AGRICULTURE AND ENVIRONMENT

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INTRODUCTION

PREFACE

Since the beginning of humanity, agricultural activities have been one of the most significant requirements for the continuity of life. Developing technology, ever-increasing population, industry, and the resulting increase in needs have made agricultural activities gain more importance both economically and in terms of people's nutrition. In developing countries, agricultural production and marketing provide almost more than half of the share of the country's economy. While agriculture was the largest economic share in developed countries in the 18th century, this share began to decline with the development of industry and technology. However, the continuous increase in the population and the global economic contraction in recent years indicate that the importance of agricultural production and marketing will gradually increase. Sustainable agricultural management will become one of the basic mechanisms of countries. The integration of many factors can achieve this sustainable management. The most important of these factors is the environment. Sustainable environmental management brings sustainable agriculture. Suppose the environment, which is already under pressure from naturally changing climate conditions, industrial production, and anthropogenic waste, is also faced with the stress of agricultural activities. In that case, it will not be possible to establish sustainable management.

The studies on agriculture and the environment carried out by many valuable academicians in this book will contribute to the areas whose importance is emphasized above. They will guide their readers in sustainable agriculture-environment management.

Editors

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CHAPTER 1

FRUIT GROWING AT HIGH ALTITUDE

Ümmügülsüm ERDOGAN

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INTRODUCTION

Altitude refers to the height of a point on earth above sea level. How high a settlement unit is located above sea level is one of the main elements that determine the geographical conditions of that settlement. In this context, the concept of elevation can affect both the physical and human characteristics of a place directly and many geographical elements indirectly (Alaeddinoğlu, 2014).

The same landforms can be located at different elevations and different landforms can be located at the same elevations. A significant part of the world geography is above 1000 m. Elevation affects many features such as climate, hydrography, soil formation and quality, diversification and distribution of vegetation (Elibüyük and Yılmaz, 2010). High or low elevation and slope are of great importance for agriculture, animal husbandry, forestry, industry and nature tourism.

High altitude is also effective in terms of solar radiation retention, increase in day-night temperature difference, lower relative humidity, and less or more water available to plants depending on the region (Korkutal et al., 2012). Highlands above 1500 m above sea level constitute almost a quarter of the world's land surface (Mengist et al., 2020) and these altitudes may be escape zones for global warming.

The average height above sea level in Turkey is 1141 m. High altitude areas (>1000 m) constitute 57% of the total area (Table 1). The high plateau region of Anatolia rises steadily eastward and is divided into valleys formed by 15 rivers, including the Tigris and Euphrates Rivers, which originate in eastern Anatolia and flow south through Syria and Iraq towards the Persian Gulf (FAO 2019). Turkey has an average slope of 17.3% and a maximum slope of 388%. The Eastern Anatolia Region has the highest elevation and the Marmara Region the lowest. 11% of Turkey is below 250 m, more than 7% is between 250-500 meters, almost 18% is between 1000-1250 m, and 3% is above 2500 m (Elibüyük and Yılmaz, 2010).

Table 1. Distribution of the area according to altitude (Türkiye) (Karagöz, 2000)

Height (m)	Proportion to total area (%)
0-500	18
500-1000	25
1000-1500	30
1500-2000	16
>2 000	11

There are different assumptions about the limit of high altitude, and areas with an altitude of 1500 m to 3500 m are commonly accepted as high altitude by measuring the oxygen distribution and air pressure in the air (Belen, 2012). In Turkey, there are 25 provinces with settlement centers at an altitude of

1000 m and 8 provinces with altitudes higher than 1500 m, namely Bitlis (1545 m), Bayburt (1550 m), Ağrı (1640 m), Van (1661 m), Hakkari (1720 m), Kars (1750 m), Erzurum (1893 m) and Ardahan (2200 m).

1. Effect of high altitude on climate

As the altitude increases, climatic conditions become different. This is due to the effects of increasing altitude on climatic elements such as temperature, humidity, precipitation, radiation and wind. Altitude has a significant effect on minimum and maximum temperatures, especially for orchards located in hilly and mountainous areas. Altitude is perhaps the most important criterion for the suitability of an orchard location, as frosts and freezing temperatures can significantly reduce profitability. There are topographical influences on air temperature (Geiger et al., 1965). Under conditions of radiational cooling, the earth, thanks to calm winds and clear skies, loses heat into the void and cools the air layer. If the vineyard is on a slope, cold and relatively dense air moves downhill. This movement can be more pronounced in mountainous areas and can even produce localized winds. The collapsing cold air displaces warm air towards higher altitudes and creates thermal exchange. Collapsing cold air collects at the base and can form frost lakes. Eighty percent or more of spring frosts and most mid-winter frosts are largely caused by radiational freezing events. Vineyards located in low-lying frost lakes are more exposed to spring and fall frosts and winter cold damage than those at higher altitudes. The effects of altitude increase on climate elements are as follows:

1.1. Temperature: Atmospheric temperatures decrease from the equator to the north and south, and at the same latitude, as the altitude increases. At high altitudes, temperature differences between day and night are high. Among the climatic changes that occur with increasing altitudes that affect the growth, development and quality of fruit trees, temperature decreases by 1 °C in dry air and 0,6 °C in humid air for every 100 m above sea level (Benavides et al., 2017). As altitude increases, the vegetation period shortens, but at higher altitudes, plants can start physiological activity at lower temperatures and reach harvest maturity before the early fall frosts (Poincelot, 1979).

1.2. Humidity: As the altitude increases, the decreasing temperature decreases the evaporation from the water surface and the water holding capacity of the air. Thus, the humidity of the environment decreases. Different temperatures are observed in areas at different altitudes at the same latitude. The partial pressure of gases such as CO_2, O_2 and N_2 and water vapor decreases (Fischer and Orduz-Rodríguez, 2012).

1.3. Precipitation: In the temperate climate zone, as the altitude increases in any ecology, the atmospheric temperature decreases and the amount of precipitation increases. As the airflow passes through high altitude regions, it leaves most of the moisture it carries in the form of precipitation.

1.4. Light intensity and radiation: At high altitudes, light intensity, visible UV, infrared radiation and wind increase with altitude. Greater solar radiation at high altitudes increases soil temperature and promotes plant growth (Fischer and Orduz-Rodríguez, 2012). Since the atmospheric layer filtering solar radiation is thinner at higher elevations, UV radiation increases by 10 to 12% for every 1000 m of altitude increase (Benavides et al., 2017). Increased light intensity increases photosynthesis, and increased photosynthesis increases secondary metabolism. In addition, increased light intensity increases resveratrol synthesis (Bajda, 2007).

1.5. Wind speed: Wind speed increases with increasing altitude (Fischer and Orduz-Rodríguez, 2012). Air movement occurs as a result of the expansion and rise of warm air at low altitude and the collapse of cold air at high altitude to areas at lower altitudes (Aslantaş and Karabulut, 2007).

1.6. Oxygen-Carbon dioxide content: The proportion of oxygen in the air does not change as the altitude increases, but oxygen molecules become rarer as the altitude increases due to the decrease in pressure. As the altitude increases, the density of the gases that make up the atmosphere decreases by 1 mm for every 10.5 m due to gravity.

2. Effect of high altitude on fruit quality

2.1. Plant development and disease resistance

Increasing UV radiation with increasing altitude causes stunting, small leaf area (Fischer et al., 2012a), thickening of the cuticle, epidermis and leaves (Fischer and Miranda, 2021), pubescence and intense coloration by synthesizing protective pigments (García-Muñoz et al., 2021). Fruits grown at high altitude have a thicker cuticle and epidermis, making them less susceptible to pathogens and insect pests (Aslantaş and Karabulut, 2007; Fischer et al., 2016). Campos and Quintero (2012) found that passion fruits grown in mountainous regions of Colombia had a thicker epidermis and were more resistant to anthracnose than those grown at lower altitudes.

At high altitudes, UV light reduces auxin production (Fischer and Melgarejo, 2014) and gibberellin synthesis in internodes (Buchanan et al., 2015), resulting in smaller plants, smaller leaf area (Fischer et al., 2012) and thicker leaves (Fischer and Miranda, 2021).

Gooseberry plants growing at 2300 m in Boyacá (Colombia) have been found to form a more superficial root system as altitude increases to 2690 m to better utilize the sun's heating of the soil (Fischer et al., 2007), the same plant can increase the number of leaf stomata to compensate for lower CO_2 and O_2 concentrations as elevation increases (Fischer and

Melgarejo, 2020) and can produce dense pubescence on all aerial surfaces to counteract high UV radiation and nighttime cold (Fischer et al., 2021).

2.1.1. Fruit size: It has been determined that the fruits become smaller and the yield of hard-shelled fruits decreases with increasing altitude in species and cultivars that require high temperature sum for ripening their fruits (Balcı, 2002; Dinis et al., 2011). In a study aiming to evaluate the effect of altitude on phenological and fruit quality characteristics of apricot genotypes (in Trans-Himalaya), it was found that for every 100 m increase in altitude, flowering was delayed by 3.3 days and fruit ripening by 7.1 days, fruit weight decreased by 0.5 g and fruit moisture content by 1.9%, and fruit TSS increased by 1.2 °Brix (Naryal et al., 2020). In Indonesia, the fruits of the 'Crystal' guava variety grown at 550 m altitude were harder, while those grown at 200 m altitude were softer and heavier (Musyarofah et al., 2020).

2.1.2. Fruit shape: The shape of fruits of plants of the same variety grown at different altitudes varies depending on altitude. Where the flowering period is cooler, fruits and fruit stalks are longer (Noe and Eccher, 1993; Eccher and Noe, 1996). Low temperatures, especially after flowering, increase the number of cells in the flower nose of the fruit and stimulate its development. Excessive synthesis of cytokinins during the cool flowering period lengthens the fruit axis. In areas with warm days and nights, the fruit remains flattened, whereas at high altitudes with cool and cold nights, the fruit is elongated. In Barlett pear grown in the Sacramento valley in California, fruit shape is flattened and fruit length increases with increasing altitude (Eccher and Noe, 1993).

