pm}0.030$ |

**Table 3.** Effect of PGPR and fertilizer applications on root growth parameters of tea cv. Hamzabey in the pot experiment during the 2013 season

\*Control: without bacteria inoculation or mineral fertilizers; N fertilizer (500 mg N per roted cutting in the form of ammonium nitrate 33%); NPK fertilizer (500 mg N + 100 mg P + 200 mg K per rooted cutting in form of compound NPK 25-5-10 fertilizer); Bacterial strains are explained in Table 1-2.

\*\* Different letters within the same column indicate significant differences according to Duncan's Multiple Range Test (p≤0.01)

#### Discussion

Bacterial association with root growth is an important new field of agricultural research. Inoculation with multi-traits bacteria increased plant height, trunk diameter, shoot and leaf weight, root fresh and dry weight, lateral root number, chlorophyll value, and Root:shoot dry weight in tea sapplings compared with the control. The positive effects of PGPR on plant growth were correlated with remarkable changes root growth and morphology, namely increasing the lateral root and root hair number and length. Previous studies also demonstrated the application of PGPR in soil have resulted in significant increase in growth of young tea bushes (Sharma et al., 2002; Chakraborty et al., 2006, 2012; Saikiaet al., 2011; Çakmakçı et al., 2014) and help in the reduction of the use of chemicals in tea plantations (Chakraborty et al., 2009). The results suggest that bacterial hormones (especially IAA) play a major role in the development of the plant root during the early stages of growth. PGPR caused high root weight, total root numbers and encouraged adventitious root formation but it's were strongly dependent on plant species. Indeed, in tea cultivation, fine roots play a vital function in determining a plant's ability to compete for soil nutrients (Razaq et al., 2017). The positive effects of PGPR on plant growth were correlated with remarkable changes root growth and morphology, namely increasing the lateral root and root hair number and length. The elongation or proliferation of root systems has been suggested to be one of the mechanisms by which PGPR stimulate plant growth. The experiment revealed that the PGPR inoculation was effective treatment to improve the root growth and change in root morphology; bacteria tested during our study have great potential being formulated and used as biofertilizer.

### 4. CONCLUSION

Selected bacterial strains stimulated overall plant growth, including shoot development, plant height, trunk diameter, leaf yield, chlorophyll, and root growth parameters of Turkish registered tea clones Hamzabey. Plant growth-promoting rhizobacteria may change root growth, increase root weight, proliferation of lateral and adventitious roots number and may have an influence on nutrient uptake potentials.

