A VIEW OF AGRICULTURE FROM AN ACADEMIC PERSPECTIVE

EDITOR Assoc. Prof. Dr. Gülşah BENGİSU



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PREFACE

Agriculture played a major role in humanity's transition to collective life. The hunter-gatherer communities that existed during the Stone Age gave way to people engaged in agriculture, giving rise to societies and states. Until the Industrial Revolution, agriculture was the main source of income for the majority of humanity. However, today there are visible developments in agriculture and the effects of technology. Significant changes took place in agriculture, especially during the 20th century. According to the Haber-Bosch process, the first artificial fertilizers were obtained using dung mixed with ammonium nitrate. Thanks to mechanization, which reduced the labor force in agriculture, a decrease in the number of workers in agriculture was observed. In response to the increase in production, unemployment increased. This book was created with the contributions of many researchers who are experts in their fields and aims to present the latest scientific developments on field crops to readers. We hope that this compilation will contribute to the agricultural sector and be a valuable resource for anyone interested in studies on field crops.

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CHAPTER 1

FINANCIAL DEVELOPMENT AND SUSTAINABLE AGRICULTURE: PERSPECTIVE ON AFGHANISTAN

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INTRODUCTION

Factors such as health – nutritive foods, improving security, reducing indigence and sustainability of environment have become staminal due to the disease epidemic. Increasing the capacity of products and food production, with sufficient focus on sustainability and safety, has turned into the most important priority, especially along with the epidemic period (Doruk Kahraman and Kahraman, 2023; Sridhar et al., 2023).

The dynamic economy of countries is one of the essential issues for managers to be self-sufficient and have power in all matters, including social, political, and cultural building blocks. Agriculture is one of the basic economic dynamics of the Afghan people, on whom the living conditions of a large part of the society are closely tied and which is known as the turning point of the society in this geography (Anonymous, 2022a; Davies et al., 2024; Khan et al., 2024). For many years, Afghanistan has become a country that immigrates to many countries of the world due to wars, internal conflicts, instabilities and security problems. Many Afghan citizens try to survive in different countries as refugees, asylum seekers or irregular immigrants (Kara and Kaya Tilbe, 2023; Mujtaba et al., 2024). Today's Afghanistan, formerly known as Khorasan, had developed agricultural and irrigation systems in the middle Ages. The Arabs were so surprised to see these systems that they named one of Basra's rivers Marghab in the 8th century. Dams were built in Sistan to divert the water in the Helmand River. Likewise, the waters of the Amu, Harirud and Kabul Rivers were used by building canals. Sistan was considered as a cereal storehouse. Khorasan people knew very well the land they used for agriculture. They would dig the soil to a depth of 2-3 meters, pound the clay in a mortar and pour water, and then filter the water and taste it. If the water tasted good, the soil was suitable for agriculture; if it was salty, they found it unsuitable for grain cultivation. They were also aware that soil fertility could be increased by using agricultural rotation systems. They divided the soil into different types such as

sandy soil, gravelly soil, yellow soil, black soil, red soil (Dupree, 1977; Meraj et al., 2024).

Almost 90% of Afghanistan's people derive most of their production from the agricultural sector. A few small factories operating especially in the cities of Herat, Kabul, and Mazar-e-Sharif do not have an increasing impact on the production process that can meet the needs of the people. Since there is no serious support for domestic industrial production in Afghanistan, factories face problems of low profits, low quality goods, lack of a reliable consumer market, lack of production standards, lack of insurance against accidents, and lack of electricity. In this state, domestic production will go bankrupt in a short time and, unable to compete with the arrival of foreign goods, it will inevitably surrender to the glory of imported goods (Anonymous, 2022b). In the past, Afghanistan was one of the major exporters of livestock products and dried fruits, but three decades of war destroyed the infrastructure in this country, limited the development of agriculture and caused serious damage to economic growth and the agricultural process. However, despite all these problems that create structural limitations for Afghanistan, agriculture can be a driving engine for economic growth and salvation in this country due to the presence of many talents and potentials in this country (Anonymous, 2022c; Aziz, 2024). Agricultural sustainability and development can be achieved by realizing and utilizing this potential.

The total area of Afghanistan is 634,547 square kilometers and it ranks 41st in the world in terms of size. Approximately 80% of the country consists of mountains, deserts, and semi-deserts, 7.2% of which is forests, 4.46% is meadows and 9.38% is residential areas. Approximately 9,610,000 hectares of land is agricultural land. A country like Afghanistan, located in an arid and semi-arid geographical region in terms of climate, struggles with the phenomenon of global "water scarcity" more than other countries in the region. Based on what has been said, due to the "water scarcity" problem, only about

half of all arable lands are cultivated and only this much area is engaged in agricultural activities. It should be noted that although Afghanistan is one of the countries with sufficient water resources, the lack of use of water resources in recent years and successive droughts have caused more than 75% of the country's water to flow to Iran. Official statistics of the Ministry of Energy and Water of Pakistan and Turkmenistan show that approximately 75 billion cubic meters of water is produced in Afghanistan every year, but people can use only 25% of it (Kabuli, 1996; Martin, 2011; Anonymous, 2022d; Hussainzada et al., 2023).

The main resources of agricultural activities are water and soil. Although Afghanistan has important underground water resources, it has not been successful in using these resources, which are vital for the country's residents, due to inaccuracies in water management and policies. In addition to the use of these weak water resources in the agricultural sector, the drilling of deep wells in major cities of this country such as Kabul destroys the underground layers. The irrigation network in Afghanistan is traditional and water wastage in this system is very high, so one of the important and effective solutions to deal with the problem of water scarcity is to create modern drip irrigation networks to minimize water wastage. Disorganization of agricultural lands and unpredictable water resources are the main problems of agricultural development in Afghanistan. Afghanistan is a country divided into two types in terms of geographical and climatic division: flat and semi-plain, and mountainous and semi-mountainous (Anonymous, 2022d; Barfield, 2022; Amiri and Habibyar, 2023). According to the information of the Afghanistan Central Bureau of Statistics, 18,000 of approximately 40,000 villages are located in mountainous regions. Approximately 12,000 villages are plain areas and the rest in semi-plain areas. The land type also differs by region. Nuristan province is 97% mountainous, Daikundi is 90% mountainous, and Herat province is 53% mountainous. As a result of these irregularities, the size of agricultural land is small and there is no opportunity to expand the cultivated area available to families. Most of these landowners cannot produce their basic food needs from these lands and are dependent on off-farm activities to obtain raw materials (Gobar, 1989; Shroder et al., 2022).

AGRICULTURAL FINANCE AND SUSTAINABLE AGRICULTURE

Sustainability has been a vital goal for all the sectors of industry, leading to the using of renewable type resources as an alternative for ecology in a rising trend worldwide (Pereira et al., 2022). Looking at the issue of sustainability from another perspective, technological development, and innovation in production in recent centuries have predominantly helped create better life conditions. On the other hand, it has also caused many difficulties for a sustainable environment. The basic source of greenhouse gas emissions is the use of fossil fuels in many areas such as providing electrical energy, agricultural production activities and transportation (Chen and Zhu, 2022). It has been stated that the concept of sustainability can be achieved by simultaneously protecting the environment, maintaining economic development by ensuring economic growth, and promoting equality (Portney, 2015). The expectation that the increase in the generally known human population will continue in the coming years and, together with the losses in arable land, will increase the role of plant disease control methods on factors such as security of food, development of economy, socially stable and landscape (Carroll et al., 2018). Sustainability innovations include improving quality at the product stage, using less input and energy, more environmentally friendly inputs, environmentally friendly packaging, recycling and reuse and eco-labelling. It aims to provide reduced emissions during the processing phase, less waste, recycling and reuse, and less input and energy (such as electricity, gas, oil, coal) consumption (Hermundsdottir and Aspelund, 2021). The fact that pesticide residues used extensively in agricultural production remain in fields, pastures, human properties, rivers, and nature for many years poses a risk to the health of people, farm animals and wildlife, and causes environmental pollution both physically and biologically (Sponsler et al., 2019).

Goals of sustainable development stated that the situation is much worse in developing countries, and that poverty and individuals being away from proper nutrition will lead to serious socio-economic losses in sectors such as human rights and education (Anonymous, 2019; Anonymous, 2023). Global epidemics and wars have negatively affected both agricultural activities and food security and have significantly increased the degree of hunger in the world (Hassen and Bilali, 2022). Changing climate conditions with the epidemic destroy crops, and extreme weather events, floods, droughts, or political turmoil have a negative impact on many sectors, especially the agricultural sector (Phillipson et al., 2020). Although efforts have long been made to develop various methods and laws to make international food security sustainable, currently approximately one in ten people face serious food insecurity (Viana et al., 2022). High food prices and lack of transparency in products lead to a lack of information, reducing consumers' demand for organic food (Ha et al., 2019). If consumers' purchasing power is eased, their expectations for safe food supply and high-quality products increase. It is estimated that approximately one-third of the increase in global food production in the last decade is due to methods applied against plant diseases (Savary et al., 2019). On the other hand, sustainability reports prepared in recent years have an important role in monitoring the economic, environmental, and social performances of businesses and in informing their stakeholders. Businesses prepare sustainability reports as a communication tool by providing information about the environment, society, and corporate responsibilities (Michelon et al., 2015).

Financial development strictly related with capital that is required for agricultural development, modern technology, and successful agricultural

solutions to be efficient. Therefore, ensuring sustainability in this regard requires financial institutions to solve the financial obstacles created in this sector with their own help and come to the aid of farmers. The sustainability reporting of many sectors was examined, and they concluded that the energy and financial sectors are making more efforts in this area (del Mar Alonso Almeida et al., 2014). Currently, the main institutions providing financial resources to Afghan farmers are traditional and local institutions that charge high interest on their loans due to the high risk of agricultural activities (Anonymous, 2022d). As it is known, people's survival depends on agriculture and agriculture is life. It is obvious that Afghanistan is an agricultural country, and agriculture has strategic importance in ensuring prosperity and selfsufficiency (Shroder, 2006). Despite of Afghanistan is an agriculturally based country; nowadays, people cannot meet their principal needs and supply of food has become the main problem of the public (Sarwary, 2023; Simangan, 2023). However, due to the increasing development of this sector, a general campaign is needed to provide a completely new agricultural reform based on sustainability in Afghanistan.

Present research aims to reveal the current problems for the development of field agriculture and keys for financial development in Afghanistan, evaluate the opportunities and present the results for sustainability.

INVESTIGATION OF THE CURRENT SITUATION OF FARMERS IN AFGHANISTAN AND THE PRINCIPLES OF SUSTAINABILITY

Afghanistan has a privilege position in world political history. The main factors about it are that the location Afghanistan country is type of intercontinental and inter-regional geographical - strategic location. Location of Afghanistan is where between Uzbekistan, China, Turkmenistan, Tajikistan, Iran, and Pakistan. Afghanistan is the most ideal crossroads for entry and exit to Central Asia, Warm Waters, and the Middle East, as well as the meeting point of Islamic, Chinese, and Indian cultures in terms of beliefs. For the control of Afghanistan, political, military, and economic conflicts called the "Great Game" took place in the 18th and 19th centuries. Afghanistan, which was previously controlled by England, came under Russian influence after it gained independence in 1918, was officially occupied by the Russians in 1979, and was cleared of occupation only after a 9-year struggle by establishing a joint front of the West and Islam. Afghanistan in the period from 1995 to 2001, when the relations between various countries and oil companies are examined, the process and reason for the occupation will be better understood, despite the fierce resistance since 2001 (Anonymous, 2022d; Ahmadi and Hikmat, 2024).

The approximate population of Afghanistan is 38,928,346 people. The data are not the latest population values of Afghanistan but are based on estimated values. As a matter of fact, it is stated as 41.1 million in some sources (Anonymous, 2024a), and as 34.3 million in others (Anonymous, 2024b). The country has the 37th largest population in the world. Approximately 85% of Afghan people are engaged in agriculture (Ball, 2008). Afghanistan has 34 cities and 398 districts. Each city in the country has its own regional government and capital. Just as there are city and district governments, regional governments also exist in the country. Kabul is the most populous city and capital of the entire country of Afghanistan. Kabul, which stands out with its economic activities and urbanization, has a population of 4 million. The total area of Afghanistan is 647,500 square kilometers. Only 2.36 million hectares of agricultural land across the country can be irrigated, and 5.46 million hectares of dry land are cultivated. Almost half of the country's arable land is cultivated (approximately 4 million hectares, corresponding to 6% of the country's total area), while most of the other half is infertile (Anonymous, 2022c; Rasouli et al., 2023). This figure, which makes most of the Afghan people directly dependent on agriculture, is a significant figure compared to

neighboring countries. Because it is possible to easily see how dependent Afghanistan is on agriculture compared to a country like India, where approximately 32% of its population is engaged in agriculture (Dupreeand and Richard, 1976; Karimi, 2023).