2.1.3. Composition of the fruit: Fischer et al. (2010) are highly temperature dependent. As a result of the low night temperatures at high altitudes, plants consume less of the photosynthetic products they produce during the day due to the lack of suitable temperature at night. Instead of structural substances, soluble sugars and proteins are stored for cold resistance. As the altitude increases, the firmness of the fruits increases, the shelf life is prolonged and they have better aroma (Osterloh et al., 1996). Walton and Jong (1990) reported that fruit ripening in kiwifruit orchards established with Hayward variety at 3 different altitudes in California conditions differed according to the regions and sugar/acid ratio was higher in fruits grown at high altitudes than in fruits grown in valleys. Voronkov et al. (2019) found that the levels of phenolic substances and polyunsaturated fatty acids in apple peel increased from 300 m to 1200 m above sea level in the Caucasus. The amount of phenols and unsaturated fatty acids increases with increasing elevation.

Phenols protect fruits against high UV radiation due to their strong antioxidant effects, while unsaturated fatty acids maintain the fluidity of fruit cell membranes within the physiological range (Voronkov et al., 2019). Phenol content, which is a very important protection mechanism against UV radiation (Cheynier et al., 2013), was found to be higher in apple (Kumar et al., 2019), cherry (Faniadis et al., 2010), peach (Karagiannis et al., 2016) and blueberry (Zeng et al., 2020) fruits grown at high altitude.

In a study conducted on apples in China, it was determined that the sugar content of fruits increased with increasing altitude, while malic acid and water content decreased (Xingjun et al., 2004). Gooseberries grown at high altitude in Colombia were found to contain higher sucrose and carotene content, while citric acid decreased with increasing altitude (Fischer, 2000). In strawberry, blackberry and blueberry fruits, a decrease in TTA, an increase in TSS and an increase in TSS/TTA ratio were determined with increasing altitude (Famiani et al., 2015). In Tahitian lemon and orange fruits, measurements at 336 m and 1038 m showed a decrease of about 15% in juice content with increasing altitude (García-Muñoz et al., 2021). The thickness of the epidermis increased in oranges grown at high altitude (Ayer and Shrestha, 2018) and the hardness of the epidermis increased in bananas (Bugaud et al., 2006).

The height of the plants above sea level may affect their vitamin content. In general, plants grown at higher altitudes contain more ascorbic acid than those grown at lower altitudes (Eşitken, 2006).

2.1.4. Fruit color and aroma: As altitude increases, increased light intensity causes fruits to be darker in color (Tonietto and Carbonneau, 2004; Fischer and Orduz-Rodríguez, 2012). In a 3-year study conducted in the Douro valley in Portugal, it was determined that the amount of anthocyanins in Touriga Nacional and Touriga Francesca varieties increased with increasing altitude. Climatic conditions at high altitude encouraged the formation of grapes with higher anthocyanin monoglycoside content (Mateus et al., 2003). It has been reported that low temperatures and high temperature differences between day and night at high altitudes cause the breakdown of chlorophyll and promote phenyl propanoid enzyme activity, which plays an important role in anthocyanin synthesis. In addition, it was observed that the lenticels on the fruits of Golden Delicious and Starkrimson varieties were less and smaller in the high altitude region. It was reported that lenticel diameter was positively correlated with anthocyanin content, while lenticel density was negatively correlated with anthocyanin content (Xingjun et al., 2004).

Increasing light intensity and UV radiation at higher altitudes cause the fruits to be darker and more intensely flavored. While the skin of Granny Smith type apple grown at an altitude of 1200 meters in Erzincan was completely green, it was found that the fruits were red when this species was grown in Erzurum at an altitude of 1850 meters (Aslantaş and Karabulut, 2007). It is also observed that grapes produced at high altitude have redder colors due to the high tannin and anthocyanin content (Korkutal et al., 2012). It has been reported that orange color in mandarins (Ayer and Shrestha, 2018), red color in strawberries (Pérez de Camacaro et al., 2017) and pomegranates (Al-Kalbani et al., 2021), peaches (Karagiannis et al., 2016), blueberries (Zeng et al., 2020) and grapes (Oliveira et al., 2019) grown at high altitude are due to the increase in anthocyanin concentrations.

2.1.5. Fruit ripening and storage life: Temperature, one of the most important climatic variables, affects the growth and development of fruit trees by regulating the length of the different phenological periods, i.e. fruit development is prolonged at high altitudes due to lower temperatures. (Parra-Coronado et al., 2015; Ramírez et al. 2018; Mayorga et al., 2020). The flowering of fruit trees of the same variety is delayed by an average of 1 day for every 33 m of altitude increase. The decrease in temperature due to altitude prolongs the vegetation period of plants and the flowering period of fruits. Although the ripening time of fruits is a hereditary factor, the ripening time for a particular variety is shorter in hot places at low altitude. Fruits grown at high altitudes, which are not problematic for cultivation, have better fruit stability, longer shelf life and better flavor resulting in late ripening. Apples grown at higher altitudes have better storage capacity than those grown in valleys (Eccher and Noe, 1993).

The lower the temperature during the growing season, the later the fruit ripens (Moretti et al., 2010). In Jamaica, the growth cycle of the Lacatan banana variety at sea level was 13 months, while at altitude the growth period increased by one month for every 100 meters of altitude (Posnette, 1980). Cape gooseberries ripen in 66 days at 2300 m and 75 days at 2690 m altitude (Fischer et al., 2007), feijoas grown at 1800 m altitude ripen in 155 days and at 2580 m altitude ripen in 180 days (Parra-Coronado et al., 2015).

2.1.6. Yield: The yield of fruit trees depends on environmental conditions as well as genetic structure. In countries where the need for chilling in fruits cannot be met sufficiently, high altitudes are advantageous. Decreasing vegetation period with increasing altitude causes fluctuations in yield even in varieties that do not show periodicity in their genetic structure.

3. CONCLUSION

The change in climatic elements such as temperature, humidity, radiation, pressure and wind due to high altitude plays an important role in crop production. In terms of human nutrition and health, the nutritional value of food ingredients is richer in plant products grown at high altitude and the relationship between high altitude and quality has been revealed by many scientific studies.

High altitudes are special areas where air pressure, oxygen content and climatic conditions differ. The decreasing air pressure and the amount of oxygen as the altitude increases have an impact on all kinds of living life. These effects of high altitude on living life bring along some advantages at habitable altitudes. The fact that living at high altitude is healthier and reduces the risk of certain diseases, and the quality and flavor superiority of agricultural products produced at high altitude have been the subject of many studies. It has been scientifically proven that these products have higher nutritional values, longer shelf life, better taste, more natural and preventive effects on disease risks. In the food market, where product diversity is quite high, there has been a trend towards foods that claim health, beauty and naturalness in recent years. These products, which create a permanent place in the minds of consumers by making a difference with their quality, taste and naturalness, are branded and the tendency to pay more for these products is increasing.

The effect of altitude on the quality components of fruit trees depends firstly on the species and variety, and secondly on whether the climatic conditions are within the range of optimal conditions for the growth of the fruit crop. Due to global warming, uplands will become more important for fruit tree planting.

The total soluble solids content of fruit increases with altitude in most cases, probably due to an increase in photosynthesis resulting from higher solar radiation. For total titratable acids there was no clear trend with altitude and this depends more on the fruit species and site conditions. The content of antioxidants in fruit flesh and epidermis increased in response to increasing altitude and increased UV light in several fruit species. Physiological disorders in the highlands include stress caused by high UV radiation and very low temperatures.

In general, there is no positive effect on fruit quality when altitude is at or above the recommended range for a fruit species. Varieties with higher photosynthetic performance and lower sensitivity to photo inhibition are better adapted to high altitudes.

The possibility of fruit damage due to extreme temperatures at high altitudes is almost negligible, and it is thought that the fact that fruit growing can be carried out in cold climates in these regions is due to the fact that the temperatures in June and September are sufficient and the sunbathing is quite high. In the ripening of fruits, regular sunbathing time is as important as temperature. Since high relative humidity is a problem in fruit growing, especially in terms of fungal diseases, it is seen that the relative humidity at high altitude is in a very suitable range for fruit growing, which means that the use of many agricultural control chemicals, especially fungicides, will not be necessary. The high bioactive content of fruits grown in high altitude conditions and in cool climates makes high altitude areas especially suitable for fruit growing. Especially against the risk of global warming, which is expected to increase further, it has come to the agenda that fruit growing should shift to cultivation areas with cool climates and high altitudes. In this direction, cultivation areas with high altitude can become a center of attraction by identifying and disseminating varieties and rootstocks suitable for ecological conditions and market demands, and determining the ideal cultural practices. Loss of our genetic potential should be prevented by protecting and multiplying existing varieties that have been adapted for thousands of years. In addition, an increase in product diversity should be ensured by planting high altitude varieties suitable for the region, which will be procured domestically and from abroad.

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CHAPTER 2

FORMATION OF MICROPLASTICS, THEIR HARMS AND BIOACCUMULATION IN SOME ANIMAL SPECIES

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INTRODUCTION

Plastic has become an indispensable part of life due to its high production quantity, lightness, easy shaping, low production cost and easy use in many areas. As a result of the increasing demand for plastic, plastic production is increasing every year.

However, a small part of the plastics produced are recycled and the remaining part is thrown into the environment. These abandoned plastic materials undergo a process of degradation into smaller, harmful fragments like microplastics and nanoplastics over time. This transformation is driven by environmental elements such as heat, humidity, temperature, and wind, leading to the emergence of environmental pollutants. Microplastics (MP) are very small particles of plastic and form a significant part of environmental pollution problems. The harms of microplastics to animals have been investigated and documented in various ecosystems and organisms. While plants are exposed to microplastics through water, soil and air, animals are also exposed through food sources in addition to these pathways. Numerous studies have indicated that microplastics induce adverse and toxic effects on humans, animals, and plants.

These damages pose a threat to humanity in terms of environmental, ecological and health aspects. These include biological damage, digestive problems, toxicity, reproductive problems and genetic deformations. Studies suggest that microplastic exposure in living things may cause health problems such as inflammation, oxidative stress and tissue damage (Galloway and Lewis 2016). In studies conducted on humans; There is a suggestion that the presence of microplastics (MPs) could potentially lead to corrosive effects, including inflammation, oxidative stress, and cytotoxicity. When examined more specifically, it has been reported that microplastics can damage DNA when in contact with environmental organisms (Mattsson et al., 2015). It has even been suggested that microplastics will negatively affect the adaptation abilities of organisms by reducing or changing their genetic diversity (Hartmann et al., 2019).

In this study, the definition, formation, damages and effects of MPs on some animal species were examined. It is also discussed what measures can be taken to protect against the harmful effects of microplastics.