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### REFERENCES

- Chakraborty U, Chakraborty B & Basnet M (2006). Plant growth promotion and induction of resistance in *Camellia sinensis* by *Bacillus megaterium*. Journal of Basic Microbiology, 46: 186-195.
- Chakraborty U, Chakraborty BN & Chakraborty AP (2012). Induction of plant growth promotion in *Camellia sinensis* by *Bacillus megaterium* and its bioformulations. World Journal of Agricultural Sciences, 8:104–12.
- Chakraborty U, Chakraborty BN, Basnet M, Chakraborty AP (2009). Evaluation of *Ochrobactrum anthropi* TRS-2 and its talc based formulation for enhancement of growth of tea plants and management of brown root rot disease. Journal of Applied Microbiology, 107: 625–634.
- Çakmakçı R (2016). Screening of multi-trait rhizobacteria for improving the growth, enzyme activities, and nutrient uptake of tea (*Camellia sinensis*). Communications in Soil Science and Plant Analysis, 47 (13-14): 1680-1690.
- Çakmakçı R, Dönmez MF, Ertürk Y, Erat M, Haznedar A & Sekban R (2010). Diversity and metabolic potential of culturable bacteria from the rhizosphere of Turkish tea grown in acidic soils. Plant and Soil, 332 (1-2): 299-318.
- Çakmakçı R, Ertürk Y, Atasever A, Kotan R, Erat M, Varmazyari A, Türkyılmaz K, Haznedar A & Sekban R (2014). Development of plant growth-promoting bacterial based bioformulations using solid and liquid carriers and evaluation of their influence on growth parameters of tea. 9<sup>th</sup> International Soil Science Congress on the Soul of the Soil and Civilization, 14-16 October 2014, Side, Book of Proceedings, 801-808.
- Çakmakçı R, Ertürk Y, Sekban R, Haznedar A & Varmazyari A (2013). The effect of single and mixed cultures of plant growth promoting bacteria and mineral fertilizers on tea (*Camellia Sinensis*)growth, yield and nutrient uptake. 1st Central Asia Congres on Modern Agricultural Tecniques and Plant Nutrition. 01-03 October, 2013, Soil Water Journal, Secial Issue for AGRICASIA, 2 (1): 653-662.
- Gebrewold AZ (2018). Review on integrated nutrient management of tea (*Camellia sinensis* L.). Cogent Food & Agriculture, 4: 1543536.
- Han WY, Ma LF, Shi YZ, Ruan JY & Kemmitt SJ (2008). Nitrogen release dynamics and transformation of slow release fertiliser products and their effects on tea yield and quality. Journal of the Science of Food and Agriculture, 88: 839-846.
- Hirono Y, Watanabe I & Nonaka K (2009). Trends in water quality around an intensive tea-growing area in Shizuoka, Japan. Soil Science and Plant Nutrition 55: 783–792.
- Kamau DM, Jhj S, Oenema O & Owuor PO (2008). Productivity and nitrogen use of tea plantations in relation to age and genotype. Field Crop Research, 108 (1): 60–70.
- Liu Z, Yang J, Yang Z & Zou J (2012). Effects of rainfall and fertilizer types on nitrogen and phosphorus concentrations in surface runoff from subtropical tea fields in Zhejiang, China. Nutrient Cycling in Agroecosystems, 93: 297-307.
- Razaq M, Salahuddin Shen H-L, Sher H & Zhang P (2017): Influence of biochar and nitrogen on fine root morphology, physiology, and chemistry of *Acer mono*. Scientific Reports, 7: 5367.
- Ruan J, Haerdter R & Gerendás J (2010). Impact of nitrogen supply on carbon/nitrogen allocation: a case study on amino acids and catechins in green tea (*Camellia sinensis* (L.) O. Kuntze) plants. Plant Biology, 12 (5): 724–734.
- Saikia DN, Sarma J, Dutta P K & Baruah DK (2011). Sustainable productivity of tea through conservation of bio-mass, addition of bio-fertilizers and reduction of inorganic fertilizer. Two and a Bud 58, 109-117.
- Sharma KL, Sharma DK. & Sharma G (2002). Long term response of integrated nitrogen nutrition with bioresources on the yield of China hybrid tea (*Camellia sinensisL.*) grown in North West Himalayas. In: Sreedharan, K., Vinod Kumar, P. K., Jayarama, Chulaki, B. M. (Eds.), Proceedings of the 15th Plantation Crops Symposium, 386–391.
- Tokuda S & Hayatsu M (2004). Nitrous oxide flux from a tea field amended with a large amount of nitrogen fertilizer and soil environmental factors controlling the flux. Journal of Soil Science and Plant Nutrition, 50 (3): 365–374.



# Comparison of Wine Aroma Compounds Produced by Local Yeasts Isolated from Purcari Wine Center

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Abstract: In recent years, consumers consider that the taste and aroma of wine are the main characteristics in the choice of wine, that define it quality. Flavor is the most important distinguishing characteristic of wine. Wine flavor is classified according to the sources of the different compounds contributing to it. This is including varietal flavor, fermentative flavor and post fermentative flavor. The aim of this study is to determine specific enological characteristics of local yeasts, potential differences in production of volatile compounds between them and their influence on wine quality. Samples of young wine were analyzed for some aroma compounds. At the same time wines were subjected to sensory evaluation. The results showed a positive influence of local yeasts on chemical and sensory properties of dry red and white wines and contribute to the formation of a very complex flavor and suggest that thay may be used to produce" Purcari" wine with a unique and desirable aroma.

Keywords: Local yeasts, aroma, fermentation, sensory evaluation.

## **1. INTRODUCTION**

The utilization of isolated local strains is an important strategy for keeping the quality and assuring the reproducibility of wine features. The utilization of strains isolated from specific wine centers could be even more interesting because of their high adaptation to their own climatic conditions and grapes. Even more, these strains are usually associated to particular wine characteristics that frequently identify specific wines and centers (Thais et.al., 2006).

Previous studies have shown that yeast strains have a large impact on wines chemical complex (Ripon, 1997) and found that the volatile composition could be an alternative method for characterization of yeasts used in wine production (Mar Vilanova, 2005).

Some authors use synthesis of different amounts of acetylmethylcarbinol, 2,3-butadiene or acetic acid as a base for studying the genetic strains variability of the genus Saccharomyces, which can serve as a way to improve the quality of wine or fermentative properties of yeasts (Romano, 1998).

During alcoholic fermentation yeasts form and other alcohols, except ethanol, so-called higher alcohols, mainly represented by n-propanol, isobutanol, isopentanol etc. They may derive directly from the corresponding amino acids or sugars in the environment. Each strain of yeast Saccharomyces genus has its own capacity to produce these secondary compounds of fermentation. Yeast strains producing of higher alcohols behave differently depending on musts studied (Pomohaci, 2000, Saint Crieg, 1999).