Unfortunately, agriculture in Afghanistan is carried out by producers with traditional systems and is far from the gifts of technology and modern agricultural methods. Apparently, producers are unable to meet the needs of a relatively small family, despite having abundant fertile land and sufficient water. For agricultural production to be highly productive and become an important industry in the country, it needs investment, attention from the government and authorities, and training and promotion of new agricultural methods. Unfortunately, official, and non-governmental organizations and international agricultural organizations responsible for this issue have not acted strongly in this field and have not been able to take significant measures. Farmers apply the same way their ancestors did 80 years ago, and their lives continue without any visible change, just like the lives of their ancestors. The small changes that took agriculture to a new stage in Afghanistan in the 1940s and 1950s failed with the onset of civil unrest. In this way, self-sufficiency will never be achieved in agricultural areas (Fazeel, 2008; Jamali et al., 2023).

The development of agriculture is a vital issue for the country, which will affect all aspects of the life, including economic, political, and even security. During travels and visits to different parts of the country, it was found that agriculture is facing similar problems in all parts of the country. If the state and non-governmental organizations try to solve these problems, agriculture will undoubtedly change, develop, and become sustainable.

In this section, the main factors affecting the sustainability of agricultural production and financial development in Afghanistan are listed.

Lack of Directors and Engineers

The difficulties and costs of administering occupied countries are obvious. The existence of professional, researcher and devoted human besides the implementation of the correct strategies to spreading and growth of agriculture snd finance in the land are the basic and critical needs for the sector. The country, which has a shortage of trained people and managers, needs to train qualified people who are experts in their fields within the understanding of security and sustainable management (Nye and Welch, 2013; Anonymous, 2022b; Miani et al., 2023).

According to business management literature, entrepreneurship is defined as extraordinary activities carried out by individuals with extraordinary mindsets with the aim of maximizing profits. Entrepreneurs perceive and accept risks differently than others by seizing unnoticed opportunities (Abu-Saifan, 2012).

Lack of Water

In agriculture and human life, water plays the role of blood in the existence of life forms, and if it is cut off for a moment, death will come. Therefore, water is considered the most basic national wealth. A country with more water is richer, and optimum use of water indicates intellectual, scientific progress and economic growth. Developed countries make maximum use of water resources in various dimensions of life, from transportation to energy production, from industry to most importantly agriculture (Anonymous, 2022b).

One of the biggest limiting factors of agriculture in Afghanistan is water scarcity. Even though Afghanistan country is rich by water resources, it faces shortage of water due to water management is not controlled and is not used propely. For example, the cities of Baghlan, Kunduz and Tahar in Afghanistan are considered the most important rice production areas. Because these regions have the necessary land and water resources for rice production. Other rice production areas in the country are the cities of Laghman, Herat and Nangarhar, which are in the temperate climate zone and have relatively warm and humid winters (Atwood, 2008).

Afghanistan is a mountainous country and rich in water resources. Nearly two hundred rivers, large and small, flow from the high mountains. The most important of these are the Amu River, Helmand River, Hari River, Kabul River, Balkh River, Baghlan River, Marghab River and Arghandab River. Unfortunately, the water of most of Afghanistan's rivers leaves this country's territory and flows into neighboring countries, and this country is the main water supplier of four neighboring countries and is considered one of their water resources. Afghanistan has experienced not only droughts, but also painful scarcity many times and for years (Anonymous, 2022b).

Most of the snow that falls in winter melts during summer season. Easter parts of Afghanistan, where the altitude is higher, an accumulation of snow that helps store water resources for a long time. Latest estimates show that the country has almost 75 billion cubic meters of potential - renewable resources of water per year, including 57 billion cubic meters of surface type water and besides groundwater that is around 18 billion cubic meters. Due to low rainfall, resources of surface water are the basic source for nutrients of the groundwater. Annual water consumption from surface waters is 20 billion cubic meters (Ahmad and Wasiq, 2004). Available data show that water resources in the country are sufficient to increase irrigation potential. However, the lack of irrigation infrastructure is seen as a very important obstacle today (Frenken and Gillet, 2012).

There is very little involvement or coordination of irrigation activities at the federal level. The supply of labor needed for the installation of canals and other infrastructure equipment for irrigation and for maintenance and repair works is provided almost entirely by the families residing here, due to the availability of land and the surface area of the villages (ICARDA, 2002). In addition to the sustainable use of agricultural lands, it is essential to bring uncultivated areas into agriculture.

Based on this, by increasing the number of irrigated lands, rehabilitating drought-tolerant plants, evaluating the lands according to their quality and determining appropriate rotation systems, a decrease in fallow areas will be achieved, thus sustainability can be achieved.

Scarcity of Agricultural Lands

The most important production factor in the agricultural sector is soil. Because it plays a fundamental role in the production process of agricultural products. Lands are also used in other sectors of the economy. One of the important problems of Afghanistan agriculture is the lack of arable land (Anonymous, 2022c). Afghanistan's agricultural lands are located mostly in the northern, western, and southwestern regions of the country, such as Kunduz, Baghlan, Belkh, Jawzjan, Faryab, Badghis, Herat, Farah, Nimroz, Helmand and Kandahar provinces (Beyoğlu, 2002). Fertile land and water resources are decreasing, so production increases must primarily result from increased efficiency and productivity. Plant breeding has contributed to significant yield increases, especially in the last 100 years. Additionally, major increases in the use of chemical fertilizers, pesticides, irrigation water, and other productivityenhancing inputs have helped increase food production and feed the growing population (Qaim, 2020).

Irregular Assignment of the Lands

The share of land in the country has not been fair, and some of the past administrations distributed these lands to parties with political and military power and strong ethnic and official connections, based on some considerations. In addition, most of the vast lands were seized by the powerful, which in turn caused a heavy tax responsibility on the people, the spread of bribery and political pressure. Thus, they caused the poor and those with less power to sell or give their lands to someone else. As the average piece size of agricultural land increases, profit and efficiency in agricultural production will increase and time loss will decrease. Land consolidation studies are very important in this respect (Anonymous, 2022c).

Inadequacy of Financial and Agricultural Policies

Considering Afghanistan is an agricultural sector-based country and the basis of country economy based on agriculture, unfortunately, this sector was not taken into consideration by some previous governments and could not be used as the country's agricultural facilities due to some administrators who did not have the knowledge. Although some measures were taken in the past for the development of agriculture, the results were not good due to the lack of competent and professional managers, the right strategy and financial development (Gazetteer and Ball, 1982). In this regard, it is of great importance that policies are especially aimed at cooperatives. The emergence of cooperatives took place gradually and is a clear sign of unity, harmony, trust, and partnership among people. Today, agricultural cooperatives are a formation that can create a leverage effect for economic development along with government policies to improve working conditions, improve production, improve social and economic conditions, and reduce poverty (Atayi, 2016).

War and Security Risk

The war and insecurity environment, which has lasted approximately 50 years, has caused great detriment to the country's agricultural configuration. This situation has evoked to requirement of import. Ahead the war during 1979, while this country was able to meet its own internal needs, it was exporting large amounts of barley, wheat, pulses, millet, vegetables and fruits to Europe, India, and the Persian Gulf countries (Hole, 2012). After 2002, with the establishment of the new Afghan government, farmers began to establish many

agricultural cooperatives to improve their living conditions, which became more difficult due to the economic problems they encountered during the wars, and to meet their needs more easily (Anonymous, 2009). The main purpose of agricultural cooperatives in Afghanistan is to cooperate with the main justifications of meeting the needs of producers, improving social, economic, and cultural foundations, and promoting economic growth based on sustainability (Rahin, 2011). However, the very long war destroyed the infrastructure, restricted agricultural development, and harmed the economic growth process. Despite all the structural problems and constraints, Afghanistan's agricultural sector still has potential for growth and development.

Lack of Knowledge of Modern Agriculture and Use of Old Methods

Agriculture in Afghanistan is done by former and traditional methods owing to the shortcomings and negligence of policies. Spring waters and streams are used to irrigate the lands, while arid lands depend on rainfall. Producers still apply ancient and traditional cultivation methods. They have little knowledge and use of new tools, technologies, and techniques such as tractors that are gps based and combine harvesters, pumping and drip irrigation, well-adapted seeds, disease – insect - weed control pesticides (Anonymous, 2022a).

So that to overcome these problems, it is necessary to support farmers in alternative activities, inform them in terms of production techniques, and implement policies to change the traditional production model. The fact that agricultural enterprises have a traditional structure and farmers cannot implement innovations in agriculture hinders sustainability. This situation causes low product diversity in daily nutrition (Abdullah et al., 2024; Cele and Mudhara, 2024; Huo et al., 2024).

Inadequate - Poor Quality Fertilization and Lack of Information on Increasing Soil Fertility

Farmers use fertilizers of animal and chemical based for crop production. Especially due to the shortage of fuel for heating purposes in the country, animal manure is used as domestic fuel, the remaining small amount is used in agriculture, and the supply of agricultural fertilizer is disrupted due to poverty and lack of chemical fertilizer. Today, many countries have established national fertilizer industries at varying scales depending on their needs or raw material resources, considering their long-term interests (Bilge and Artukoğlu, 2019).

Fertilizers are substances that restore plant nutrients lost from the soil in plant production and increase the fertility of the soil. Fertilizers are effective in increase to yield in plant production as well as improving food quality. Fertilizers, among the inputs used in plant production, provide a yield increase of over 40% (Eraslan et al., 2010). In addition, plants with sufficient nutritional element content are resistant to diseases, while plants with inadequate nutritional element content increase their susceptibility to diseases. Nutrient element, nutritional status of the plant, type of plant and disease agent are the factors that affect the resistance of the plant to the disease. Micronutrients reduce the cell penetration and infection of pathogens by affecting both the cell wall strength and the structural integrity of membranes and by having a direct toxic effect on the pathogen (Çakmak et al., 2008).

Insufficient Breeding Studies and Genetic Pool

For meet the demands of the increasing population, the need for sustainable solutions that will protect plants from various stress factors, adapt to the ecology in which they are grown, and have high and stable yield and quality qualities is of critical importance. In addition to identifying and protecting genetic resources, it is important to carry out studies that increase productivity and improve quality. Characterizing the genetic diversity and

morphological characteristics of existing genotypes, that is, the success of any breeding program for improvement, depends on the genetic variability and selection efficiency in the base population. For successful selection, it is necessary to examine the nature of the relationship of the traits of interest to other traits of interest, as well as the genetic variability available for them. Breeding programs rely heavily on the variation found in genetic resources or plant germplasm, as this increases the likelihood of obtaining suitable varieties. Over the last decade, recent techniques have been noted to serve well the goals of molecular based - biotechnological plant breeding through the sustained release of plant genome sequences. From past to present, breeders have tried to transfer the resistance resources in wild plant species to cultural forms. Various plant disease resistance genes present in wild relative species of cultivated plants have been pyramided in commercial varieties. If breeding is started with many hybrid plants, the rate of finding genotypes with desired characteristics increases (Irish et al., 2008; Yılmaz et al., 2010; Correll et al., 2011; Doruk Kahraman, 2021; Kahraman and Gökmen, 2022; 2023; Singh et al., 2022).

Feeding the world's population will require significant amendments in the global scale for food systems if it is to occur sustainably. The basic need is to produce more food by using the same or a smaller number of natural resources and to be sustainable. After a general assessment of the situation of global hunger and hidden hunger, the establishment of food security and agricultural development, especially in developing countries where poverty, hunger and malnutrition are high, it is obvious that the importance of classical and modern breeding practices and the protection of biodiversity will increase (Devaux et al., 2020).

The diversity and genetic variability of a species is the most important step in understanding its use in breeding programs. As with all plant products, the development of new varieties that can meet changing producer and consumer demands is a priority in agriculture. Breeding studies that provide the infrastructure for the fast and accurate development of new varieties and seeds are required.

Lack of Cooperative Awareness and Financial Literacy

Other important problems of farmers in this country are lack of information and industry, the absence of a proper market to sale and marketing of agricultural products, adequate vehicles and facilities for transportation and storage for agriculturally based products, absence of support by government, security problems (Doruk Kahraman and Gökmen, 2021; Hassan et al., 2024; Li et al., 2024; Salman and Wang, 2024).