1. Definition and Formation of Microplastics

Plastics are petroleum-derived semi-organic materials that can easily take the desired shape (Güler, 2013). Plastics, which are widely used in the packaging industry, can remain unchanged after becoming waste, or they can be broken down into small pieces with heat generation and advanced biodegradation (Lehner et al., 2019). Through research findings, it has been established that plastics undergo a transformation into increasingly detrimental microplastics (MP) and nanoplastics (NP) over time, influenced by external factors like ultraviolet irradiation, hydraulic erosion, and biological degradation (Wang et al., 2021). Microplastics (MPs) are characterized as artificially produced solid particles exhibiting either regular or irregular shapes and falling within the size range of 1 µm to 5 mm (Frias and Nash, 2019), are found in freshwater, ocean (Zantis et al., 2021), soil (Yang et al., 2021) and even in the air (Gasperi et al., 2018).

The most common plastic wastes are polypropylene, polyethylene, polyvinylchloride, polystyrene and polyethylene terephthalate (Andrady, 2011).

Figure 1. Degradation of plastics

Approximately 300 million tons of plastic are produced every year, and it has been reported that only a certain part of it can be recycled and approximately 10% of it ends up in the oceans (https://plastik-pollution.org/).

Plastics, which are very durable in the natural environment and take many years to decompose in nature, have very slow biodegradation times. It has been determined that plastics thrown into nature by us humans decompose and shrink under the influence of various external conditions such as biodegradation, hydraulic erosion, thermal degradation, physical abrasion, exposure to ultraviolet radiation and winds, and become more harmful microplastics and nanoplastics. Abiotic factors, including light, temperature, air, water, and mechanical forces, along with biotic pathways initiated by living organisms, contribute to the degradation of plastics. Reports suggest that abiotic degradation takes place prior to biodegradation, primarily attributed to the limited bioavailability of plastics (Andrady, 2017; Wang et al., 2021).

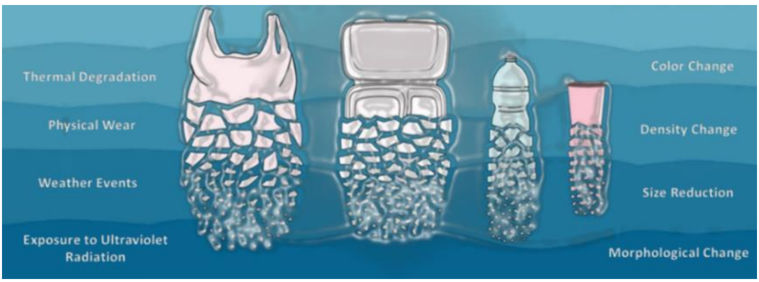

Figure 2. Decomposition of Plastics

Source: https://www.istockphoto.com/tr/ill%C3%BCstrasyon/microplastic-pollution

2. Ways of microplastics reaching living things and their harms

The natural environment harbors a significant volume of plastic waste, a consequence of factors such as extensive plastic manufacturing and inadequate recycling practices. These wastes are carried to rivers and seas by wind, rain and erosion, causing intense plastic pollution in coastal areas.

Studies on plastic pollution are generally carried out on aquatic creatures or aquatic ecosystems. While microplastics (MPs) have been extensively identified in aquatic environments, their occurrence in soil ecosystems has been largely overlooked (Jin et al., 2022).

Microplastics (MPs) are present not only in natural settings like soil, seawater, and freshwater but have also been identified in seafood and beverages. This raises concerns about the potential ingestion of MPs by animals and humans through the food chain (Sucharitakul et al., 2021).

Land ecosystems, especially agricultural ecosystems, where humans operate more intensively, are always more likely to be polluted with plastic waste (Piehl et al., 2018). Industrial and agricultural activities contribute significantly to the annual production of substantial quantities of heavy metals, pesticides, and plastic products. This invariably leads to environmental pollution and poses a threat to the health of animals (Zhong et al., 2021; Liu et al., 2023). Contrary to traditional agricultural practices, the extent of pollution is increasing in conventional agricultural practices, which are mostly carried out for commercial purposes. Plastic mulching, silage films, sewage, greenhouses, fertilizer and compost applications, sewage sludge, biological wastes, etc. during agricultural production stages cause more intense plastic pollution (Piehl et al., 2018; Jin et al., 2022).

Depending on their size and type, MPs have the ability to penetrate into plant cells such as seeds, roots, leaves and fruits (Dietz and Hertz, 2011). Activities such as natural disasters, physical properties in the soil (cracks, pores), external human intervention (agricultural practices such as plowing, harvesting, digging, etc.) and animal intervention (animals scratching, dragging the soil) cause MPs to somehow enter the soil with horizontal and vertical movements. and causes them to settle on plant roots (Gabet et al., 2003). The size of nanoplastics prevents the transmission of nutrients in plants by allowing MPs to enter plant tissues and cells and blocking not only root surfaces but also vascular tissues (Schwab et al., 2016).

While plants are exposed to microplastics through soil, water and air, animals are also exposed through food sources in addition to these pathways. In this way, through the food chain, humans and land animals necessarily interact with MPs in some way (Oliveri et al., 2020; Esposito et al., 2022).

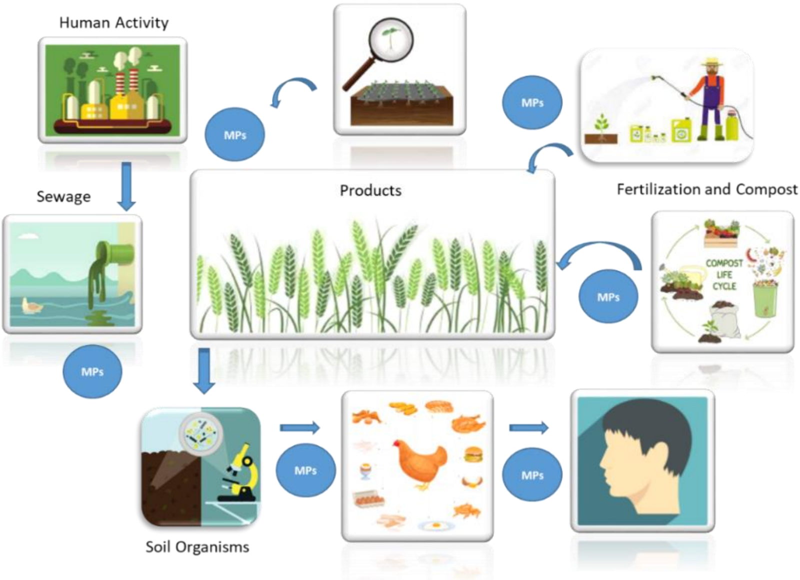

Figure 3. Pathways of exposure of living organisms to microplastics

Animal exposure levels to MPs may vary depending on environmental conditions, the region where they are raised, and various other factors. A few of these exposure routes and the most important ones are; These can be listed as feed, water and irrigation water, soil, entry into the food chain, and environmental exposure. The feed consumed by animals to meet their food requirements is contaminated with MPs during the processing and storage of feed raw materials. In addition, drinking water and irrigation water are also contaminated with MPs.

Animals can come into contact with MPs accumulated in the soil by digging or eating the soil and transfer them to their bodies. Similarly, microplastics (MPs) can infiltrate the food chain via water, soil, and plants within their surrounding environment.

Reports indicate an annual accumulation of an estimated 1.15-2.41 million tons of plastic waste within the agricultural ecosystem (Lebreton et al., 2017). The threats posed by MPs have recently caused great concern among the scientific community. In addition to penetrating and accumulating in the body of aquatic organisms, MPs can directly or indirectly affect the physical properties of the aquatic organism by changing the penetration of light, its chemical properties by consuming oxygen, and its biological ecosystems by binding to different biotas (Manzoor et al., 2022).

It has been reported that MPs rarely have lethal effects on animals and plants in aquatic and terrestrial ecosystems, but instead cause general oxidative stress, including gene expression, which will affect all living things (Yu et al., 2020).

MPs clinging to plant roots prevent the growth of roots and buds by preventing the plant from receiving the water it needs and negatively affecting the respiration required for photosynthesis. Apart from affecting photosynthetic processes, it holds the capacity to influence the antioxidant system, potentially causing changes in the functionality of antioxidant enzymes. (Gao et al., 2019; Yin et al., 2021). In addition, they also affect element compositions.

It has been reported that approximately 0.9 x 10^4 to 7.9 x 10^4 MPs can accumulate per year in an adult human body exposed to MPs through routes such as ingestion, inhalation and dermal contact (Wu et al., 2022). Research findings indicate that prolonged exposure to microplastics could be linked to chronic inflammation, cancer, and autoimmune diseases, possibly due to the immune system's suppression (Prata et al., 2020). It has been suggested that MPs and nanoplastics may have serious negative effects on endocrine disruption, reproduction and growth (Shen et al., 2019). Bisphenols and phthalates, found in synthetic MPs, have been linked to endocrine disorders and numerous health issues, including diabetes, cancer, and obesity (Blackburn and Green 2022; van Raamsdonk et al., 2020). Another study suggests that microplastic exposure may cause negative effects on the immune system of organisms, leading to changes at the genetic level (Lu et al., 2016).

Indications of MPs in animals encompass reduced feed intake, induction of oxidative stress, emergence of morphological disorders leading to physiological and digestive system issues, elevated osmotic pressure, release of toxic chemicals, and diminished energy distribution within animal populations (Chae et al., 2018).

There are various studies showing that microplastic exposure can change the gene expression of organisms and that these changes can lead to potential effects on health. A study on microplastic exposure showed changes in liver gene expression of fish and that these changes affected genes related to the immune system and metabolism (Rochman et al., 2017). Another study conducted on marine organisms shows that microplastic exposure can change the expression of some genes (genes related to stress response, immunity and energy metabolism) (Browne et al., 2008), and also shows that terrestrial organisms exposed to microplastics experience changes in immune system gene expression. These changes can affect the immune responses of organisms (Sussarellu et al., 2016).

Studies have reported that due to the inadequacy of analysis methods, only particles above the nano size can be examined, which may cause the potential health effects arising from smaller particles to be overlooked (Bouwmeester et al., 2015).

3. Studies investigating microplastics on bioaccumulation in some animal species

Over the past 10-15 years, many comprehensive studies have been conducted on the sources, fate and toxicity of MPs in the aquatic ecosystem. However, there are almost no studies on their accumulation, uptake, penetration in the terrestrial (soil) ecosystem, and their effects and risks on the photosynthetic components of the terrestrial ecosystem (Evangeliou et al., 2020; Ullah et al., 2021).