Esters formation in wine is made in two ways: biological esterification occurs during alcoholic fermentation, malolactic and / or esterification of acetic by chemical or enzymatic reactions that occurs very slowly during storage / aging of wine. Both ways may occur depending on the technological and almost equal participation in the formation of esters in wine (Tirdea, 2007).

## 2. MATERIALS AND METHODS

The research was carried out in the Laboratory of Biotechnology and Wine Microbiology, Scientific-Practical Institute of Horticulture and Food Technologies (Moldova).

### **Strains of Yeast**

In the present paper were studied local yeast strains (FNFTP-1, FNFTP-6, F-75-FTP-5, Ch75P-3ÎF), isolated in the wine center "Purcari". Studies performed on the morphological, cultural and physiological-biochemical properties permitted, using identifier by Kudreavţeva (Burian, 2003), establishing that yeast strains identified belong to the species Saccharomyces cerevisiae. As a control were studied industrial dry active yeast: Oenoferm freddo, (Germany).

As a raw material was used grape must of the variety Aligote harvest of 2017. Initial physico-chemical characteristics of the grape are presented in Table 1.

| Table 1 | . Physico-chemical | characteristics of | f the grape | (harvest 2017) |
|---------|--------------------|--------------------|-------------|----------------|
|---------|--------------------|--------------------|-------------|----------------|

| Grape variety | Sugar, g/L | Titratable acidity, g/L tartaric acid | pН   | Potential OR, mV |
|---------------|------------|---------------------------------------|------|------------------|
| Aligote       | 210,0±0,5  | 6,8±0,1                               | 3,09 | 216,9            |

### **Chemical Analysis**

The analyses of wine aroma compounds were performed by gas chromatography method.

### **Sensory Analysis**

The wines from 2017 harvest season were subjected to sensory evaluation by a panel comprising 10 members of the Scientific-Practical Institute of Horticulture and Food Technologies, all of them highly experienced in wine sensory testing.

Wines of each yeast treatment were compared by a paired sample test to determine aroma differences among different varieties.

## 3. RESULTS AND DISCUSSION

### **Chemical Analysis**

Comparative analysis of aromatic content of Aligote dry white wine (harvest 2017) achieved by classical technology using different yeast strains allowed the establishment of significant differences. The results obtained are shown in Table 2.

The results presented in Table 2 demonstrate that the content of volatile substances in dry white wines Aligote (harvest 2017) varies depending on the strain of yeast used.

| Compounds               | Strain yeast |         |            |           |                            |  |  |
|-------------------------|--------------|---------|------------|-----------|----------------------------|--|--|
| Compounds               | FNFTP-1      | FNFTP-6 | F-75-FTP-5 | Ch75P-3ÎF | Oenoferm freddo, (Germany) |  |  |
| Acetic aldehyde         | 10,4         | 13,8    | 12,5       | 12,7      | 13,4                       |  |  |
| Ethyl acetate           | 19,8         | 24,9    | 20,4       | 23,4      | 22,6                       |  |  |
| Methyl alcohol, g/L     | 0,02         | 0,02    | 0,02       | 0,02      | 0,015                      |  |  |
| Higher alcohols         |              |         |            |           |                            |  |  |
| 2-butanol               | <0,5         | <0,5    | <0,5       | <0,5      | <0,5                       |  |  |
| n-propanol              | 9,9          | 10,1    | 10,0       | 12,9      | 10,1                       |  |  |
| Isobutanol              | 20,7         | 20,3    | 22,4       | 31,3      | 23,0                       |  |  |
| n-butanol               | <0,5         | <0,5    | <0,5       | <0,5      | <0,5                       |  |  |
| Isopentanol             | 141,9        | 151,1   | 155,6      | 161,1     | 148,0                      |  |  |
| $\Sigma$ Higher alcohol | 173,7        | 182,5   | 189,0      | 206,3     | 182,1                      |  |  |

Table 2. Content of volatile substances (mg / L) in dry white wines Aligote

Acetic aldehyde concentration limit values vary from 10.4 up to 13.8 mg / L. Obviously yeast strain has a significant influence on the content of acetic aldehyde, which can be explained by the specific characteristics of each yeast strain to eliminate relatively large or small quantities of this substance. For example: the use of yeast strain FNFTP-1 acetic aldehyde concentration is 10.4 mg / L (minimum), and the use of yeast strain FNFTP-6 acetic aldehyde concentration is 13,8 mg / L (maximum).

A less significant influence yeasts have studied the content of n-butanol and 2-butanol, where the determined values were below 0.5 mg / L.