Cooperatives are important organizations that contribute to sustainable economic growth, stability, and employment quality. Looking at the big picture on the subject, when fishing cooperatives are included, approximately 32% of cooperatives worldwide consist of agricultural cooperatives (ICA, 2019). As a result of the widespread use of the cooperative system and understanding its importance, its contribution to producers and the country's economy will be significant. For example, in the European Union, the business volume of agricultural cooperatives is over 210 billion Euros, and cooperatives provide 55% of agricultural inputs and market 60% of agricultural products. Cooperatives in Canada produce 35% of the world's total sugar production. In Uruguay, cooperatives produce 90% for whole production of milk, around 34 percent of whole production of honey and 30% of production of wheat (Todaro, 2012; Engin, 2018).

Systems and tools such as data analytics to supply for farmers with accurate information on requirements such as rainfall and cycle of water, plant nutrition and fertilization, introduction, and use of modern techniques, informing on the optimum period to harvest to optimize output and enabling selection of the best crops to plant. Financial support sources for access should become widespread (Eastwood et al., 2021).

The world population has increased significantly in the last decade and is projected to reach 9.5 billion by the year of 2050s. Therefore, considering the increasing globally population, ensuring security of food is feasible by designing advanced agricultural systems that can be developed to the highest-level efficiency and production by the minimum required inputs (Paravar et al., 2023).

In addition to all these issues, it is of great importance to implement sustainable agriculture principles and provide training on sustainable development – financial literacy.

Organization in Afghanistan's agricultural sector is still underdeveloped and unstructured. The biggest reason for this is long-lasting wars and internal conflicts. It is known that when producers cannot come together and act together in cooperative formations, they are forced to sell their products to traders at low prices. Cooperatives are democratic organizations that rely on and derive their power from the people. Education is an issue that should be at the forefront in the work of all organizations within the public-based cooperative movement.

MATERIAL AND METHOD

Agricultural products produced in the country, which is in a strategic location, are mostly aimed at the domestic market and agricultural self-sufficiency has not been achieved. According to published FAO data in 2019, the values of field crops production of Afghanistan were 2,872,571 ha for the cultivation area, and approximately 6.7 million tons were produced. The featured field crops are primarily wheat, corn, rice, legumes, barley, cotton, potatoes, sesame, and flax. Harvested areas for the wheat get a share 80 percent amongst field crops harvested area which is equal to 72 percent of total production. Other field crops (corn, rice, legumes, barley, cotton, potatoes, sesame, and flax) equal to almost 18 percent amongst the harvested area

(533,920 ha) that is equal to a share of 27% in the production amount (1,818,990 tons). Lack of managers and engineers, insufficiency of water, negligence of agricultural policies, war and insecurity environment, inadequacy of modern technical information, deficiencies in fertilization and plant nutrition, use of population characterized seeds that have lower yield and inadequacies in cultivation techniques - breeding studies, storage - marketing - training and publication issues. It is known that producers are experiencing significant difficulties due to deficiencies.

This research was realized according to face-to-face interviews with randomly chosen producers from the northern regions of Afghanistan in 2022, with the aim of make evaluations regarding agriculturally sustainable production and sustainable financial development. Within the scope of the study, 20 questions were asked to a total of 100 randomly selected farmers to evaluate their situation in field crop production. The data obtained are expressed as percentage (%) (Kahraman, 2017; 2023). The data obtained within the framework of the research results are listed below.

RESULTS AND DISCUSSION

In this section, the questions asked to the research participants and the answers received are listed. Abbreviations are as follows: "QUE" for Question, "ANS" for Answer.

QUESTION (QUE) 1): In how much area do you produce field crops? [1 decare (da) = 1000 square meters]

ANSWER (ANS) 1):

a) From 0 to 50 da = 45%

b) From 50 to 100 da = 34%

c) From 100 to 150 da = 9%

d) From 150 to 200 da = 7%

e) More than 200 da = 5%

According to the research results, it was determined that many of the farmers (45%) participating in the survey had very small lands (0-50 da). 34% of the producers have an area of 50-100 decares, 9% have an area of 100-150 decares, 7% have an area of 150-200 decares, and 5% have agricultural production on an area of more than 200 decares.

QUE 2): How many years have you been dealing with field crop production?

ANS 2):

- a) From 0 to 5 years = 22%
- b) From 5 to 10 years = 36%
- c) From 10 to 15 years = 15%
- d) From 15 to 20 years = 16%
- e) More than 20 years = 11%

According to the results, 22% of the surveyed producers have been farming for at most 5 years, 36% have been farming for 5-10 years, 15% have been farming for 10-15 years, 16% have been farming for 15-20 years and 11% have been farming for more than 20 years.

QUE 3): Do you apply crop rotation? ANS 3): a) Yes = 24% b) No = 76% It was determined that 24% of the farmers applied crop rotation, while the remaining 76% did not apply crop rotation. This situation is one of the main reasons for the low unit area yield values.

QUE 4): Which plants do you rotate with?

ANS 4):

- a) Wheat = 55%
- b) Potato = 5%
- c) Barley = 20%
- d) Oat = 5%
- e) Alfa alfa = 11%
- f) Tomato = 4%

According to the results of the research, it was determined that he used the plant in rotation many of the farmers (55%) who participated in the survey planted wheat, 20% planted barley, 11% planted alfalfa, 5% planted potato and oat, and 4% planted tomatoes.

QUE 5): Do you use certified seeds in the production of field crops?

ANS 5): a) Yes = 26% b) No = 74%

It was determined that most of the farmers participating in the survey (74%) did not use certified seeds in the production of field crops, while 26% used certified seeds.

QUE 6): Why don't you use certified seeds in the production of field crops?

ANS 6):

a) Due to lower yield = 14%

b) Expensive = 79%

- c) They are not tolerant for diseases = 6%
- d) Since I have my own seeds = 1%

According to the results of the research, the farmers stated that most of the farmers participating in the survey (79%) do not use certified seeds because the seeds are expensive, 14% because of low yield, 6% because they are not resistant to diseases, and 1% because they have their own seeds.

QUE 7): Where do you get the seeds for field crop production?

ANS 7):
a) Own production = 39%
b) Seed dealer = 33%
c) Other farmers = 28%

The results of the research showed that 39% of the farmers stated that they obtained seeds from their own production, 33% from seed dealers, and the remaining 28% from other farmers.

QUE 8): What is your seed sowing method in field crop production?
ANS 8):
a) Spreading = 87%
b) By seeder = 13%

While 87% of the producers who participated in the survey reported that they sowed by spreading method, the remaining 13% reported that they sowed with a seeder.

QUE 9): Do you fertilize field crops? ANS 9): a) Yes = 86% b) No = 14%

Research results determined that many of the farmers (86%) participating in the survey fertilized field crops.

QUE 10): Which fertilizers do you use in field crop production?

ANS 10):

a) Animal manure = 32%

b) Chemical manure = 68%

Most of the farmers (68%) stated that they use chemical fertilizers.

QUE 11): Which irrigation method do you use in field crop production?

ANS 11):

a) Drip irrigation = 25%

b) Flood irrigation = 49%

c) Sprinkler irrigation = %26

According to the results of the research, many of the farmers participating in the survey (49%) reported using flood irrigation, 26% using sprinkler irrigation, and the remaining 25% using the drip irrigation method.

QUE 12): Do you have any income other than field crop production? ANS 12): a) Yes = 57% b) No = 43%

Research results showed that 57% of the farmers stated that they had income other than production, and the remaining 43% stated that they had no income other than field crop production.

QUE 13): Do you have the habit of regularly following agricultural news on the Internet, TV, or newspapers?

ANS 13): a) Yes = 61% b) No = 39%

It was determined that 61 percent of the producers participating in the research acquired the habit of regularly following agricultural information through channels such as the internet, TV, and newspapers.

QUE 14): Do you apply the agricultural news you watch or read in your business?

ANS 14): a) Yes = 75% b) No = 25%

According to the results of the research, it was understood that many of the farmers (75%) who participated in the survey applied the agricultural news they watched or read in their businesses.

QUE 15): What kind of agricultural machinery, tools and equipment do you own?

ANS 15):
a) Seeder = 3%
b) Plow = 47%
c) Tractor = 50%

It was determined that 47% of the farmers owned a plow, 50% owned a tractor, and only 3% owned a seeder.

QUE 16): In your opinion, what are the most important problems of farmers regarding field crop production? (Responses were received in order of priority)

ANS 16):

a) Lack of Directors and Engineers = 29%

b) Lack of water = 21%

c) Security problems = 37%

d) Poverty = 13%

It was revealed that most of the farmers had problems due to security problems (37%) and lack of directors and engineers (29%).

QUE 17): Would you like to produce field crops again in the coming years?

ANS 17):

a) Yes - Reason: We produce until we get a better crop, if I don't produce, we won't have food = 87%

b) No - Reason: I cannot produce because I do not have the opportunity = 13%

It was determined that most of the farmers (87%) participating in the survey cultivated until they obtained a better product in order to meet their food needs. The statement of the remaining 13% stated that they will not be able to produce field crops in the coming years because they do not have the opportunity.

QUE 18): How do you comment input prices in field crop production?

ANS 18):

- a) Cheap = 47%
- b) Expensive = 50%
- c) Normal = 3%

Research results implicated that 50% of the producers participating in the survey evaluated the input prices in field crop production as expensive, 47% evaluated the input prices in field crop production as cheap, and the remaining 3% evaluated the input prices in field crop production as normal.

QUE 19): What are the people and institutions you exchange information with regarding field crop production?

ANS 19):

a) Ministry of Agriculture = 57%

b) Agricultural Engineer = 37%

c) Nobody = 6%

Results of the present research showed that 57% of the farmers reported that they consulted with the Ministry of Agriculture and 37% with Agricultural engineers regarding production.

QUE 20): After the seed sowing, how many times do you go to the field to check it until harvest?

ANS 20): a) 3 times = 3% b) 5 times = 30% c) 8 times = 38% d) Never = 29%

According to the results of the research, it was determined that most of the farmers participating in the survey (38%) went to their filed for 8 times or 5 times (30%), but 29% did not carry out control to the field.

The solutions and programs mentioned above can only be implemented under the shadow of security, a strong government, valuable and servant-qualified administrators, comprehension, and national specification, on the other hand all efforts will be in vain (Boz, 2004).

In Afghanistan, the agricultural sector acts a decisive statue in development of economic owing to most of the people earn their basic needs from the land. If the leaders of Afghanistan are serious about the human welfare, the only solution to evolve the prosperity of the plurality in a short time is to first take steps to increase the production of farmers and then expand the market network by guaranteeing the purchase of these products (Cultural, 2010). Considering agricultural facilities in Afghanistan, in case of using efficiently, it won't be enough just to provide for their own needs in terms of food requirement, also would be able to export toward the other countries in the shortest time (Omrani et al., 2011). If 6 million hectares of arable land is opened to cultivation by building dams, canals and streams, all cultivated areas in the country will reach 14 million hectares (Kohzaad, 2002).

CONCLUSIONS

Based on the results of the research, the important issues for the development of agriculture and achieving sustainable development in Afghanistan are summarized as follows.

- 1- Hiring qualified managers and training qualified people.
- 2- Storing and holding of water, reducing fallow areas.
- 3- Planning in the inheritance system and integration of the fields.
- 4- Carrying out reforms in terms of ownership and management of agricultural lands.
- 5- Development of deep-rooted policies for agricultural sustainability and financial development, state support for farmers and expansion of banks that will invest in this sector.
- 6- Urgently stop of war, confusion, and chaos with the support of all states and the restructuring of security.
- 7- Introducing farmers to new agricultural technologies and modernizing agriculture.
- 8- Establishing the fertilizer supply chain, understanding the principles of plant nutrition, and introducing and extending the principles of sustainability in field use and finance.

- 9- Using classical and modern breeding methods, developing new plant varieties with high adaptability, efficiency, and quality high added value, demanded in the world markets - and placing them in rotation systems.
- 10- Establishing storage systems for agricultural products, establishing a marketing network, and preparing transportation routes, popularizing, and adopting cooperatives, carrying out agricultural - financial literacy education and publication activities more effectively, carrying out studies for the development of farmers and encouraging modern agriculture.

It is essential to implement sustainable education projects for farmers to inform them about new farming methods that have given way to traditional agriculture in Afghanistan. Afghan farmers need to have knowledge about the types and methods of spraying, irrigation methods and cultivation stages of different products to combat diseases and pests. One of the important deficiencies is that the products are not protected from pests in different seasons since pesticides are not distributed to farmers at low prices and on time.