In a study conducted by Lusher, (2015) to investigate the effects of MPs on marine organisms, it was determined that MPs had negative effects on seabirds, fish, mussels and other marine organisms. In addition, a study by Wright et al. (2013) revealed that microplastics damage the digestive systems of marine organisms and these organisms cannot remove ingested microplastics.

MPs especially affect the intestines, which are responsible for digestion and absorption in the digestive system, and the microbiota community found there on a large scale (Cheng et al., 2021).Although the number of studies on MP accumulation in various living species is quite high, the number of studies on farm animals is quite limited.

As a result of the intense interaction of all habitats with MPs, these particles inevitably contaminate animal bodies and products. MPs decomposed over time can enter the digestive tract through nutrition and eventually be absorbed or excreted by the body (Liu et al., 2022a). As a matter of fact, in studies conducted in previous years, the presence of MP in chicken feces provides evidence that poultry ingested these substances (Wu et al., 2021).As a result of a study conducted on chickens collected from seven regions in the Philippines, the presence of MP was observed in five regions in their intestines and three regions in the gizzards (Leon et al., 2022).

In a 2023 study by Zou et al., which explored the adverse effects of MPs exposure on the growth performance and intestinal microbiota of chickens, the findings indicated a notable decline in the growth performance of chickens. Moreover, the gut microbiota exhibited a significant reduction in alpha diversity, ultimately resulting in an imbalance of intestinal microbes. Studies on farm animals, which are part of the terrestrial ecosystem, have yielded striking results.

Lwanga et al. (2017) in a study where they examined the microplastic contents of soil, worm manure and chicken manure, 0.87 ± 1.9 particles g −1 in soil, 14.8 ± 28.8 particles g −1 in worm manure and 129.8 ± 82 particles g −1 in chicken manure.

Zhang et al. (2023) evaluated the amount of MP accumulated in the soil by organic pig and cow manures during an 11-year field study; They determined it to be 3547 kg^{-1} in pig manure and 4520 kg^{-1} in cow manure. With the use of these manure composts, the MP concentration in the soil increased every year and ranged between 144-316 kg^{-1} on average. The amount of MP accumulated in the soil after many years of compost application was determined as 1.73×10^8 to 7.22×10^8 products ha^{-1}. According to the results obtained, MPs accumulated as a result of organic fertilizer compost in the soil: eroded, decomposed, and also decreased in size and number.

The smallest particulate form of MPs (<0.1 μm; i.e. nanoplastics), whose toxic effect increases as their size decreases (Banerjee and Shelver 2020), can cross cell membranes, the placenta and the blood-brain barrier and enter systems (Prietl et al., 2014).

In a 28-day study with 30 Arbor Acres chickens, MPs were added to the diet at a rate of 200 mg/kg and as a result, reports indicated noteworthy reductions in both the body weight and average daily weight gain of chickens within the groups exposed to MPs. In addition, a decrease in antioxidant capacity and intestinal microbiota diversity, and abnormalities in intestinal villus morphology have been observed (Li et al., 2023a).

Liu et al. (2022b) determined MP levels in chicken eggs obtained from supermarkets in their study on MP transfer to animal products.As a result, it was determined that there were 11.67 ± 3.98 particles of MP per egg and that this MP presence was due to polyethylene through infrared measurements. Additionally, it has been reported that the presence of MP in egg yolk is higher than in egg yolk.

Susanti et al. (2021) in their study to prove microplastic contamination on 25 ducks, they detected the presence of 11-49 MP-1 in the intestines.

Tanaka et al. (2013) reported in a study conducted on shearwater birds that plastics passed into the tissues of the birds.

Similarly, Bahrani et al. (2023) observed MP concentrations in tissues such as meat, liver and rumen of sheep and cows.

In their examination of microplastic (MP) contamination originating from packaging in ready-made food sales and the use of polyethylene-based plastic cutting boards for food preparation, Habib et al. (2022) reported microplastic contamination levels ranging from 0.014 ± 0.024 to 2.6 particles per gram of chicken meat and approximately ± 2.8 particles per gram in fish. Again, Habib et al. (2022) conducted a similar study, they found that the MP size in raw meat purchased from Middle Eastern butchers and supermarkets was 1279.2 ± 835.0 µm, and washing the meat with water for 3 minutes before cooking reduced the MP number to 0.07 MP/g.Additionally, they determined that 875 g of polyethylene was lost from a used and expired cutting board.

On mulching, which is one of the general problems in the production phase in order to obtain more productive crops in agriculture, Beriot et al. (2021) investigated in their study to what extent sheep swallowed and carried MPs. According to the results they obtained from the vegetable field where they first took 6 samples, they determined 2×10^3 particles·kg −1 MP in the soil. It has been reported that there are 10^3 particles kg $^{-1}$ MP in sheep feces and the possibility of its transmission (in 1000 flocks) can be 10^6 particles ha^{-1}.

Wu et al. (2021) examined the distribution of MPs from 3 different species in the feed and fertilizers of 19 cattle and poultry using FTIR microscopy. As a result, polypropylene and polyethylene types of MPs, including colored particles and fibers, were determined as 115 types in fertilizer and 18 types in feed. It has been suggested that the application of these fertilizers to agricultural soils will increase the number of MPs day by day.

In the investigation conducted by Li et al. (2023b), the study involved the utilization of 10 animals to assess the existence and characteristics of microplastic (MP) particles within the lung tissue of both domesticated and fetal pigs in their native habitat. According to the results determined by the polarized light microscope LDIR imaging system, 12 particles/g with sizes of 115.14 µm to 1370.43 µm were observed in domestic pigs, while half of these values (6 particles/g) were determined in fetal pigs. The most abundant MP type in domestic pig lungs was polyamide (46.11%), while polycarbonate was the most abundant polymer in fetal pigs (32.99%).

The animals most exposed to microplastics are fish. In a study conducted in the Mediterranean, it was found that 23.3% of all fish, whose main source is fiber, contained plastic parts (Giani et al., 2019).

Another study conducted in the northern and central Adriatic Sea reported that 95% of fish contained microplastics

(Pellini et al., 2018). Similarly, it has been reported that approximately 56% of fish in the Tyrrhenian and Ligurian Seas have MP in the gastrointestinal tract (Sbrana et al., 2020), and 46.8% of fish in 20 regions in France, Italy, Greece and Spain (Tsangaris et al., 2020) .

Suggestions propose that the ingestion of microplastics, facilitated by their small size, could adversely impact the feeding behavior, reproduction, and energy mechanisms of marine organisms. Furthermore, this ingestion may potentially elicit inflammatory responses, histological changes, DNA damage, cytotoxicity, physical harm, and increased mortality (Bour et al., 2018; Ribeiro et al., 2018). al., 2019).

Studies have found that histological changes occur in the liver and intestines of fish exposed to microplastics and cause oxidative stress (Espinosa et al., 2019). It has been determined that microparticles are especially prevalent in the gastrointestinal tract and muscular system (Zitouni et al., 2020).

Marco et al. (2022) conducted a study with zebrafish to examine the accumulation and toxicity of MPs, and observed a delay in the incubation period, changes in larval development, and deformations in body and tail development in zebrafish exposed to MPs. Moreover, heightened expression levels of genes associated with oxidative stress (sod1, sod2, and cat) and cellular detoxification (gst and cyp) were observed in zebrafish larvae following exposure to microplastics.

Lu et al. (2018) found that body, liver and lipid weight decreased and intestinal functions were impaired in mice exposed orally to 1000 µg/L 0.5 and 50 µm polystyrene MP. Furthermore, microplastics have been documented to induce various pathological processes in the intestine, such as microbial dysbiosis, metabolic disturbances, oxidative stress, as well as hepatic and renal toxicity.

Some studies on sea bird species have shown that; MPs cause decreased appetite, damage and obstruction of the intestines, decreased body fat accumulation, physiological disorders, and also have negative effects on the morphometric characteristics and blood calcium levels of birds (Lavers et al., 2014; Lavers et al., 2019; Weitzel et al., 2021).

CONCLUSION AND RECOMMENDATIONS

Microplastics produced from petroleum-based materials pose a major threat to food and agriculture.Agricultural soils are the main storage sites of microplastics, and the behavior of these microplastics can be affected by various environmental factors and absorb these microplastics in different ways.They become destructive for all living organisms due to their toxicity, resistance to physical and biochemical degradation, and properties such as absorption.

The adoption of environmentally sound waste disposal practices, the reduction of plastic usage, and efforts to minimize the risk of plastic contamination in the surrounding environment are essential measures to safeguard living organisms from the adverse effects of microplastics.

The first step to prevent MPs from reaching livestock should be to keep MPs away from agricultural land and feed. However, since it is almost not possible to do this today, we should instead focus on advanced techniques and applications developed in the light of science. Feed processing equipment should be cleaned regularly to prevent contamination during the production process. It should be ensured that the feed ingredients you use are of high quality and quality control measures should be taken to minimize the risk of contamination. In order to protect against the negative effects of MPs on ecosystems, it is important to contribute to awareness and search for solutions at the social level, as well as your personal efforts. If the presence of MPs is suspected or there are concerns about their impact on the livestock enterprise, experts in environmental science and agriculture who can provide guidance and potentially perform testing should be consulted. These simple and immediate measures will help reduce the likelihood of MPs entering the diet.

In addition to these simple measures, complex and future-oriented measures should be taken. In agricultural production, physically and biologically degradable MPs should be preferred instead of toxic MPs that cannot be easily degraded.

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CHAPTER 3

EVALUATION OF THE GROUNDWATER QUALITY IN KÖSE-KELKİT REGION (GÜMÜŞHANE-TÜRKİYE)

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INTRODUCTION

Groundwater, especially in arid and semi-arid regions, is vital for drinking and use. The use of unhygienic drinking water in developing countries poses a danger to human health (Rajendran et al., 2019). The water quality is rapidly deteriorating due to significant increases in industrialization and urbanization and the spread of agricultural activities (Rajendran et al., 2019). Deterioration of groundwater quality is mainly caused by multifaceted interactions of geology, hydrogeology, topography, drainage systems, hydrometeorology and anthropogenic activities (Kim et al., 2005). Water quality is evaluated with different techniques, and hydrogeochemical methods, water quality indices, and GIS are commonly used (Güler et al., 2002; Appelo & Postma, 2005; Babiker et al., 2007; Selvam et al., 2014; Barzegar et al., 2016; Rajendran et al., 2019; Gao et al., 2020).