Isobutanol concentration in dry white wines vary depending on the strain type of yeast used and the variation range of values is quite wide and is up from 20.3 to 31.3 mg/L.Maximal concentrations of isobutanol have been established in yeast strain used Ch75P-3ÎF.Changes in concentrations of n-propanol in wine raw material is within the range 9.9 to 12.9 mg/L.

Isopentanol concentration in dry white wines studied is about 60% of the sum of the higher alcohols, and the difference value is 19.2 mg / L. The highest concentration of isopentanol was found in wine achieved with local yeast strain (Ch75P-31F).

Another important component that forms the must fermentation is ethyl acetate, which directly influence the organoleptic properties of wine obtained.

It is known that ethyl acetate is part of the group mean fatty acid esters, and most of the ester group enanthic assign a strong sense of fruit wine.

Therefore, ethyl acetate directly participates in the formation of wine aroma obtained. In addition, ethyl acetate affects the taste of wine. At concentrations higher than the olfactory perception, he gives a stringent flavor. All wines contain healthy ethyl acetate, formed during fermentation, up to 160 mg / L.

In our case, values of ethyl acetate are in the range of 19.8 to 24.9 mg / L, respectively lowest concentration was found in wine obtained by using local strain yeast (FNFTP-1) and highest in wine obtained by using yeast strain FNFTP-6, but this difference is insignificant in this period.

The analysis of complex volatile dry white wines studied, we can conclude that in all wines the methyl alcohol content is about 0.02 mg/L, which proves that nature of yeast does not affect methyl alcohol concentration.

### **Sensory Analysis**

The results of sensory evaluation of wines are shown in figure 1, and even without significant differences they indicate a substantial effect on the aroma of "*Purcari*" wines as a result of fermentation with different local yeast strains, which is in accordance with literature data had the most typical aroma, whereas aroma of Oenoferm Freddo wines was not so typical. According to our results, more intense aroma of local yeasts wines was due to numerous factors, and we suppose that one of them could be a lower amount of total higher alcohols. The aroma differences among produced wines could also be linked to a different yeast  $\beta$ -glucosidase activity. Laffort et. al. suggested that specific yeast strain  $\beta$ -glycosidases can affect the aroma of wines



Figure 1. Sensory profiles of white wine Aligote obtained with different yeasts strain.

### 4. CONCLUSION

The current study reports the use of local strain yeasts in winemaking and compared with imported Oenoferm Freddo, our results indicated interesting enological properties of local FNFTP-1 and FNFTP-6 strains. Higher alcohols, esters, aldehydes, volatile acids and other substances formed during fermentation of must in the manufacture of dry white wine contribute to the formation of complex flavors. Specified yeast strains are able to positively influence the aromatic content of wines. Our results show that knowledge of the biochemical properties of yeast strain used for producing dry white wines can have a decisive role.

In the future this yeast should be better studied and selected.

#### REFERENCES

Burian N., 2003. Practicescaia microbiologhia vinodelia.Simferopoli.Tavrida.

- F. Laffort, H. Romat, P. Darriet, 1989. Revue des Oenologues, 53: 9–12
- Mar Vilanova, Isabelle Massneuf-Pomarede, 2005. Characterization of yeast strains from Rias Baixas (NW Spain) and their contribution to the fermentation of Albarino wine. Annals of Microbiology, 55 (1): 23-26.
- Pomohaci N., Sârghi C., Stoian V., Valeriu V. Cotea, M. Gheorghiță, I. Nămoloșanu, 2000. Enologie, volumul 1. Ceres, București.
- Riponi, C., Carnacini, A., Antonelli, A., Castellari, L. & Zambonelli, C.,1997 . Influence of yeast strain on the composition of wines for the production of brandy. Journal of Wine Research, 8: 41–55.
- Romano, P., Brandolini, V., Ansaloni, C. & Menziani, E., 1998. The production of 2,3-butanediol as a differentiating character in wine yeasts. World Journal of Microbiology and Biotechnology, 14: 649–653.
- Saint Crieg De G., Nathalie Provost C., Vivas N., 1999. Comparative study of poliphenol scavenging activities assessed by different Methods. J. Agr. and Food Chem., V. 47, № 2: 425-431.
- Thais M. Guimaraes, Danilo G. Moriel, Iara P.Machado et.al., 2006. Isolation and characterization of Saccharomyces cerevisiae strains of winery interest. Brasilian Journal of Pharmaceutical Sciences, vol.42, n.1:119-126.

Țîrdea C., 2007. Chimia și analiza vinului. Editura "Ion Ionescu de la Brad", Iași.





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