Another important deficiency is the lack of a precise agricultural calendar for different regions of the country so that different products can be grown at certain times and in certain areas. The existence of such a calendar will greatly help increase the productivity of a country's agricultural lands and thus ensure sustainability. Another important problem is that there are no active agricultural cooperatives in the country that will provide the necessary facilities to farmers. The lack of modern high-capacity silo that can store different products is an important problem for the farmers. The construction of silos all over the country is vital for a modern agriculture.

It is necessary to establish short-term training workshops to provide farmers with basic training, especially on irrigation, planting, and harvesting. Similarly, workshops teaching farmers how to make the most of existing facilities are required. There are no agricultural banks to solve farmers' financial problems. There is a marketing weakness regarding the sale of agricultural products in domestic and foreign markets. Another problem is that chemical fertilizers and waste are not distributed correctly and on time. The extreme poverty of the villagers and their lack of access to new agricultural facilities and the fact that producers still use the basis of several hundred years ago are serious problems. It is of great importance that an accurate and comprehensive agricultural strategy is urgently created and that the government takes seriously the agricultural sector in Afghanistan as a key sector of economic growth.

There is a great need to obtain accurate information about Afghanistan's agricultural lands based on a professional survey. Data on the distribution of available land and irrigated land are still unclear and there is no specific research in this area. Accurate information based on accurate surveys forms the basis of agricultural policies - cooperative awareness and financial development. Addressing the above-mentioned issues systematically and multidisciplinary to ensure sustainability will make serious contributions to Afghanistan agriculture, financial development, human welfare, complete independence and so freedom.

REFERENCES

- Abdullah, H. M., Islam, M. N., Saikat, M. H., & Bhuiyan, M. A. (2024). Precision agriculture practices from planting to postharvest: scopes, opportunities, and challenges of innovation in developing countries. *Remote Sensing in Precision Agriculture*, 3-26.
- Abu-Saifan, S. (2012). Social entrepreneurship: definition and boundaries. *Technology innovation management review 2012; 2(2): 22-27.*
- Ahmad, M., & Wasiq, M. (2004). Water resource development in Northern Afganistan and its implications for Amu Darya Basin(No. 36). World Bank Publications.
- Ahmadi, R., & Hikmat, C. A. (2024). The Factors of the Fall of the Republic Government and Political Crisis in Afghanistan: A Survey of Public Attitudes. *Journal of Contemporary Philosophical and Anthropological Studies*, 2(1).
- Amiri, Z., & Habibyar, S. (2023). A Descriptive Look at the Factors Influencing the Geopolitics of Afghanistan: The Geopolitics of Afghanistan. *Journal for Research in Applied Sciences and Biotechnology*, 2(2), 176-181.
- Anonymous. (2009). Afghanistan, Tarım, Sulama ve Hayvancılık Bakanlığı, Amar Sabti Kooperatifha ba Meyarat Jadid. MAİL: https://www.mail.gov.af/dr/node/1097.
- Anonymous. (2019). https://unstats.un.org/sdgs/report/2019/The-Sustainable-Development-Goals- Report-2019.pdf.
- Anonymous. (2022a). https://moec.gov.af Afgan Ekonomi Bakanlığı (Erişim tarihi: 16.12.2022).
- Anonymous. (2022b). https://www.mail.gov.af Afgan Tarım Bakanlığı (Erişim tarihi: 11.12.2022).

- Anonymous. (2022c). https://momp.gov.af/dr/ Afgan Maden Bakanlığı (Erişim tarihi: 11.12.2022).
- Anonymous. (2022d). https://directory.ooyta.com/en/about/aima-afghanistanislamic-medical- association-1.html Afgan İstatistik Bakanlığı (Erişim tarihi: 11.12.2022).
- Anonymous. (2022e). https://www.findatour.com (Erişim tarihi: 11.12.2022).
- Anonymous. (2023). https://www.undp.org/sustainable-development-goals.
- Anonymous. (2024a). https://www.mfa.gov.tr/afganistan-kunyesi.tr.mfa.
- Anonymous. (2024b). https://tr.tradingeconomics.com/afghanistan/population.
- Atayi, A. (2016). The First National Conference of Afghanistan Agricultural Cooperatives. Dari.wadsam: https://dari.wadsam.com.
- Atwood, R. (2008). Afghanistan's hidden treasures. *National Geographic*, *213*(6), 130-145.
- Aziz, A. (2024). Strategic Dimensions: CPEC's Influence on Pakistan's New Geo-economics Narrative. *Jahan-e-Tahqeeq*, 7(1), 136-146.
- Ball, W. (2008). The Monuments of Afghanistan, p. 45-50 (in English).
- Barfield, T. J. (2022). Afghanistan: A cultural and political history.
- Ben Hassen, T., & El Bilali, H. (2022). Impacts of the Russia-Ukraine war on global food security: towards more sustainable and resilient food systems? *Foods*, 11(15), 2301.
- Beyoğlu, A. (2002). Afganistan Üzerine Araştırmalar. p 3-12 (in English).
- Bilge, B., & Artukoğlu, M. (2019). Türkiye'de Son Yıllarda Gübrede Uygulanan Politikalara Genel Bir Bakış. *Tarım Ekonomisi Dergisi*, 25(2), 275-281.
- Boz, İ. (2004). Tarım Sektörünün İktisadi Kalkınmadaki Rolü. İçinde, Kalkınma Ekonomisi: Seçme Konular, Ed: Sami Taban-Muhsin Kar, Ekin Kitabevi Yayınları, Bursa, p137-158.
- Çakmak, Ö., Aysan, Y., & Erdem, H. (2008). Farklı düzeylerde çinko beslenmesi altındaki domates bitkilerinde bakteriyel solgunluk

hastalığı üzerine bitki büyüme düzenleyicilerinin etkisinin araştırılması. 1070273 *no'lu tübitak araştırma projesi sonuç raporu. 49 s. In: Adana.*

- Carroll, C. L., Carter, C. A., Goodhue, R. E., Lawell, C.-Y. C. L., & Subbarao,
 K. V. (2018). A review of control options and externalities for
 Verticillium wilts. *Phytopathology*, 108(2), 160-171.
- Cele, T., & Mudhara, M. (2024). Impacts of Crop Production and Value Chains on Household Food Insecurity in Kwazulu-Natal: An Ordered Probit Analysis. *Sustainability*, 16(2), 700.
- Chen, Y., & Zhu, Z. (2022). Liability structure and carbon emissions abatement: Evidence from Chinese manufacturing enterprises. *Environmental and Resource Economics*, 83(2), 481-507.
- Correll, J., Bluhm, B., Feng, C., Lamour, K., Du Toit, L., & Koike, S. (2011). Spinach: better management of downy mildew and white rust through genomics. *European journal of plant pathology*, *129*, 193-205.
- Cultural, P. (2010). Training Resource, Afganistan, Alexander the Great, p 330-323 BC (in English).
- Davies, S., Akram, I., Ali, M. T., Hafeez, M., & Ringler, C. (2024). The economywide impacts of increasing water security through policies on agricultural production: The case of rice and sugarcane in Pakistan. Intl Food Policy Res Inst.
- Del Mar Alonso-Almeida, M., Llach, J., & Marimon, F. (2014). A closer look at the 'Global Reporting Initiative'sustainability reporting as a tool to implement environmental and social policies: A worldwide sector analysis. Corporate Social Responsibility and Environmental Management, 21(6), 318-335.
- Devaux, A., Goffart, J.-P., Petsakos, A., Kromann, P., Gatto, M., Okello, J., Suarez, V., & Hareau, G. (2020). Global food security, contributions

from sustainable potato agri-food systems. *The potato crop: Its agricultural, nutritional and social contribution to humankind*, 3-35.

- Doruk Kahraman, N. (2021). Mikrospor kültürü ile haploit bitki üretimi. International Anatolian Congress on Medicinal and Aromatic Plants, 2021, Konya, Turkiye.
- Doruk Kahraman, N., & Gokmen, S. (2021). Turkey's bulgur durum wheat production potential. 2nd International Congress of the Turkish Journal of Agriculture-Food Science and Technology (TURJAF 2021),
- Doruk Kahraman, N., & Kahraman, A. (2023). Economic sustainability in agriculture: Importance of cool-season cereals. *International Congress* of Finance and Tax (pp. 213-219),
- Dupree and, L., Richard, S. (1776). Afganistan Kabul ve Ziraat p 3-29 (in English).
- Dupree, N. (1977). An historical guide to Afghanistan: Afghan Air Authority, Afghan Tourist Organization. Verlag: Afghan Air Authority, Afghan Tourist Organization p. 492. Retrieved 2010-08-22 (in English).
- Eastwood, C. R., Edwards, J., & Turner, J. A. (2021). Anticipating alternative trajectories for responsible Agriculture 4.0 innovation in livestock systems. *Animal*, *15*, 100296. https://doi.org/10.1016/j.animal.2021.100296
- Engin, M. (2018). Tarım Kredi Kooperatiflerinde Çok Amaçlılık Politikalarının, Kooperatif Ortakları Açısından Değerlendirilmesi. Namık Kemal University Graduate School of Natural and Applied Sciences Department of Agricultural Economics. Basılmamış Yüksek Lisans Tezi. Namık Kemal Üniversitesi Fen Bilimleri Enstitüsü Tarım Ekonomisi Ana Bilim Dalı, 99, Tekirdağ.
- Eraslan, F., İnal, A., Güneş, A., Erdal, İ., & Coşkan, A. (2010). Türkiye'de kimyasal gübre üretim ve tüketim durumu, sorunlar, çözüm önerileri ve

yenilikler. Tmmob ziraat mühendisleri odası, ziraat mühendisliği vii. Teknik kongresi, 11, 15.

- Fazeel, F. (2008). Tarım Ürünleri Pazarlamasının Temelleri, Shahrno Kabil, Kabil Üniversitesi, Sabour Basım ve Endüstriyel Matbaa, p 1 (in Persian).
- Frenken, K., & Gillet, V. (2012). Sulama suyu ihtiyacı ve ülkeye göre su çekilmesi. FAO.
- Gazetteer, A., & Ball, W. (1982). Afganistan Volume, Recherche Surles Civilisations, Paris.
- Gobar, M. (1989). Tarihin yolunda Afganistan. Kabul-Afganistan. (in Persian).
- Ha, T. M., Shakur, S., & Do, K. H. P. (2019). Rural-urban differences in willingness to pay for organic vegetables: Evidence from Vietnam. *Appetite*, 141, 104273.
- Hassan, A., Akhtar, S., & Ishaq, M. (2024). Analysis of livelihood asset of agricultural households influencing livelihood strategies in Khyber Pakhtunkhwa Pakistan. Sarhad Journal of Agriculture, 40(1), 54-63.
- Hermundsdottir, F., & Aspelund, A. (2021). Sustainability innovations and firm competitiveness: A review. *Journal of Cleaner Production*, 280, 124715. Abdullah, H. M., Islam, M. N., Saikat, M. H., & Bhuiyan, M. A. (2024). Precision agriculture practices from planting to postharvest: scopes, opportunities, and challenges of innovation in developing countries. *Remote Sensing in Precision Agriculture*, 3-26.
- Hole, F. (2012). Neolthic Age in Afganistan. Kabul-Afganistan. p 5-15 (in Persian).
- Huo, D., Malik, A. W., Ravana, S. D., Rahman, A. U., & Ahmedy, I. (2024). Mapping smart farming: Addressing agricultural challenges in datadriven era. *Renewable and Sustainable Energy Reviews*, 189, 113858.
- Hussainzada, W., Cabrera, J. S., Samim, A. T., & Lee, H. S. (2023). Water resource management for improved crop cultivation and productivity

with hydraulic engineering solution in arid northern Afghanistan. *Applied Water Science*, *13*(2), 41.