In Kelkit and Köse districts (Gümüşhane-Turkey), located within the Upper Kelkit valley, local people mostly make their living from agriculture and animal husbandry. In this sense, it meets its water needs mainly from groundwater, including drinking water. This study aimed to determine the Kelkit and Köse districts' and surroundings' groundwater quality and evaluate their usability as drinking, domestic use, and agricultural use water.

1. STUDY AREA

The Kelkit-Köse region, located on the border of Bayburt and Gümüşhane provinces, is situated between 40°03′45″-40°13′00″ northern latitudes and 39°12′03″-39° 42′ 13″ eastern longitudes (Figure 1). The altitude of the study area, which has a total area of 445 km^2, varies between 1408-1580 m above sea level, and the land slope is generally low.

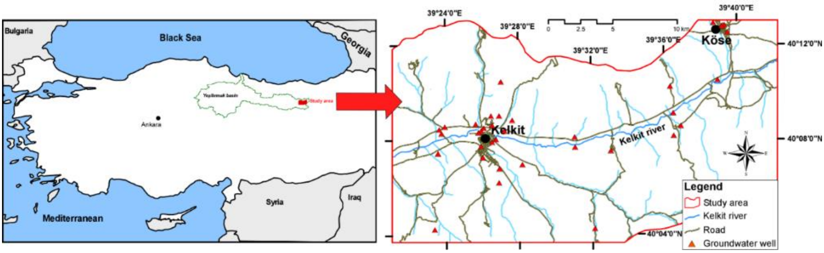

Figure 1. Location map and groundwater sampling points of the Kelkit-Köse region.

In the study area, where the transitional climate between the Eastern Black Sea and Eastern Anatolia Regions prevails, summers are hot and dry and winters are cold and rainy (Wikipedia, 2023). The average annual temperature of the study area is 9.6 °C and the average annual precipitation is 460.3 mm (MGM, 2023).

The study area is located in the orogenic belt called the Eastern Pontides. In this region, the basic unit is the Köse granites, which are a metamorphic rock type and crop out in the district within the borders of the study area (Topuz et al., 2010). Köse granites are unconformably overlain by the Şenköy Formation, which is characterized by andesite, basalt, lava and pyroclastics, sandstone, siltstone, claystone, and mudstone (Evcimen, et al., 2020). The Berdiga Formation, represented by clayey limestone and massive carbonates towards the top, lies harmoniously on the Şenköy Formation (Evcimen et al., 2020). Berdiga and Şenköy Formations are overlain by Kelkit Formation and Quaternary alluviums, respectively (Arslan & Aliyazicioglu, 2001).

Aquifer types in the study area are generally divided into two groups. These are the alluvial aquifer consisting of unconsolidated material extending along the Kelkit Stream and the semi-consolidated aquifer hosted by the Kelkit Formation (Yıldırım, 2023). 42.95% of the Kelkit district and 39.54% of the Köse district are agricultural areas (Gümüşhane Provincial Directorate of Environment and Urbanization, 2021) and water supply for irrigation purposes is provided by the Kelkit Stream and the wells drilled in the aquifers (Yıldırım, 2023).

2. METHOD

2.1. GROUNDWATER SAMPLING AND ANALYSIS

A total of 20 groundwater samples were collected throughout the study area in November 2022. Physical parameters (pH, EC (µS/cm) and temperature (℃)) of water samples were measured in situ in the field. Water samples were stored in 250 mL HDPE (high-density polyethylene) plastic bottles at +4 ℃ and transferred to the laboratory environment. Two 250 mL HDPE bottles were used for each sample, and 2 mL nitric acid was added to one bottle to prevent adsorbing cations. Analyzes of chemical parameters (major anion-cation and trace element) were performed in the laboratory using ICP-MS and spectrophotometer methods in the Bayburt University, Central Research Laboratory.

2.2. HYDROCHEMICAL CLASSIFICATION

Various diagrams have been developed to visualize chemical analyses, determine the facies of waters, and determine their suitability for use (Hem, 1985). Before the hydrogeochemical classification process, the cation-anion charge balance must be checked (Freeze & Cherry, 1979). The error in this load balance is recommended to be below 5%. In the charge balance calculation (Equation 1), ion concentrations are converted to meq/L (Freeze & Cherry, 1979).

$$Charge\ Balance\ (\%) = \frac{total\ cations - total\ anions}{total\ cations + total\ anions} \times 100 \qquad Equation\ 1$$

The Piper diagram (Piper, 1944) is widely used to interpret the hydrochemical evolution of groundwater. The facies of water is determined through the Piper diagram (Figure 2), in which the concentrations of major anions and cations (meq/L) are used.

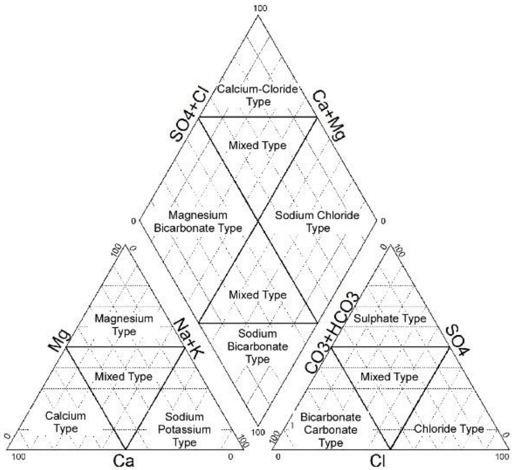

Figure 1. Piper diagram.

The US Salinity Laboratory diagram was used to evaluate the usability of water consumed, mainly for irrigation purposes. The purpose of this diagram (Figure 3a), whose axes are formed by Sodium Adsorption Rate (SAR) and electrical conductivity (EC) values, is to classify sodium and salinity hazards (United States Salinity Laboratory Staff, 1954). High sodium (Na^{+}) concentrations in irrigation water reduce soil and plant productivity (Todd, 1980). For this reason, it is essential to know the SAR rate, calculated with the formula in Equation 2.

$$SAR = \frac{Na^{+2}}{\sqrt{\frac{Ca^{+2} + Mg^{+2}}{2}}} \qquad Equation\ 2$$

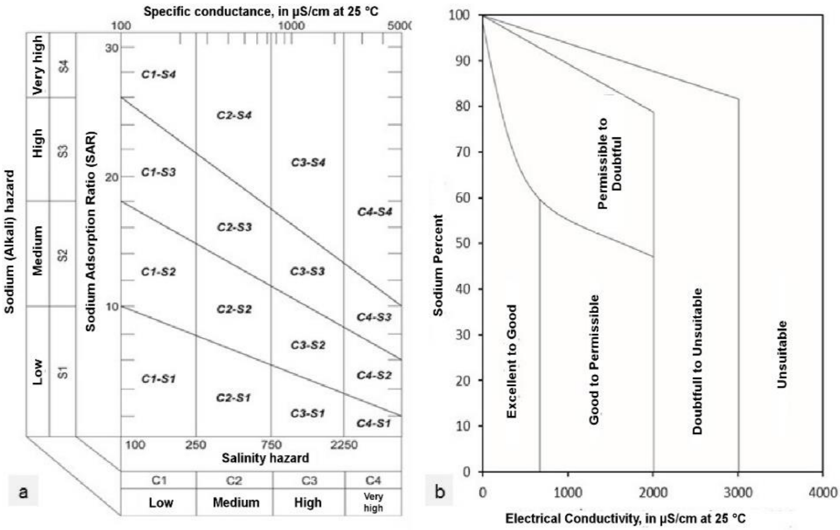

Figure 2. The US Salinity Laboratory diagram (a), Wilcox diagram (b).

The US Salinity Laboratory diagram divides water into four classes according to SAR (S1, S2, S3, and S4) and salinity amount (C1, C2, C3, and C4). These classes are detailed in terms of the suitability of waters for soil and plants.

Wilcox (Wilcox, 1948) used sodium percentage (%) and electrical conductivity (µS/cm) values to evaluate the suitability of groundwater for irrigation. Sodium percentage indicates sodium hazard calculated with Equation 3 (Wilcox, 1948). The diagram created with these two values (Figure 3b) divides water into five classes. These classes are "excellent to good, good to permissable, doubtful to unsuitable, permissable to doubtful, and unsuitable."

$$\%Na = \frac{Na^{+}}{Ca^{+2} + Mg^{+2} + Na^{+} + K^{+}} \times 100 \qquad \text{Equation 3}$$

2.3. WATER QUALITY INDEX

The Water Quality Index (WQI) is an effective tool that estimates water quality for drinking purposes by examining various chemical parameters (Adimalla, 2019). These parameters are chosen based on data availability, expert opinion, and environmental impact significance (Uddin et al., 2021). In this study, pH, TDS, TH, Na^{+}, Cl^{-}, SO_4^{-2}, NO_3^{-}, and F^{-} parameters were used. Determination of WQI consists of three stages (Horton, 1965; Brown et al., 1970);

i. A weight value (wi) between 1 and 5 is assigned to the parameters to be applied in WQI. In assigning these weight values, the importance of quality in drinking water is taken into account.
ii. At this stage, the relative weight (Wi) value of each parameter is calculated using the equation below (Equation 4).

$$W_i = W_i \div \left(\sum_{n=1}^{n} w_i \right) \qquad \text{Equation 4}$$

Where; Wi is the relative weight, wi is the weight value assigned to each parameter, and n is the number of parameters.

iii. The quality indix is obtained by dividing the concentration (mg/L) of each sample by the drinking water standard determined by the World Health Organization (WHO, 2017). The quality degree is calculated using the equation below (Equation 5).

$$Q_i = \left(\frac{C_i}{S_i}\right) \times 100 \qquad \textit{Equation}\ 5$$

Where; Qi is the quality degree, Ci is the concentration of each parameter measured in mg/L, and Si is the drinking water standard of each parameter determined by WHO.

To calculate the WQI value, the subindex (SI) of each parameter is calculated, and the sum of these subindexes gives the WQI result of the sample. WQI value is determined by following Equation 6 and Equation 7.

$$SI = W_i \times Q_i \qquad \textit{Equation}\ 6$$

$$WQI = \sum_{n=1}^{n} SI \qquad \textit{Equation}\ 7$$

Where; SI is the subindex of each parameter, and Qi is the quality rating. Waters are divided into five classes according to the calculated WQI value (Table 1).

Tablo 1. Water classes according to WQI.