- ICA. (2019). Exploring The Cooperative Economy. monitor.coop: https://monitor.coop/sites/default/files/publication-files/wcm2019final-1671449250.pdf.
- ICARDA. (2002). International Center for Agricultural Research in the Dry Areas, 2002. Needs Assessment on Soil and Water in Afghanistan. Aleppo.
- Irish, B., Correll, J., Feng, C., Bentley, T., & de Los Reyes, B. (2008). Characterization of a resistance locus (Pfs-1) to the spinach downy mildew pathogen (Peronospora farinosa f. sp. spinaciae) and development of a molecular marker linked to Pfs-1. *Phytopathology*, 98(8), 894-900.
- Jamali, A., Lalzai, F., & Jamal, N. (2023). Marketing Constraints and Price Perspectives for Onion in Khost Province, Afghanistan. *Journal for Research in Applied Sciences and Biotechnology*, 2, 1-7.
- Kabuli, M. (1996). Ariana ansiklopedisi, Afganistan tarihi. Kabul-Afganistan (in Persian).
- Kahraman, A. (2017). Evaluation of Farming Legumes in United States of America. Yuzuncu Yıl University Journal of Agricultural Sciences, 27(1), 51-57.
- Kahraman, A. (2023). Evaluation of legume farming in Turkey and agricultural sustainability. *Legume Research-An International Journal*, 46(2), 166-170. https://doi.org/10.18805/LRF-697
- Kahraman, A., Doruk, N., & Gökmen, S. (2022). Konya Kurak Koşullarında Makarnalık Buğdayda Bazı Fenolojik ve Morfolojik Özellikler ile Verim ve Verim Unsurlarının Belirlenmesi. Bahri Dağdaş Bitkisel Araştırma Dergisi, 11(1), 40-48.

- Kahraman, N. D., & Gökmen, S. (2023). Determination of some quality characteristics of durum wheat under dry conditions in Konya. *Selcuk Journal of Agriculture and Food Sciences*, 37(1), 64-71.
- Karimi, A. (2023). Afganistan antepfistiği ve çam fistiği sektörünün mevcut durumu Bursa Uludag University (Turkey)].
- Kohzaad, A. A. (2002). AfghanistanHistory. *Kabul-Afganistan.p* 42, (in *Persian*).
- Li, P., Abbas, J., Balsalobre-Lorente, D., Wang, Q., Zhang, Q., & Shah, S. A.
 R. (2024). Impact of sectoral mix on environmental sustainability: How is heterogeneity addressed? *Gondwana Research*, *128*, 86-105.
- Martin, M. A. B. (2011). Geopolitical analysis of Afghanistan. Prebie 3(2): 7.
- Meraj, J., Ahmed, P., & Abbas, M. G. (2024). Understanding the Dynamics: The Impact of Exchange Rate Fluctuations on Pakistan's Economic Landscape. *Journal of Development and Social Sciences*, 5(1), 211-223.
- Miani, A. M., Dehkordi, M. K., Siamian, N., Lassois, L., Tan, R., & Azadi, H. (2023). Toward sustainable rural livelihoods approach: Application of grounded theory in Ghazni province, Afghanistan. *Applied Geography*, 154, 102915.
- Michelon, G., Pilonato, S., & Ricceri, F. (2015). CSR reporting practices and the quality of disclosure: An empirical analysis. *Critical perspectives on accounting*, *33*, 59-78.

- Mujtaba, G., Shah, M. U. H., Hai, A., Daud, M., & Hayat, M. (2024). A holistic approach to embracing the United Nation's Sustainable Development Goal (SDG-6) towards water security in Pakistan. *Journal of Water Process Engineering*, 57, 104691.
- Nye Jr., J. S., Welch D. A. (2013). Küresel çatışmayı ve İşbirliğini Anlamak, Türkiye İş Bankası Kültür Yayınları, *3. Basım. Çev. Renan Akman, İstanbul.*
- Omrani, B., & Leeming, M. (2011). Afghanistan: A companion and guide (2nd ed.). *Hong Kong: Odyssey Illustrated Guides*.
- Paravar, A., Piri, R., Balouchi, H., & Ma, Y. (2023). Microbial seed coating: an attractive tool for sustainable agriculture. *Biotechnology Reports*, e00781.
- Phillipson, J., Gorton, M., Turner, R., Shucksmith, M., Aitken-McDermott, K., Areal, F., Cowie, P., Hubbard, C., Maioli, S., & McAreavey, R. (2020). The COVID-19 pandemic and its implications for rural economies. *Sustainability*, 12(10), 3973.
- Portney, K. E. (2015). Sustainability. The MIT press essential knowledge series the Massachusetts Institute of Technology (MIT) press, *Cambridge, Massachusetts. Sustainability.*
- Qaim, M. (2020). Role of new plant breeding technologies for food security and sustainable agricultural development. *Applied Economic Perspectives and Policy*, 42(2), 129-150. https://doi.org/10.1002/aepp.13044129.

Rahin, M. (2011). Modiriyet dar Rahbari Kooperatifhai Ziraati. Karna Dergisi.

Rasouli, H., Vaseashta, A., & Belhassan, K. (2023). Mechanical Analysis of Khair Abad Village, Surskhrud District, Nangarhar Province, Afghanistan. *International Journal of Earth Sciences Knowledge and Applications*, 5(1), 103-120.

- Salman, M., & Wang, G. (2024). The impact of National Environmental Policy on Pakistan's green economic development: evidence from regression discontinuity design. *Environment, Development and Sustainability*, 1-30.
- Santo Pereira, A. d. E., de Oliveira, J. L., Savassa, S. M., Rogério, C. B., de Medeiros, G. A., & Fraceto, L. F. (2022). Lignin nanoparticles: New insights for a sustainable agriculture. *Journal of Cleaner Production*, 345, 131145.
- Sarwary, M., Samiappan, S., Khan, G. D., & Moahid, M. (2023). Climate Change and Cereal Crops Productivity in Afghanistan: Evidence Based on Panel Regression Model. *Sustainability*, 15(14), 10963.
- Savary, S., Willocquet, L., Pethybridge, S. J., Esker, P., McRoberts, N., & Nelson, A. (2019). The global burden of pathogens and pests on major food crops. *Nature ecology & evolution*, 3(3), 430-439.
- Shroder, J. (2006). Afganistan ve Ziraat. Kabul-Afganistan (in Persian).
- Shroder, J. F., Eqrar, N., Waizy, H., Ahmadi, H., & Weihs, B. J. (2022). Review of the Geology of Afghanistan and its water resources. *International Geology Review*, 64(7), 1009-1031.
- Simangan, D., Bose, S., Candelaria, J. L., Krampe, F., & Kaneko, S. (2023). Positive peace and environmental sustainability: Local evidence from Afghanistan and Nepal. *Environment and Security*, 1(3-4), 142-162.
- Singh, A., Uttamrao, T. M., Topgyal, T., Sharma, M., Chand, G., Sharma, M.,
 & Gupta, M. (2022). Genetic diversity analysis of common bean (Phaseolus vulgaris L.) based on SSR marker.
- Sponsler, D. B., Grozinger, C. M., Hitaj, C., Rundlöf, M., Botías, C., Code, A., Lonsdorf, E. V., Melathopoulos, A. P., Smith, D. J., & Suryanarayanan,
 S. (2019). Pesticides and pollinators: A socioecological synthesis. *Science of the Total Environment*, 662, 1012-1027.

- Sridhar, A., Balakrishnan, A., Jacob, M. M., Sillanpää, M., & Dayanandan, N. (2023). Global impact of COVID-19 on agriculture: role of sustainable agriculture and digital farming. *Environmental Science and Pollution Research*, 30(15), 42509-42525. https://doi.org/10.1007/s11356-022-19358-w
- Todaro. (2012). Tosiye İktisadi dar Keshwarhai Jahan Sevom. Roshangaran Dergisi, Tahran.
- Viana, C. M., Freire, D., Abrantes, P., Rocha, J., & Pereira, P. (2022). Agricultural land systems importance for supporting food security and sustainable development goals: A systematic review. Science of the Total Environment, 806, 150718.
- Yılmaz, G., Kandemir, N., & Yanar, Y. (2010). Bazı Patates Melezlerinden Yeni Klonların Seçimi ve Başçiftlik Yerel Patates Çeşidinin Moleküler Karakterizasyonu. Sonuç Raporu, TÜBİTAK–TOVAG. Proje(106).
- Yılmaz, K., & Tilbe, F. K. (2023). Tarım ve Hayvancılık Sektöründe Çalışan Afgan Göçmenlerde Ekonomik Konumlanma ve Entegrasyon: Trakya Bölgesi Örneği. *MANAS Sosyal Araştırmalar Dergisi*, 12(4), 1413-1424.

CHAPTER 2

MOLECULAR STUDIES IN HAZELNUT

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INTRODUCTION

The hazelnut which belongs to *Corylus* genus in *Betulaceae* family is found in southern Europe in Spain, Sicily and Greece starting from Türkiye in Asia and spreading from the Caucasus to İran in the east and from Taurus Mountains in Anatolia to Syria and Lebanon in the south. Hazelnut grows in 36°- 41° north latitudes and under specific climate conditions. Of the 13 commonly recognized species of *Corylus*, only the European species (*C. avellana*) is widely planted for commercial production. According to 2022 FAO data, hazelnut has been mostly cultivated respectively in Türkiye (765.000 tons), Italy (98.670 tons), Azerbaijan (72.105 tons), USA (70.310 tons) and Georgia (33.400 tons). It is also cultivated in Chile (62.557 tons), China (24.696 tons), Iran (13.407 tons) and Spain (8.040 tons). Türkiye is the biggest hazelnut producer in the world, supplying solely about 70 % of world production.

Hazelnut (Corylus avellana) has impressive health benefits, wide range of usage areas and high economical importance in international trade. Hence, studies aiming to protecting and improving genotypes and boost production efficiency in hazelnut cultivation have been becoming increasingly more popular. In this context, breeding activities which are one of the most effective tools in solving many problems in plant production also in the foreground. New technologies in plant breeding provide significant advantages both in terms of time and in explaining issues where classical breeding methods are insufficient. Molecular methods are at the forefront of these technologies. Nowadays, with the development of molecular methods, many problems have been tried to be solved by the help of molecular studies. Molecular markers are used in plant studies for a wide of range of purposes including identification of genotypes, determination of genetic relations and diversity, investigation of genetic structures of germlasms, registration of varieties, identification of breedling lines and hybridity and varietal purity tests, assessment of plant resistance and adaptation capability, DNA marker mapping and study of simple inherited traits

with Marker-Assisted Selection (MAS) process, Quantitative Trait Loci (QTL) analyses and gene pyramiding. In the past 25 years, germplasm collections have been enlarged and a substantial body of information on the genetic control of traits generated. There has been an increase in marker assisted selection studies which have been conducted for many features. This situation has brought a different dimension to hazelnut breeding trials.

Molecular markers have allowed studies of genetic diversity in the genus Corylus, construction of linkage maps, and marker-assisted selection for disease resistance. Genome and transcriptome sequences have been mined for DNA markers, and provide a foundation for the isolation of important genes. Application of classical methods in breeding studies conducted on identification of diseases and pests that could impact on hazelnut production, of characterization isolates. molecular interspecies hybridization. determination of pollen-stigma incompatibility, identification of hazelnut genotypes and determination of genetic relations is too much time-taking and provides insufficient data. Such drawbacks can be eliminated by application of molecular markers

In this section the general status of molecular studies on hazelnut in the world is summarised and evaluated, according to study purposes.

1.1. Molecular Studies on Genetic Mapping of Hazelnut

Hazelnut is an economically important tree nut for which demand currently exceeds supply. In response to this economic opportunity, plantings have expanded in many current production areas, and orchards have been established in areas new to hazelnut production. Nearly all world production is based on selections from the local, wild vegetation, and public breeding programs to improve *Corylus avellana* were not initiated until the 1960s. In the past 25 years, germplasm collections have been enlarged and a substantial body of information on the genetic control of traits generated.

Molecular markers have allowed studies of genetic diversity in the genus *Corylus*, construction of linkage maps, and marker-assisted selection for disease resistance. Genome and transcriptome sequences have been mined for DNA markers, and provide a foundation for the isolation of important genes.

SSR markers are multiallelic, codominant, and suitable for automated analysis. In addition, primer sequences and data can be shared among laboratories. More than 330 SSR markers have been developed for hazelnut (Bassil et al., 2005a, 2005b; Boccacci et al., 2005; Gürcan et al., 2010a, 2010b; Gürcan & Mehlenbacher, 2010a, 2010b; Peterschmidt, 2013; Sathuvalli & Mehlenbacher, 2013) and the majority have been placed on the linkage map. The first linkage map consisting of 10 LGs covering 452 cM was constructed based on molecular analysis and field observations.

Mehlenbacher et al. (2006) have constructed a linkage map of European hazelnut by RAPD and SSR markers using 144 seedlings from the cross 'OSU 252.146' X 'OSU 414.062'. The maternal map included 249 RAPD and 20 SSR markers and spanned a distance of 661 cM. The paternal map included 271 RAPD and 28 SSR markers and spanned a distance of 812 cM. The maps were found to be quite dense, with an average of 2.6 cM between adjacent markers. In the same study, the S-locus, which controls pollen-stigma incompatibility, was placed on chromosome 5S where 6 markers were linked within a distance of 10 cM. Polymorphisms in coding regions are useful as molecular markers. Particularly SNPs are suggested to be applied in high efficiency genotyping studies. Gürcan & Mehlenbacher (2010a) developed polymorphic microsatellite markers for European hazelnut from the sequences of ISSR fragments and SSR flanking regions. 25 ISSR primers were used to generate fragments for cloning. Of the 70 loci developed from ISSR and SSR flanking sequences, 50 segregated in the mapping population and were assigned to linkage groups. Beltramo et al. (2014) reported that, a F1 progeny of the cross 'Tonda Gentile delle Langhe' × 'Hall's Giant' was obtained and plants of the

progeny were analyzed at 92 SSR loci. The initial results indicated a large variability of the traits for vigour and time of budburst. Bhattarai & Mehlenbacher (2018) mined the 'Jefferson' genome sequence to develop new polymorphic SSR markers with repeat motifs of 4, 5, or 6 bp and further saturated the reference hazelnut genetic linkage map.

The first QTL analyses for hazelnut were presented by Beltramo et al. (2016). Fifteen QTLs were identified, including at least one major QTL for each of the three traits. A major QTL on LG_02 for time of leaf bud burst explained about 50 % of the PV. This is an initial step in the identification of chromosomal regions carrying genes of interest, which will be important for breeding programs and allow marker-assisted selection.

Valentini et al. (2021) discussed a rational approach for mapping QTL for phenology-related traits, such as the time of male and female flowering, dichogamy, and the period required for nut maturation, with the aim of identifying quantitative trait loci (QTL) and specific genes associated with plant phenology. Overall, 71 QTLs were detected, of which 20 were identified as contributing to the time of male flowering, 15 to the time of female flowering, 25 to dichogamy, and 11 to the time of nut maturity.

1.2. Molecular Studies on Identification of Hazelnut Genotypes and Determination of Genetic Relations

Isoenzymes began to be used over time due to difficulties such as not fully knowing the genetic control mechanism of phenotypic traits, insufficient variation and the long time it takes for the desired phenotypic traits to appear at the appropriate growth stage, but the impact of environmental factors on biochemical properties led plant breeders to molecular markers. The identification of both existing and existing genotypes obtained through selection studies is the most common use of molecular markers in plants. Research on this subject is also frequently encountered in hazelnut. The taxonomy of *Corylus* has been investigated since the mid-nineteenth century, with the number of recognized species dependent on the emphasis placed by various authors on certain anatomical and morphological characters (Whitcher & Wen, 2001). Due to its outcrossing, highly heterozygous nature, substantial genetic diversity can be found in the pool of existing cultivars. This wide diversity, expressed as morphological, phenological, and DNA sequence variability, has been discussed by numerous authors, including more recently Mehlenbacher, (1991); Mehlenbacher, (1997); Boccacci & Botta (2010); Boccacci et al. (2008); Ferreira et al. (2010). World germplasm collections hold numerous cultivars, many of which are well characterized and, in recent years, have become more readily available for use in breeding and research efforts (Boccacci et al., 2008).

Corylus species were classified into 4 groups to examine genetic relationships based on ITS region of the nuclear ribosomal DNA and chloroplast matK gene. The groups were assigned as follows: Group 1: *C. avellana*, *C. maxima*, *C. americana*, *C. heterophylla*; Group 2: *C. colurna*, *C. chinensis*, *C. jacquemontii*; Group 3: *C. cornuta*, *C. california*, *C. sieboldiana* and Group 4: *C. ferox*. Groups 1 and 2 included the shrub species while Group 3 consisted of the tree species (Erdoğan, 1999).

Galderisi et al., (1999) aimed to identify six hazelnut cultivars widespread in the Campania region (south Italy) by using RAPD technique. The analysed hazelnut cultivars were successfully distinguished by their RAPD fingerprints using the DNA primers U2, U3, U4, U11 and U14.

Miaja et al. (2001) performed RAPD-marker based characterization of 19 Italian hazelnut varieties and genotypes by using 30 RAPD primers. The dendrogram had two major groups of cultivars. The cultuvars 'Tombul', 'Imperiale de Trebizonde', 'Fructo Rubro' and 'Jean's' were in the same group all of which have tubular husks. In order to compare the genetic profiles of hazelnut cultivars 'Tonda di Giffoni', 'Negret', 'Cosford' and 23 clones of 'Tonda Gentile delle Langhe' and to detect any intra-varietal variability, Valentini et al. (2001) used 20 RAPD primers in 10 bp in size. The intra-varietal variability was detected with the 16 primers and no polymorphism was identified between 'Tonda Gentile delle Langhe' clones.

Nas et al. (2004) were conducted to assess clonal stability of hazelnuts generated from axillary buds cultured in vitro for long-term using RAPD. RAPD analysis did not reveal any somaclonal variation between donor plants from which in vitro cultures were initiated and micropropagated plants and no somacional variation was detected among in vitro-propagated plants. However, polymorphism (15.6%) was detected between the parent plant and its in vitro-propagated progenies.

Gantner & Okon (2009) assessed the genetic variability between six hazelnut cultivars which grow in Poland using RAPD markers. 30 decamer primers were selected from 80 primers. These primers yielded a total of 243 bands, of which 146 were polymorphic. The phylogenetic tree showed that only three cultivars clustered together.

Kafkas & Doğan (2009) investigated genetic relationships among 18 Turkish hazelnut by using 25 RAPD primers, 25 ISSR primers, and eight AFLP primer pairs which generated a total of 434 polymorphic marker loci. Genetic similarity index values ranged from a high of 0.96 for 'Kan' and 'UzunMusa' to a low of 0.73 for 'Yassi Badem' and 'Kalinkara'.

In another study (Martins et al., 2009), Turkish and Portuguese hazelnut genotypes and three testers ('Butler', 'Merveille' and 'Longue d'Espagne' of hazelnut) were screened using 20 RAPD markers and 18 ISSR markers. The dendrograms generated with RAPD, ISSR and RAPD plus ISSR data, grouped the 18 genotypes in two main clusters. The first cluster, which contains all

hazelnut genotypes, could be divided into four sub-clusters. The second cluster was formed only by the Turkish hazelnut.

Yılmaz (2009) aimed to define the pomological, morphological and genetic characterization of 16 Turkish hazelnut varieties and 64 genotypes by using RAPD and SSR methods in molecular characterization. The highest polymorhism (98 %) was detected with SSR followed by RAPD (81 %). Similarity coefficients in overall genotypes were between 0.12-0.98 in SSR and 0.64-0.97 in RAPD.

Ferreira et al. (2010) aimed to describe phenotypic variation in nut and husk traits and investigated genetic relationships among selections and cultivars using ISSR markers. The results suggested that the local accessions are closely related to each other, but are relatively distant from the standard cultivars of eastern Spain, Italy and the USA.

Demir (2014) determined the molecular profile of hazelnut cultivars and accessions grown in Türkiye, and assessed their genetic relationships using 15 Turkish hazelnut cultivars and 12 hazelnut accessions. Genetic relationships of hazelnut cultivars and accessions were assessed using 22 RAPD primers. The best polymorphism was obtained from OPAD-02 (5 polymorphic bands) primer. UPGMA cluster analysis of the data separated the 27 genotypes into two main groups. Most of the hazelnut cultivars were settled on the first group while 'Kalınkara', 'İncekara' and 'Mincane' cultivars and 'FAE-190' accession were placed on the second group. Depending on the genotypes, similarity ratios ranged between 0.364-0.974, with a mean value of 0.697. Overall, the results demonstrated a high level of polymorphism among hazelnut cultivars and accessions in Türkiye.

Mohammedzedeh et al. (2014) investigated the phenotypic variation in morphological traits and genetic relationships among genotypes and cultivars of hazelnut using ISSR and RAPD markers. The genotypes were found to be phenotypically diverse. 10 ISSR and 15 RAPD primers, which generated 140 and 188 polymorphic fragments, respectively, were able to distinguish genetic variation among genotypes. The dendrograms based on both molecular data showed two main clusters with high variability between genotypes.

Karakaya et al. (2023) aimed to assess phenotypic variation in nut traits and genetic diversity revealed by inter simple sequences repeat (ISSR) markers in hazelnut germplasm resources from the Eastern Black Sea Region. Molecular marker analysis yielded a total of 80 bands, 74 of which were polymorphic, from 9 inter simple sequence repeat primers. The rate of polymorphism varied between 66.7 % and 100.0 %. Similarity index of hazelnut cultivars and accessions was computed between 0.46 and 0.88. Study suggested that the accession H-1 with superior nut traits could be good genetic material for the development of new cultivars in future hazelnut breeding efforts.

SSR markers have been used to fingerprint cultivars and characterize germplasm collections (Boccacci et al., 2006, 2008; Gökirmak et al., 2009; Gürcan et al., 2010b; Sathuvalli & Mehlenbacher, 2013). Most of the world's cultivars are selections from local wild vegetation. In hazelnut, SSR markers were initially developed from DNA libraries enriched for specific repeats (Bassil et al., 2005a, 2005b; Boccacci et al., 2005; Gürcan et al., 2010a). Based on simple sequence repeat markers, most cultivars have been assigned to one of four major geographical groups: Central European, Black Sea, English or Spanish-Italian (Boccacci et al., 2006; Gökirmak et al., 2009). SSR markers are widely used in plant genetics and breeding, including studies of genetic diversity and evolution, association, gene flow, genetic mapping, gene tagging, gene cloning, cultivar identification, parentage analysis, identification of duplicates, marker-assisted selection (MAS), and quantitative trait locus (QTL) analysis (Hearne et al., 1992; Bassil et al., 2003; Ellegren, 2004; Kafkas et al., 2009; Parida et al., 2009; Erdoğan et al., 2010; Gürcan et al., 2010b; Testolin & Cipriani, 2010; Boccacci et al., 2013; Mohammedzedeh et al., 2014; Zong et al., 2015; Öztürk et al., 2017a, b., Bhattarai & Mehlenbacher, 2018; Fiore et al., 2022; Karakaya et al., 2023).

Bassil et al. (2003) used 12 SSR primers to detect differences among 19 hazelnut genotypes. The primers were successful for identification of single alleles in in nine genotypes. It was suggested that they would be useful for establishment of clonal fingerprints, description of cultivars and genome mapping.

Gökırmak et al. (2005) used 10 pairs of SSR primers to investigate genetic diversity in 272 cultivars of European hazelnut. Preliminary PCR results indicated that the SSR loci used in the study were highly polymorphic.

Boccacci et al. (2006) studied on 78 hazelnut cultivars from various gene pools at 16 SSR loci in order to identify the genotypes and investigate their genetic relations. The northern European cultivars were found to evidently differentiate from the southern European ones and from the Turkish cultivars.

Boccacci et al. (2008) used 16 SSR markers to fingerprint 18 hazelnut cultivars from northeastern Spain. Their microsatellite profiles were characterized and used to study the genetic diversity in 33 genotypes including local Spanish germplasm. In this study no new case of synonymy was detected. High genetic diversity (0.7) was observed in 33 genotypes, although a high number of them displayed a close genetic relationship.

Gökırmak et al. (2009) used 21 SSR primers to investigate genetic diversity in 270 clonal accessions of European hazelnut (*C. avellana*). The resulting dendrogram in the study revealed four major geographical groups as: Central European, Black Sea, English and Spanish-Italian.

In the study of Boccacci and Botta (2010), microsatellite data were obtained at 16 SSR loci for 75 accessions from Spain, Italy, Türkiye, and Iran. An excess of heterozygosity was observed in Spanish and Turkish gene pools, while a heterozygosis deficit was observed in Iranian samples. The Italian accessions from Liguria region (North-West Italy) resulted genetically closer to varieties of Turkish origin than to Italian ones.

Later, Gürcan & Mehlenbacher (2010a) developed SSRs from the sequences of inter simple sequence repeat (ISSR) markers and flanking regions. Gürcan et al. (2010a) investigated genetic diversity in 88 accessions from Türkiye, Georgia, Azerbaijan and compared with cultivars from Spain and Italy using 12 microsatellite loci. A high level of genetic diversity ($H_e = 0.71$, $H_o = 0.70$) was observed in the Black Sea accessions. Six Turkish accessions in the US hazelnut collections were found to be synonyms of cultivars in the Turkish collection in Giresun.

Although about 525 SSR markers are publicly available in hazelnut, the majority contain di-nucleotide motifs which suffer from PCR artifacts that prevent automated scoring (Testolin & Cipriani, 2010).