WQI	Water Quality
<50	Perfect water
50-100	Good water
100-200	Poor water
200-300	Very poor water
>300	Not suitable for drinking purposes

2.4. HEAVY METAL EVALUATION INDEX (HEI)

The Heavy Metal Evaluation Index (HEI) (Edet & Offiong, 2002), which provides information about the general quality of water according to heavy metals, is a useful index for interpreting the pollution level (Prasanna et al., 2012). HEI is calculated with Equation 8, and heavy metals Ba, Cd, Cr, Cu, Ni, Sb, As, Pb, Fe, Mn, and Zn are used in the calculation.

$$HEI = \sum_{i=1}^{n} \frac{H_C}{H_{mac}} \qquad Equation\ 8$$

Where; Hc is the observed value (mg/L) of each parameter and Hmac is the maximum acceptable concentration of each parameter (Edet & Offiong, 2002). The values determined by the World Health Organization (WHO, 2017) were considered for maximum acceptable concentration values. Waters are divided into three categories according to HEI values: low pollution (<10), medium pollution (10-20), and high pollution (>20).

3. RESULTS

3.1. GROUNDWATER CHEMISTRY

Descriptive statistics of physical, chemical, and heavy metal parameters of the groundwater of the Kelkit-Köse region are presented in Table 2.

Table 2. Descriptive statistics of physico-chemical parameters of the Kelkit-Köse region.

Parameter	Unit	Groundwater (*n* = 20)					
		Mean	**Median**	**Minimum**	**Maximum**	**Range**	**SD**
pH		7.36	7.27	6.81	9.02	2.21	0.46
TDS	mg/L	672	629	389	1278	889	229
EC	µS/cm	830	788	499	1596	1097	272
TH	mg/L	362	371	87	738	651	132
Ca^{+2}	mg/L	97.7	102.3	32.8	177.1	144.4	32.0
Mg^{+2}	mg/L	28.7	28.6	0.1	71.8	71.7	15.9
Na^{+}	mg/L	52.1	31.9	3.7	325.9	322.1	72.0
K^{+}	mg/L	3.3	1.8	0.4	19.6	19.2	4.8
Cl^{-}	mg/L	26.6	19.0	1.9	125.9	124.0	28.1
HCO_3^{-}	mg/L	324.3	332.0	0.0	533.9	533.9	103.1
CO_3^{-}	mg/L	0.8	0.0	0.0	15.0	15.0	3.4
SO_4^{-2}	mg/L	94.8	43.0	2.4	754.4	752.0	165.2
NO_3^{-}	mg/L	29.1	22.8	0.0	200.5	200.5	42.2
NO_2^{-}	mg/L	0.5	0.0	0.0	2.4	2.4	0.6
F^{-}	mg/L	0.9	0.6	0.2	5.7	5.5	1.2
As	µg/L	9.8	1.3	0.2	159.7	159.5	35.4
Ba	µg/L	148.7	144.2	32.4	289.1	256.7	72.7
Cd	µg/L	0.0	0.0	0.0	0.1	0.1	0.0
Cr	µg/L	0.6	0.3	0.0	3.6	3.6	0.8
Cu	µg/L	0.5	0.5	0.0	1.6	1.6	0.5
Fe	µg/L	39.8	3.8	0.0	263.1	263.1	67.4
Mn	µg/L	12.8	0.8	0.0	203.9	203.9	45.4
Ni	µg/L	0.8	0.2	0.0	7.3	7.3	1.6
Sb	µg/L	0.1	0.0	0.0	0.1	0.1	0.0
Pb	µg/L	0.0	0.0	0.0	0.7	0.7	0.2
Zn	µg/L	5.9	0.0	0.0	73.8	73.8	16.8

3.2. HYDROCHEMICAL CLASSIFICATION

A Piper diagram was used to determine the facies of the study area groundwater samples (Figure 4). Considering the cation triangle of the Piper diagram, the majority of the groundwater samples are in the "calcium type" water class. While two samples belong to the "mixed type" water class, two samples belong to the "sodium-potassium type" water class. According to the anion triangle, all samples except one are in the "bicarbonate-carbonate type" water class. Eighteen groundwater samples classified in the rhombus, which reflects the combination of cation and anion triangles, are "calcium-magnesium-bicarbonate" type waters. One sample has high salinity and is in the "sodium-chloride type" water class. Another sample belongs to the “sodium-bicarbonate type” water class.

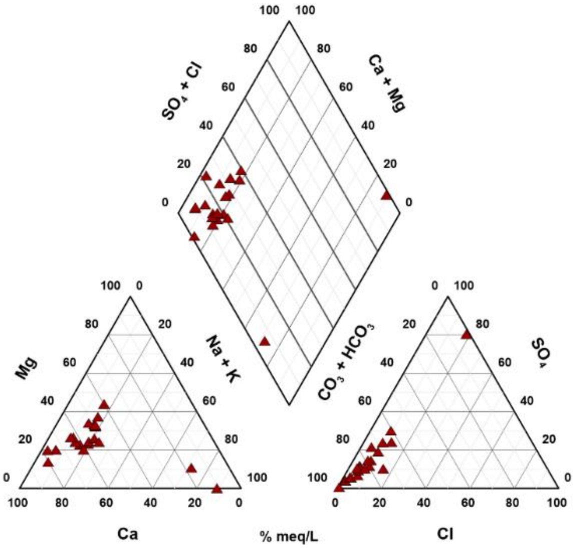

Figure 3. Piper diagram of the groundwater of the Kelkit-Köse region.

The US Salinity Laboratory diagram, which evaluates the usability of water as irrigation water, was used to assess the groundwater of the study area (Figure 5a). SAR values of groundwater in the study area vary between 0.09-15.21, while EC values vary between 449-1596 µS/cm. The majority of the samples are in the C2-S1 and C3-S1 classes. These waters are low in sodium and have medium and high salinity. While water in the C2-S1 class is used for moderately salt-tolerant plants, salt control, and special methods must be applied to use water in the C3-S1 class. The salt tolerance of the plants in which the sample in the C3-S2 class will be used, and the soil in which this groundwater will be used must be coarse textured or rich in organic matter.

According to the Wilcox diagram, which is another diagram in which the usability evaluation of irrigation water, five of the samples are in the "excellent to good" class, thirteen in the "good to permissible" class, and two in the "permissible to doubtful" class (Figure 5b).

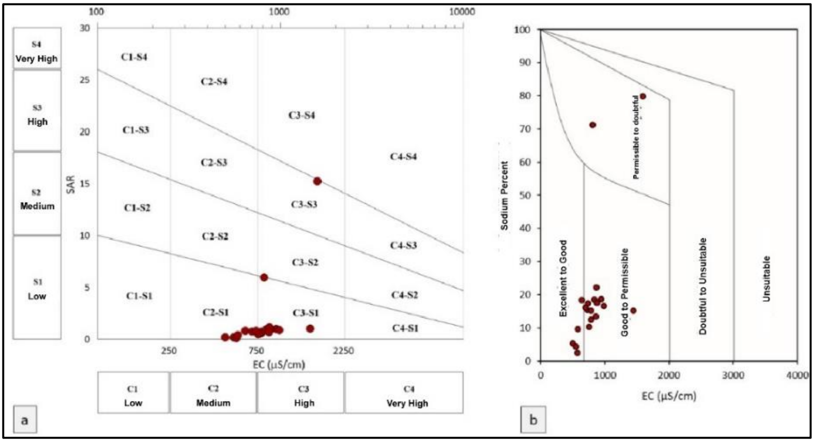

Figure 4. US Salinity Laboratory diagram (a) and Wilcox diagram (b) of the groundwater of the Kelkit-Köse region.

3.3. WATER QUALITY INDEX

Water Quality Index (WQI) and Heavy Metal Evaluation Index (HEI) were used to determine the quality of water used for drinking purposes by examining various chemical parameters and evaluating water pollution according to the heavy metals it contains. According to the WQI values, 13 out of 20 groundwater samples are in the "excellent water" category, six in the "good water" category, and one in the "not suitable for drinking purposes" category. The pH, TDS, Na^+, SO_4^{-2}, and F^- parameters of water unsuitable for drinking exceeded the drinking water limit values determined by the World Health Organization. According to HEI values, 19 groundwater samples are in the "low pollution" class, and "medium pollution" was observed in one sample. The As value of the sample in the medium pollution class is 16 times the WHO standard value.

4. CONCLUSION

In this study, groundwater sampling was conducted in the Kelkit-Köse region, and groundwater samples were obtained from 20 wells. The hydrogeochemical classification of the obtained groundwater was applied, their facies were determined, their usability as irrigation water was evaluated, and their quality was interpreted with the help of various indices (WQI and HEI).

According to the created Piper diagram, the majority of waters are "calcium-magnesium-bicarbonate" type waters. The waters evaluated with the US Salinity Laboratory diagram were mainly distributed in the C2-S1 and C3-S1 classes. Waters are divided into three classes in the Wilcox diagram created using sodium percentage (%Na) and EC (µS/cm) values. These are "very good-good", "good-usable" and "usable-doubtful".

According to the WQI and HEI data calculated by taking the limit values of water used for drinking purposes determined by WHO as reference, two samples are not considered appropriate to be used for drinking purposes. While high salinity was detected in one of these two samples, the As value of the other was high. It is predicted that if these waters are used for long-term drinking purposes, they will have negative effects on human health.

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CHAPTER 4

MICROBIOME and INFLUENCING FACTORS in NEONATAL CALVES

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INTRODUCTION

Calves reared using scientific breeding techniques form the basis of efficient cattle production and are important for the future and sustainability of cattle farms. Proper care, feeding, and health practices during the postnatal period can significantly impact growth, development, health, and performance in adulthood. On the other hand postnatal calf losses are a persistent problem with high morbidity and mortality rates worldwide and cause financial losses to cattle farms.

The gut microbiome in calves is a crucial aspect in achieving optimal health and performance. This is mainly because the gut microbiota is significantly affected by feeding and management regimes, resulting in alterations to feed conversion and immunological reactions. A beneficial microbiome may be established by implementing feed or management practices, particularly early stage.

The microbiome research area has originated from microbial ecology research, serving as a foundation for various industries including, human medicine, mathematics, food science, bioeconomics, biotechnology and agriculture (Berg et al., 2020).