In the study of Boccacci et al. (2013), phenotypic variation in nut and husk traits was decribed and genetic relationships were investigated using ten SSR markers, 57 well-known references cultivars, and 19 wild accessions in five countries (Portugal, Spain, Italy, Slovenia, and Greece). The results indicated the existence of three primary centers of diversity in the Mediterranean basin: northwestern Spain (Tarragona) and southern Italy (Campania) in the West and Black Sea (Türkiye) in the East. Moreover, the data suggested the existence of secondary gene pools in the Iberian (Asturias) and Italian (Liguria and Latium) Peninsulas, where local varieties were recently domesticated from wild forms and/or from introduced ancient domesticated varieties.

Leinemann et al. (2013) assessed the within and between-population differentiation of hazelnut, 20 natural populations from Germany, Italy and Hungary. Results for overall isozyme variability with Na 2.46 alleles per locus, allelic diversity (Ne) 1.39, expected heterozygosity He 21 % and 79 % polymorphic loci were in accordance with the findings of previous studies. The respective values for AFLPs were lower, but both marker systems revealed the

same level of about 3.5 % differentiation between populations. For cpSSR only the Italian sample showed within-population variation and the two haplotypes were completely differentiated from all other populations expressing a unique genetic structure with one single haplotype. Among the three marker systems AFLPs showed the best ability to differentiate between populations. While chloroplast markers were able to clearly distinguish only the Hungarian population, the nuclear markers revealed clear spatial genetic structures. The correlations between geographic and genetic distance was high for AFLPs. The correlograms illustrated this effect for all populations as well as for the German populations.

Sathuvalli & Mehlenbacher (2013) developed SSR markers from bacterial artificial chromosome (BAC) sequences. SSRs have also been developed from sequences of the hazelnut transcriptome (Colburn et al., 2017) and genome (Bhattarai & Mehlenbacher, 2017), and sequences in public databases (Boccacci et al., 2015; Gürcan & Mehlenbacher, 2010b).

Zong et al. (2015) applied 10 polymorphic SSR markers to evaluate the genetic diversity and population structure of 348 C. mandshurica individuals among populations in China. According to the coefficient of genetic differentiation (Fst = 0.1215), genetic variation within the populations (87.85%) were remarkably higher than among populations (12.15%). The results further showed that there was obvious genetic differentiation among populations from Northeast China to North China. There are also studies on determination of hazelnut genotypes and genetic relations based on primer screening.

Boccacci et al. (2005) developed 18 microsatellite loci in the European hazelnut using three enriched genomic libraries. They were evaluated on a set of 20 accessions of this species on the basis of number of alleles (mean: 7.1), expected heterozygosity (mean: 0.67), power of discrimination (mean: 0.77) and polymorphism information content (mean: 0.64). All primer pairs

amplified in all species, except for CaT-C505 in *Corylus ferox* and CaT-A114 in *Corylus californica*.

Helmstetter et al. (2020) used double digest restriction enzyme-associated DNA sequencing (ddRAD-seq) to investigate the genetic diversity and domestication of cultivated and wild hazelnuts in Türkiye. This included 200 individuals from cultivated and wild hazelnut trees collected in the Black Sea region in Türkiye, along with related Corylus species and specimens from the United Kingdom, Georgia, and Italy. Population genetic analyses revealed that cultivated hazelnuts showed elevated heterozygosity compared to wild individuals, and that genetic similarity did not correlate well with cultivar names. This might be due to somatic mutations, propagation of a group of clones that physiologically look alike but are actually genotypically different. Marker screening has previously been performed for Turkish hazelnut varieties, using microsatellite markers were to investigate their genetic diversity (Kafkas et al., 2009; Erdoğan et al., 2010; Gürcan et al., 2010b; Öztürk et al., 2017a, b).

SNP markers were used to provide a higher resolution for DNA fingerprinting of diverse varieties, and to understand the population structure of cultivated hazelnut trees in Türkiye (Oztolan-Erol et al., 2021). Representatives of nine commercial hazelnut varieties collected from multiple locations both from the Giresun Hazelnut Research Institute collection and private orchards were sequenced, and their SNP profiles analyzed using population genetics methods. In total 1,048,575 SNPs were discovered across all individuals, greatly increasing the number of known nucleotide polymorphisms in hazelnut. Bryant et al. (2010) developed a set of SNPs for European hazelnut by exploiting the exceptional depth and breadth of transcriptome sequencing made possible by high-throughput RNA sequencing (RNA-seq). 5,398 SNPs were identified in the study. Previously, Torello Marinoni et al. (2018) identified 9,999 SNPs using a Genotyping-by-

Sequencing approach, and generated saturated linkage maps for a segregating population of two parents, Tonda Gentile delle Langhe and Merveille de Bollwiller. The SNPs reported here are also potentially valuable for genetic and QTL mapping, although only a minority of loci (10,645) were consistently retrieved from all individuals. This indicates a high level of genetic diversity between individuals, and high levels of heterozygosity found in cultivated accessions also necessitate careful selection of potential molecular markers.

1.3. Molecular Studies on Resistance to Disease Causative Agents and Diseases in Hazelnut

Molecular marker techniques are frequently used to determine the resistance status and levels of plant species to cold, drought and certain diseases, and to explain resistance mechanisms.

Several defects have been reported in hazelnuts, such as the presence of blemishes, areas of discoloration, or stains in marked contrast with the rest of the kernel (Teviotdale et al., 2002). Hazelnut defects, defined as "rotten hazelnuts" by commercial evaluation, negatively impact kernel availability on the market as well as economics. The identification of the causal agents is critical in order to define and apply preventive actions, improve hazelnut yield and quality, and thus increase the market value.

In order to investigate the fungi associated with defective hazelnuts with a special focus on the role of *Diaporthe* spp.; to characterize *Diaporthe* strains based on molecular techniques, using multi-locus phylogenetic species identification by means of ITS, tub, and EF1- α ; and identify, at the sub-genus level; to analyze the genetic diversity of eastern filbert blight pathogen (*Anisogramma anomala*); to characterize Apple mosaic virus is aimed in hazelnut molecular studies.

Davis & Mehlenbacher (1997) tried to identify a marker gene resistant to Eastern filbert blight disease in hazelnut by using RAPD technique. For this purpose, 500 primers were screened in 100 F1 progeny developed from the cross 'Willamette' x 'VR 6-28'. Of these primers, OPH-171200, OPH-19600, UBC-152800 and UBC- 173500 were found to be linked to resistance gene.

Lunde et al. (2000) surveyed 90 hazelnut genotypes for response to the eastern filbert blight pathogen (*Anisogramma anomala*) by using primer UBC-152800 which was linked to the resistance gene. Primer UBC-152800 was absent in six genotypes: 'Closca Molla', 'Ratoli', 'Potomac', 'Yoder-5', 'Grand Traverse', and 'Medium Long', which did not display signs of the pathogen or symptoms of disease.

In the other study (Mehlenbacher et al., 2004), a total of 1,110 decamer primers were screened for RAPD markers linked to a dominant allele in hazelnut (*Corylus avellana*) that confers resistance to eastern filbert blight caused by *Anisogramma anomala*. Twenty RAPD markers linked in coupling, and five markers linked in repulsion, were found. Seven markers were sufficiently robust to allow their use in marker-assisted selection. These included AA12(850) which showed no recombination, and six markers on one side of the resistance locus: 173(500), 152(800), 122(825), 275(1130), H19(650) and O16(1250). The 16 markers closest to the resistance locus were cloned and sequenced.

Chen et al. (2005) identified five AFLP markers linked in coupling to resistance to Eastern filbert blight caused by *Anisogramma anomala*. B2-125 was located on one side of the resistance locus at a distance of 4.1 cM, while A4-265 (9.2 cM), C2-175 (5.9 cM) and D8-350 (2.5 cM) were on the other side, and A8-150 cosegregated with resistance. Sathuvalli & Mehlenbacher (2010) suggested that a dominant allele at a single locus from the obsolete pollenizer 'Gasaway' confers complete resistance to EFB (Eastern Filbert Blight) disease. New SCAR and SSCP markers were developed from 36 BAC end sequences and a fine map of the resistance region was constructed. The map spaned 3.7 cM and had an average interval of 0.3 cM between markers. Sathuvalli et al. (2011) studied segregation for disease response in two progenies from crosses

of Ratoli with susceptible selections and identified linked DNA markers. About half of the seedlings were resistant, suggesting control by a dominant allele at a single locus. A total of 900 RAPD primers and 64 AFLP primer combinations were screened. Four RAPD markers and two ALFP markers were identified and a linkage map was constructed. Based on co-segregation with SSR markers, Ratoli resistance was assigned to linkage group 7 while Gasaway resistance is on linkage group 6. Koma et al. (2021) discuss the genetic resistance to control the disease Eastern Filbert Blight caused by *Anisogramma anomala* (Peck) E. Müller. New simple sequence repeat (SSR) markers were developed for the resistance region on LG6, and new sources of resistance were investigated. In total, 42 new SSR markers were developed from four contigs in the genome sequence of cv 'Jefferson' released by the Oregon State University breeding program. These new LG6 resistance sources and SSR markers may be useful in breeding new cultivars.

Sathuvalli & Mehlenbacher (2010) revealed that the European hazelnut could serve as a model plant for the Betulaceae. A bacterial artificial chromosome (BAC) library for 'Jefferson' hazelnut was constructed using the cloning enzyme MboI and the vector pECBAC1 (BamHI site). The library consists of 39,936 clones arrayed in 104,384-well microtitre plates with a mean insert size of 117 kb. It provides a valuable resource for the map-based cloning of two important genes, the resistance gene from 'Gasaway' that confers resistance to eastern filbert blight caused by the fungus *Anisogramma anomala* (Peck) E. Müller and the S locus that controls pollen-stigma incompatibility. Fine mapping at the disease resistance locus showed that markers W07-375 and X01-825 flanked the resistance locus. The S locus is flanked by markers 204-950 and KG819-200.

Ertunç et al. (2011) studied molecular characterization of Apple mosaic virus (ApMV) in Turkish and Ukrainian Isolates and compared these isolates

to determine genome differences. The findings revealed 99% similarity between the isolates of two countries.

In the other study conducted by Vitale et al. (2011), isolate characterization of *Fusarium lateritium*, reported as the causal agent of nut gray necrosis (NGN) on hazelnut was undertaken in a study to investigate how morphological and molecular diversity was associated with host and geographic origin.

Peterschmidt (2013) focused on developing new microsatellite markers from hazelnut transcriptome sequences and on disease resistance from three accessions ('Culpla', 'Crvenje' and OSU 495.072) which showed no disease symptoms following a series of inoculations. They utilized the transcriptome sequence from 'Jefferson' hazelnut to mine for microsatellites, align with the genomic sequence, design primers, screen for polymorphism, and characterize and map polymorphic markers. A total of 1432 microsatellites were mined from the transcriptome sequence, and the most frequently found motifs were AG (35.8%), AT (13.3%), and AAG (12.7%), and 382 primer pairs were designed. Screening showed that 119 markers were polymorphic. Fifty-three markers that segregated in the mapping population or in three alternate populations were mapped and assigned to linkage groups. Diaporthe spp. fungi appears as necrotic spots on kernel surfaces and causes internal browning that is visible after cutting the nut in half (half-cut). In a previous study in the Caucasus region, *Diaporthe* spp. was identified as the crucial genus involved in causing hazelnut defects. Only three strains were identified at the species level in the study, but this suggested that *Diaporthe eres* was responsible for the visible brown spots on the kernel surface and the internal discoloration observable after the nut was cut in half (*Battilani et al. 2018*). *Diaporthe eres* was also recently reported by other authors as associated with hazelnut trunk cankers in Oregon (Wiman et al., 2019), while Diaporthe foeniculina was mentioned by Guerrero et al. (2019) as causing black tip and necrotic spots on hazelnut kernels in Chile

and Diaporthe rudis was detected in hazelnut kernels with visible mold in Oregon (Pscheidt et al., 2019). Since their discovery, Diaporthe spp. and their asexual stage *Phomopsis* spp. have been identified based on morphology and host association (Uecker, 1988; Ferreira et al., 2015). However, the association between host and species is not reliable within the *Diaporthe* genus as an identification criterion (Gomes et al., 2013; Udayanga et al., 2014a,b). It has been observed that the same *Diaporthe* spp. colonizes different hosts, and the co-occurrence of different species is commonly reported in the same host (Rehner & Uecker, 1994; Mostert et al., 2001; Guarnaccia et al., 2016; Guarnaccia & Crous, 2017). Additionally, many studies have recently claimed that morphology is generally not conclusive for identification at the species level due to the high complexity of the *Diaporthe* genus (Santos et al., 2010; Udayanga et al., 2011; Dissanayake et al., 2017a,b). Therefore, by using molecular approaches, substantial progress regarding the identification and characterization of emerging pathogens in the Diaporthe genus has been realized (Santos & Phillips, 2009; Diogo et al., 2010; Luongo et al., 2011; Udayanga et al., 2012a,b; Thomidis et al., 2013; Gao et al., 2017; Guarnaccia & Crous, 2017).