Although the terms microbiota and microbiome are frequently used interchangeably, they do have unique differences. The microbiota is the community of microorganisms residing in an organism, functioning as a fundamental, delicate superorganism composed of billions of fungi, bacteria, and protozoa. It includes all the unique species coexisting with living organisms. The microbiota contains 10 times more cells and 10^{12} times more genes compeared to its host body. The microbiome encompasses all microorganisms populations and their genetic material and is distinct from the microbiota (Figure 1). This system offers multiple beneficial effects for the host, serving as a protective shield from transient bacteria, aiding in the fragmentation of food and recuperation of energy from the diet, offering nutrient metabolites for enterocytes, and exerting crucial regulation on the host's immune system (Küllük and Dalgın 2021). The microbiome encompasses both the microorganisms involved and encompass their theatre of activity, that lead to the creation of specific ecological niches.

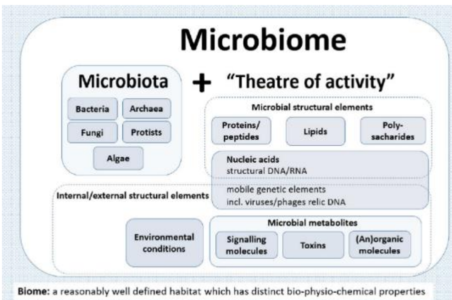

Figure 1. A diagram illustrating the components of the microbiome (Berg et al., 2020)

The microbiome encompasses a range of molecules generated by microorganisms, comprising structural components, metabolites, and host-produced molecules that are influenced by their environmental context (Berg et al., 2020). Thus, all genetic components (viruses, phages, extracellular DNA etc.) are contained in the microbiome.

The rumen microbiome is intricately associated with feeding and rearing systems. Because only the microbiota in the rumen can ferment compounds that are indigestible by other mammals. Therefore, it is essential to establish and improve the microbiota in the gastrointestinal tract (GIT) during the neonatal period to ensure healthy rearing. Microbiome variation in ruminants changes significantly during the initial weeks of life, which has a crucial impact on immune development. This change occurs rapidly and a key factor in ruminant health.

Factors Effecting The Neonatal Calves Gut Microbiota

Microbial colony formation and gut development are crucial for the proper functioning and growth of the GIT in young ruminants, with weight gain and feed efficiency being directly affected by these processes. This text aims to discuss some of these key factors in the context of ruminants. Some of the key factors that impact the microbiota of neonatal calves are discussed below.

Embryonic period

Maternal nutrition during the embryonic period is essential for the future health of newborns as it regulates developmental processes in utero. The gut microbiota may role-play as a mediator in this process. The bacteria transmission from mothers to their infants is necessary for the healthy microbiome development, which can influence child growth, immune system matures and even neurodevelopment. Likewaise, early establishment of the postnatal gut microbiota is critical for optimal metabolic activity, lasting health and the immune system. However, the observation of microorganisms in the newborn meconium suggests that the calves hindgut microbial colonisation may have begun pre-natal and that indigenous bacteria are transferred from the mother to the fetus via the placental barrier in utero. (Funkhouser and Bordenstein, 2013). With this, the fetal gastrointestinal tract and surroundings during the third trimester of pregnancy are sterile, and GIT colonisation has been reported to continue throughout the birth process after a rupture of the membranes (Malmuthuge and Griebel, 2018). On the other hand, Elolimy et al. (2019) supported the idea of bacterial influx from the weir to the fetus by the finding of bacteria in the newborn calves hindgut immediately after birth. Thus, It is not yet clear whether colonisation for mating takes place before birth (Seferovic et al., 2019). The risk of immune and metabolic problems in the newborn may be increased by antimicrobial treatment during pregnancy. The maternal microbiota crucially influences gut immune system development in the growing fetus, and this is linked to the distribution of microbial metabolites. Supplementation of methionine in the diets of dairy cows in late gestation altered the fecal microbiota and metabolism and positively affected the health and functionality of the hindgut of calves, despite no significant differences in feed intake. Changes in hindgut microbiota may also contribute to the provision of essential nutrients and prevention of pathogen colonization (Elolimy et al., 2020). Additional advantages of providing methionine from the dam to the newly born calf have been documented, such as faster development of metabolic pathways in the liver and improved innate immune function (Wang et al., 2019).

Maternal

The neonatal microbiome is initially colonised primarily through mother-infant microbial exchange, likely occurring through contact with the maternal's vagina, udder and milk in the neonatal stage (Klein-Jöbstl *et al.*, 2019; Stephens *et al.*, 2016). Vaginal delivery appears to have a significant impact on the microbial species development in the GIT due to the sequence of exposure, however fleeting, to microbes present in the vaginal flora of the mother (Taschuk and Griebel, 2012). Regardless, overall, it seems that exposing an infant to the microbial environment of the mother is helpful in regard to lifelong immune homeostasis (Torow and Hornef, 2017). According to a study by Guzman et al., (2015) young calves may have methanogenic archaea in their gastrointestinal tract on the day of birth. These microorganisms are anaerobic and cannot survive for an extended time outside the body. Additionally, the faecal microbiome of calves shows similarity to that of the vaginal microbiome in mothers, suggesting potential transmission via direct maternal interactions. Afterwards, the newborn could potentially come into contact with the microbiota found in colostrum and the skin of the mother. The hindgut's bacterial community is greatly impacted by the vaginal microbiota. Yet, the transfer of microbiota through the udder skin differed from vaginal or colostrum transfer, as numerous bacteria are present both on the maternal skin and in the surroundings. This means that they can be passed on from both the teat skin and the surrounding habitat (Yeoman et al., 2018).

Feeding with colostrum and milk

Diet has a noteworthy impact on the gut microbiota development and is considered to be a fundamental element in its regulation. Thus, implementing an appropriate feeding program in the initial stages of life of a calf will ensure the early development of its gut microbiota. This early development possesses a significant impact in the later stages of the calf's life.

Feeding colostrum to ruminants is highly recommended to ensure sufficient early nutrition and mucosal immunity development. Feeding colostrum to ruminants is highly recommended to provide sufficient early feed conversion and mucosal immunity development (Hang, 2019). Feeding newborn calves with colostrum has been found to increase *Lactococcus* levels and potentially reduce the spread of *Mycobacterium avium subsp. paratuberculosis* in calves that are infected with these bacteria (Stabel et al., 2019).

Milk contains 40 oligosaccharides and lactoferrin, which act as antibacterial, anti-inflammatory, and iron chelating agents. These components may inhibit the attachment of pathogens to the intestinal epithelium. It was observed that increased milk intake after birth led to improved health status in calves. An increase in *Lactobacillus* density was detected in the faeces of calves fed a higher amount of milk. This was related to an improved gut flora and a reduction in inflammation (Alimirzaei et al., 2020). An increase in the quantity of milk was found to enhance the height, width and surface area of villi in the small intestine of pre-ruminants. This could be imperative for rumen developmen

Water intake

Latest research has indicated that ingestion of water is conducive to improving the rumen development.

Wickramasinghe et al. (2019) conducted a study which showed that calves who consumed water from birth had increased improved feed efficiency, better fiber digestibility and body weight in comparison with those who water supply commenced at a later stage. A high concentration of *Faecalibacterium prausnitzii* and *Bifidobacterium* was observed in the intestinal flora of calves with early water consumption. Faecalibacterium bacteria are associated with increased digestibility of acid detergent fibre. In conclusion, early water consumption of calves has a positive impact on rumen development and digestibility (Arshad *et al*., 2021).

Calf Starter Feed

The initial few weeks of a calf's life have a very important function in their digestive system development and immune systems. Generally, calves begin consuming solid food in the form of starter feed between 2-3 weeks old. The appetizing initial feed of quickly fermentable carbohydrates stimulates the development and setting up of gut microbiota, particularly bacteria that degrade starch (Amin et al., 2021). Since milk goes directly to the abomasum, bypassing the rumen, it is crucial to feed solid foods to facilitate the development of rumen microbiota. Feeding pre-weaning calves with starter feed encourages rumen development and increases amounts of fermentable carbohydrates and resulting in higher levels of volatile fatty acids and acetate in the caecum. In summary, an increase in rumen weight and papilla size was observed with starter feeding in pre-weaning calves, which was ultimately associated with better healthy life and growth performance (Sun *et al*., 2018; Amin *et al*., 2021; Arshad *et al*., 2021).

Weaning age

Stress factors at weaning are known to have various negative impact on growth performance and feed intake. Weaning involves decreasing milk intake and increasing solid feed consumption for calves, usually between 6 and 10 weeks of age. In one research, Calves weaned at 6 weeks old exhibited a swift alteration in beta diversity of their rumen and faecal microbiome, along with inferior growth rates. Conversely, the calves weaned at 8 weeks old demonstrated a gradual modification in beta diversity, greater rumen development and feed intake (Mao et al., 2017)

Early weaning can be associated with higher mortality in calves, while delayed weaning may result in higher feed costs and reduced growth rates due to digestive tract issues, impeding performance later in life. An optimal weaning age should be chosen to ensure proper gut and rumen development before weaning and mitigate any adverse effects (Amin et al., 2021).

Dietary changes

The rumen development in neonatal calves is a crucial aspect of calf nutrition. Focusing on rumen development is therefore of paramount importance for effective calf nutrition. It plays a pivotal role in ensuring a healthy transition from milk or milk substitute feeds to more efficient and economical feeding practices. To guarantee healthy development and swift weight gain, it is crucial to develop the rumen of the calf as early as possible. Prolonged milk feeding disturbs the development of the rumen papillae.

Neonatal calves have an underdeveloped reticulo-rumen and are fed primarily on milk-based diets; as the calves mature, their diet gradually shifts from liquid to solid feeds. During this stage, there is a gradual increase in the quantities of fibre-degrading bacteria such as *Prevotella* and *Ruminococcus*, resulting in their dominance in the rumen. Therefore, the transition to the rumination phase due to the addition of solid diets is the critical point of GIT microbial growth. İn addition, If young calves do not receive feed during the transition phase, they may have difficulties adapting to the diet and physiology when they are first exposed to feed.

Conclusions

To improve the growth performance and healthy life of neonatal calves, the gut microbiota development is very critical. Feeding and rearing practices on farms have a significant impact on the gut microbiota. The feding and rearing strategies applied yield positive results in feed efficiency and immune system efficiency. These feeding strategies are necessary to sustain the performance of neonatal calves and to influence rumen development and beneficial microbes colonisation in a desirable manner. This is because the pre-weaning period has long-time and lasting effects on the formation of rumen microbiota. Yet, additional researches are needed to clearly find out the system of activity of influencing factors the gut microbiota development of neonatal calves.

Furthermore, it is crucial to identify pathways and key factors related to the interaction between host and microbes by considering the influence of microorganisms on the host's physiology in the first days of its life. Additionally, understanding the impact of host genetics on microbial populations is essential to achieve this aim.

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CHAPTER 5

MEDICINAL AROMATIC PLANTS GROWING IN THRACE REGION

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INTRODUCTION

It is known that there are approximately 422,000 plant species in the world and 52,885 of them consist of medicinal and aromatic plants. China is home to the greatest number of species of aromatic and medicinal plants, followed by those from Malaysia, Vietnam, the USA, India, and Vietnam. It is estimated that Turkey is home to about 500 different species of fragrant and medicinal plants. (Gül and Çelik, 2016; Temel et al., 2018).

Herbal product-based treatment techniques have been around for a while. Plant parts, including leaves, flowers, stems, roots, and all of them, are utilized either fresh or dried. Plants can be applied directly or boiled, and they can even occasionally be drunk as tea. Among the most popular ways involve boiling or brewing plants to release some of its active ingredients into water, which can then be consumed. Herbal and fragrant plants with strong scents are also utilized as spices. Grannite, thyme, red pepper, rosemary, and black pepper are a few of the most used spice plants in Turkey. Many of the fragrant and medicinal plants found in Turkey are harvested straight from the wild. The most often harvested aromatic medicinal plants in nature include thyme, bay, sage, rosemary, linden, and rosehip, according to research. (Faydaoğlu ve Sürücüoğlu, 2011; Yüzbaşıoğlu ve Kızıloğlu, 2019).

One of Turkey's seven geographical regions, Thrace is made up of the entire Istranca and Ergene sections that make up the Marmara Region, nearly half of the Çatalca-Kocaeli section, and the portion of the Southern Marmara section that corresponds to the Gallipoli Peninsula (Darkot, 1955).

The Thrace Region's diverse biological features, including its rivers, mountains, coastline, and three seas, contribute to its vast plant diversity. This diversity is demonstrated by the presence of about 2450 taxa from 145 groups in Thrace's natural vegetation. The abundant natural vegetation in the area contains fragrant and medicinal plants, which are harvested and used as a source of revenue (Özhatay et al., 1996).

The aim of this study is to identify some medicinal and aromatic plants growing in the Thrace region and to reveal the intended use and economic value of these plants.

Some Medicinal Aromatic Plants Growing in Thrace Region

Thyme

Thyme is an important medicinal aromatic plant that is traded and widely used in Turkey and has many genera, all belonging to the Lamiaceae family. Some of the thyme species are; It is known as *Origanum*, *Thymbra*, *Coridothymus, Satureja* and *Thymus* (Viuda-Martos et al., 2010).

It is among the species with high economic value and most exported. *Origanum onites* (ball thyme, Izmir thyme), *Origanum vulgare* subsp. *hirtum* (Istanbul thyme, black thyme), *Origanum minutiflorum* (Milkmen's thyme, highland thyme, Toka thyme), *Origanum majorana* (White thyme, Alanya thyme). 60% of the 52 *Origanum* species known in the world are distributed in Turkey. This is strong evidence that Turkey is the gene center of *Origanum* species (Başer, 2002).

Among the thymes growing in the Thrace region *Satureja cuneifolia* Ten., *Thymus longicaulis* C. Presl. subsp. *longicaulis*, *Origanum vulgare* subsp. *hirtum* (Figure 1.) and *Origanum vulgare* subsp. *vulgare*, *Thymus longicaulis* C. Presl subsp. *longicaulis* var. *subisophyllus* (Borbas) Jalas (Kültür, 2007; Kartal ve Güneş, 2017)

Figure 1. *Origanum vulgare* subsp. *Hirtum* (Başer, 2022).

Salvia fruticosa Mill and *Salvia officinalis*

Anatolian sage (*Salvia fruticosa* Mill.) (Figure 2.) is known as a medicinal and aromatic plant with commercial importance in our country. The increasing use of Anatolian sage in the field of health due to the active ingredients it contains and the consumption of the leaves of the plant as tea are increasing the popularity of the Anatolian sage plant day by day. This plant, belonging to the Lamiaceae family, is of great importance in pharmacology and perfumery industry due to its pleasant, pungent odor.

Figure 2. *Salvia fruticosa* Mill. (Anwar et al., 2017)

Anatolian sage (*Salvia fruticosa*) is a shrub that grows in Thrace, Western and Southwestern Anatolia, and has soft, densely hairy and grayish leaves. The essential oil (apple oil) obtained from the leaves carries up to 60% 1,8-cineole and is more valuable than medicinal sage (*Salvia officinalis* L.) (Figure 3.) in this respect (Baytop 1996; Bayram 2001).

Figure 3. *Salvia officinalis* L. (Ghorbani and Esmaeilizadeh, 2017).

Since Anatolian sage is distributed in the Marmara, Aegean and Western Mediterranean regions, its adaptation is high in regions with a temperate climate, especially in the coastal zone. Although it grows in different types of soil to which many plants cannot adapt, it prefers loamy-clay alluvial and medium textured soils.

Hypericum perforatum

Hypericum perforatum L. (Figure 4.) is a plant that is sometimes referred to as St. John's wort, sorrel, swordwort, binbirdelikotu, yeastwort, kanotu, and sheepwort, despite being a member of the *Hypericeae* (Syn. *Guttiferae*) family. The genus *Hypericum* contains between 350 and 400 species worldwide, including 70 species found in Turkey. Tropical climate plants such as *H. perforatum* L. are noted for their therapeutic aromatic properties. They can be found growing on roadsides, riversides, in uncultivated fields, in areas with rainy winters and dry summers, and in acidic-neutral soils with pH values ranging from 0 to 7 (Baytop, 1999).

The herb *H. perforatum* L., which was widely used in European folk medicine throughout the Middle Ages, was also commonly used in Anatolian folk medicine. It was traditionally used to cure jaundice, TB, and wounds, burns, and skin ailments. Additionally, it is recognized for usage in the management of conditions including liver cancer and gallstones (Moffat, 2014). This interest in *H. perforatum* L. persists to this day, with reports from clinical trials and scientific investigations carried out in the last few years indicating that it has significant benefits in the treatment of rheumatism, diabetes, cancer, hepatitis, bronchitis, dysentery, and throat infections (Gülben et al., 2008; Burunkaya, 2020). *H. perforatum*, which has a wide range of uses, has a very high economic value. The *H. perforatum* plant has many different uses. The most well-known of these are; St. John's wort tea, St. John's wort oil, St. John's wort decoction, St. John's wort tincture and St. John's wort extract (Ceylan et al.,2005; Çırak and Kurt, 2014).

Figure 4. *Hypericum perforatum* L. plant a. flower parts, b. root parts, c. stem parts and d. sample image collected from locations (Gıdık et al., 2022)

H. perforatum plant, which has been used for many different treatment methods in folk medicine for centuries, has proven its usefulness through studies and has become a plant used in pharmacological medicine. Research on St. John's wort and its compounds is increasing day by day due to its antioxidant, anti-inflammatory, anti-ulcerogenic and antidepressant effects (Kaya and Can, 2018).

***Urtica dioica* L.**

The most widespread species of the Urticaceae family, *Urtica dioica*, (Figure 5.) is also referred to as stinging nettle and is one of the most researched medicinal plants in the world. It is a perennial herbaceous plant that has long been used to treat a variety of illnesses. The plant thrives in both temperate and tropical wasteland regions globally, adapting well to a wide range of conditions. The Latin term urere, which means "to burn," is where the name *Urtica originates*, and it refers to its stinging hairs. *Dioica* is the most common species, so named because the plant typically produces either male or female flowers. There are two types of this plant: dioecious blooms, small oval, greenish-yellow achene fruit, and oval, long, petiolate, elongated leaves with toothed sides. (Upton, 2013).

Figure 5. Urtica dioica (Ahmed and Parasuraman, 2014)

The plant features a tuft of hair at the apex and stinging hairs. Many touch-sensitive non-stinging hairs on the leaves and stems, along with needles that can inject chemicals into the skin, such as serotonin, histamine, acetylcholine, moroidin, leukotrienes, and probably formic acid, are present. The irritating substances cause stinging, wheezing, or pain. Nevertheless, when cooked, this edible plant loses its irritating qualities, the heat does actually eliminate the juice's burning quality, and the young shoots can be employed in cooking (Kavalali, 2003).

With its extensive medical history, U. dioica is most frequently used to treat inflammatory conditions. This plant has been used to cure arthritis in traditional medicine. Pre-clinical tests have shown that it has antiasthmatic, antidandruff, astringent, depurative, diuretic, galactogogue, hemostatic, and hypoglycemic properties (Ahmed and Parasuraman, 2014).

Lavandula angustifolia L.

Lavender, or *Lavandula angustifolia* (Figure 6), is endemic to the hilly regions of the Mediterranean, ranging from Spain to France and Italy. It grows best at elevations above 1500 meters. There are two subspecies: ssp. pyrenaica, which is found in the Pyrenees, and ssp. angustifolia, which is indigenous to the French and Italian Alps. The essential oil composition of these subspecies has been found to differ somewhat, with ssp. angustifolia being the more economically significant of the two and wild ssp. pyrenaica being deemed unsuitable for regular commercial grade lavender oil (Passalacqua et al., 2017). This species is hardy in cultivation due to its origins in the mountains. It blossoms from mid-June to mid-July and develops as a shrub that can reach a width of 50 cm (Despinasse et. al., 2020).

Figure 6. *Lavandula angustifolia* (Betlej et al., 2023)

The essential oil obtained by water distillation from fresh lavender flowers and the dried buds separated from the dried stems after harvest are evaluated. Its essential oil, which is important for the perfume, cosmetics, taste and fragrance industries, is used as an antidepressant, antiseptic, antibacterial, diuretic, diaphoretic, stimulant, rheumatism pain reliever, antiseptic and expectorant (Arabacı ve Bayram, 2005).

CONCLUSION

Medicinal aromatic plants include important plants whose usage areas and economic value have increased in recent years. Thrace Region has ecological characteristics where plants with high diversity and economic value, which are included in Turkey's vegetable production, can be grown. Particularly thyme, sage, nettle, lavender and St. John's wort are among the prominent medicinal aromatic plants in the region. There are not enough studies on medicinal aromatic plants that have high economic value, a wide variety of uses and wide cultivation areas. More comprehensive studies should be conducted in this area.

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