Arciuolo et al. (2020) investigated the fungi associated with defective hazelnuts in Türkiye, with a special focus on the role of *Diaporthe* spp., characterized Diaporthe strains based on molecular techniques, using multilocus phylogenetic species identification by means of ITS, tub, and EF1- α ; and dentified at the sub-genus level, the *Diaporthe* strains isolated from hazelnut kernels. Study presented that despite the heterogeneous nature of the hazelnut cultivable mycobiota, *Diaporthe* spp. are the only fungal species strongly associated with both internal and external defects in hazelnut kernels. A comparison of strains from different geographic origins showed that different species were responsible for similar symptoms (clade I versus clade III). Finally, a majority of Turkish *Diaporthe* strains are related to *Diaporthe eres*, a well-known plant pathogen that has been previously reported in *C. avellana*. Future studies focusing on improved molecular-based species classification, particularly of those strains belonging to clades I and III, will prove to be valuable to clarify their role as causative agents of hazelnut defects, assist with developing control strategies, and increase the quality and quantity of available product that meets market requirements.

1.4. Molecular Studies on Incompatibility Alleles in Hazelnut Genotypes

Hazelnuts are diploid (2n=2x=22), monoecious, dichogamous, windpollinated and self incompatible, and thus cross-pollinated in neture (Bhattarai & Mehlenbacher, 2018).. Cross-pollination is enforced by sporophytic incompatibility (Thompson, 1979), which is controlled by a single locus (the S-locus) with multiple alleles (haplotypes). The genetics of incompatibility in Corylus was first studied based on diallelic crosses, and its self-incompatibility was shown to be of the sporophytic type and controlled by a single multiallelic S-locus (Thompson, 1979). Reports based on other experimental methods also supported this conclusion (Hampson et al., 1996, Heslop-Harrison et al., 1986). Because of the importance of SSI in breeding programs and commercial pollination, many efforts have focused on testing the compatibility of hazelnut crosses using fluorescence microscopy, identifying S-alleles, and analyzing the dominance relationships among them (Mehlenbacher, 1997; Erdogan et al., 2005; Vicol et al., 2009). At present, 33 S-haplotypes are known in hazelnut (Mehlenbacher, 2014). SSI in hazelnut was first explored based on the hypothesis that the hazelnut genome contains homologs of Brassica S-locus genes. However, hybridization with an SLG probe was weak, indicating that the S-genes cloned from Brassica would not be useful for exploring SI in hazelnut (Hampson et al., 1996).

Pomper et al. (1998) identified RAPD markers for self-incompatibility alleles that will allow marker-assisted selection of desired S-alleles in hazelnut.

DNA was extracted from young leaves collected from field-planted parents and 26 progeny of the cross OSU 23.017 $(S_1S_{12})\times VR6-28$ (S_2S_{26}) $(OSU23\times VR6)$. A total of 250 primers were screened, and one RAPD marker each was identified for alleles S_2 (OPI07₇₅₀) and S_1 (OPJ14₁₇₀₀). Both the S_1 and S_2 markers were successfully cloned and 18 bp primers yielded the sole OPJ14₁₇₀₀ product, while 24-bp primers yielded OPI07₇₅₀ as well as an additional smaller product (700 bp) that was not polymorphic but was present in all of the S-genotypes examined. Bassil & Azerenko (2001) identified RAPD markers for self-incompatibility and cross pollen-grain incompatibility for in S1, S2, S3 alleles in hazelnut. The findings suggested that primers OPJ141700, OPI07750 and OPN201300 can be used for S₁, S₂, S₃ alleles.

Martins et al. (2012) identified the S-alleles present in a collection of Portuguese landraces. Ten major Portuguese hazelnut landraces were submitted to controlled pollinations in the field, with 18 genotypes whose S-alleles are known. Three landraces were revealed to have *S*2 allele, two have *S*5, and four have one of the *S*3, *S*5, *S*10, and *S*18 alleles.

Ives et al. (2014) reported that pollen-stigma incompatibility in European hazelnut (*Corylus avellana* L.) is of the sporophytic type and under the control of a single locus with multiple alleles (haplotypes). The S-locus was previously assigned to linkage group 5 (LG5) and linked DNA markers were identified. The loci that control leaf color and style color are linked to the S-locus. Segregation for leaf and style color and S-alleles were investigated in two progenies, the loci were mapped, and the two new maps with the LG5 reference map were compared using SSR markers. The color loci and the S-locus mapped to LG5 between SSR markers B028 and B774. The SSR markers closest to the S-locus are KG819, KG847, and BR259. In progeny 05050, which segregated for style and leaf color, no recombination was observed between the two traits. The style color locus was placed very close to SSR marker B028 in both progenies. On the reference map, RAPD DNA markers 564-500M, 345-

1050dF, and 204-950dF and ISSR marker 815-540dF were very close to the Slocus. It is suggested that the identification of closely linked markers will facilitate the map-based cloning of the S-locus and color loci in hazelnut.

To determine the mechanism of SI in Corylus, Li et al. (2020) carried out a comprehensive transcriptomic analysis for Ping'ou hybrid hazelnut (*Corylus heterophylla* × *Corylus avellana*). The identified sequences showed relatively high homology with all of the key genes involved in the SSI mechanism in Brassica. Li et al. (2020) cloned two full length cDNAs (ChaSRK1 and ChaSRK2) from Ping'ou hybrid hazelnut that were homologs of Brassica SRK using RACE techniques based on the unigenes selected from transcriptional data. It is examined the expression patterns of these genes using quantitative real-time PCR (qRT-PCR) and dual-color fluorescence in situ hybridization (FISH). Furthermore, it is cloned the homologs of ChaSRK1/2 from other cultivars and species using a pair of degenerate primers.

Hou et al. (2022), highlighted the molecular mechanism of saprophytic self-incompatibility (SSI) in some *Corylus* spp. that remains currently largely unknown. From self-pollination experiments ('Dawei' _ 'Dawei') and cross-pollination experiments ('Dawei' _ 'Liaozhen No. 7') and later an RNA-Seq analysis, the mechanism of pollen-stigma interactions was investigated to identify genes that may be responsible for SSI, discovering 19,163 up- and 13,314 downregulated genes, some of these potentially involved in pollen stigma interactions and SSI mechanisms.

1.5. Molecular Studies on Gene Expression in Hazelnut

Within the scope of recent studies carried out in hazelnut, researches on topics such as gene expression analyzes performed on seeds at different maturation stages, the molecular mechanism of embryo abortion, analysis of some proteins that cause food allergy and are in the composition of hazelnuts, identification of genes effective in the unsaturated fatty acid biosynthesis pathway through genome evolution analysis is being carried out. According to Rigola et al. (1998), unlike most of the angiosperms, which produce ovules during floral development such that they are ready for polen at anthesis, hazelnut ovary development is delayed and triggered by compatible pollination. In order to elucidate the mechanisms regulating this unusual process and the role of the MADS box genes in ovary development, a cDNA library from pollinated styles of hazelnut was screened with a mixture of MADS box genes from different plant species. CaMADS1 (*C. avellana* MADS box), a floral-specific MADS box gene, was isolated, and characterized as belonging to the sub-family of the AGAMOUS genes. A precise correlation was found between ovary development and CaMADS1 expression.

Santino et al. (2003) reported that plant lipoxygenases (LOXs) are a class of dioxygenases which display diverse functions in several physiological processes such as growth, development and response to biotic and abiotic stresses. Even though LOXs have been characterized from several plant species, the physiological role of seed LOXs is still unclear. With the aim to better clarify the occurrence of LOXs and their influence on hazelnut seed quality, researchers carried out the biochemical and molecular characterization of the main LOX isoforms expressed during seed development. Gene expression analysis conducted on seeds at different maturation stages showed that LOXs are mainly expressed at early developmental stages. CavPrx represented a first putative stigmaspecific peroxidase of hazelnut, whose expression profile changes during flower development, with a maximum level in mature stigmas pollinated with compatible pollen (Beltramo et al., 2012). The identification of this new putative stigmatic peroxidases in hazelnut is a further opportunity to analyze and understand the specific function of stigmaspecific class III peroxidases and their role in flower fertility. Food allergy is increasingly prevalent in our societies and represents today an emerging public health problem affecting children, adolescents, and adults. This pathological reaction of the immune system is triggered by the exposure to a particular food

protein in sensitized individuals and the clinical symptoms can range in severity from mild to life-threatening (Yu et al., 2016, Benede et al., 2016, Dhondalay et al., 2018).

The research of Wei et al. (2021) discusses the regulation of auxin in ovary development, which is thought to be related to auxin response factors (ARFs). It is suggested that ChARF3 (*C. heterophylla* ARF3) may regulate ovary initiation and ovule development by mediating genes related to auxin biosynthesis and transport, cell division and proliferation, and flower and fruit development.

Hazelnut (Corylus avellana L.) is one of the most commonly consumed nuts worldwide, either raw or roasted, being largely used by the food industry in a variety of processed foods including cakes, creams, chocolates, and confectionary products. Moreover, hazelnuts are generally regarded as "heartprotective" foods and are considered important in human nutrition and health due to their protein, fatty acid, vitamins, essential minerals, essential amino acids, phenolics and dietary fiber composition (Costa et al., 2016). Nevertheless, they are one of the most common triggers of IgE-mediated food allergies, being a major source of allergens capable of inducing mild to severe allergic reactions (Nitride et al., 2013). Sensitization to hazelnut allergens vary depending on the geographic origin and age of the patients. The objective of the study (Blanc et al., 2015) was to further investigate the allergenic activity of hazelnut allergens using sera from patients recruited in various European regions and presenting different sensitization patterns to hazelnut proteins. Hazelnut allergic patients from Mediterranean countries are mainly sensitized to the nsLTP Cor a 8 whereas patients from France and Switzerland are sensitized to pollen-related allergens. Interestingly, an intermediate profile was evidenced in patients from Madrid. Hazelnut 7S globulin (Cor a 11) and 11S globulin (Cor a 9) were found to be minor allergens, recognized only by patients from Mediterranean countries. The biologic activity of the 4 tested allergens,

analysed by HR assay, further confirmed the sensitization patterns, but also demonstrated the very high elicitation potency of Cor a 8.

In last few years, special attention has been given to food-induced allergies, in which hazelnut allergy is highlighted. Hazelnut is one of the most commonly consumed tree nuts, being largely used by the food industry in a variety of processed foods. Considering the great number of reports addressing hazelnut allergens, with an estimated increasing trend, Costa & Mafra (2016) intended to assemble all the relevant information available so far on the following main issues: prevalence of tree nut allergy, clinical threshold levels, molecular characterization of hazelnut allergens (Cor a 1, Cor a 2, Cor a 8, Cor a 9, Cor a 10, Cor a 11, Cor a 12, Cor a 14, and Cor a TLP) and their clinical relevance, and methodologies for detection of hazelnut allergens in foods. Due to hazelnut genetic diversity with the existence of many varieties, varietydependent differences in the IgE-binding properties may be suspected. Nevertheless, the results of the study of Ribeiro et al. (2020) do not support this hypothesis; although some minor differences were found at the level of genes encoding important allergens, namely Cor a 8, Cor a 9 and Cor a 14 for the 13 hazelnut varieties under study, these results were not reflected at the level of IgE-reactivity analysis using sera from allergic individuals. Cor a 1.04 and Cor a 9 were the predominant immunoreactive allergens within the tested sera, while no IgE-binding was observed for Cor a 8. From this study, none of the hazelnut varieties seems to be promising in terms of potential hypoallergenicity.

Liu et al. (2021) discussed the unclear mechanism underlying the adaptation of C. heterophylla to extremely low temperatures. Through genome evolution analysis, 17 expanded genes were identified, which were found to be significantly enriched in the unsaturated fatty acid biosynthesis pathway (ko01040). It was deduced that the expansion of these genes may promote a

high unsaturated fatty acid content in kernels and improve the adaptability of C. heterophylla to the cold climate of north-eastern China.

RESULT

It is seen that preliminary molecular studies on hazelnut primarily focused on genetic mapping, investigation of genetic relations, identification of genotypes from selected or diverse germplasms, determination of phylogenetic trees, morphological and phenological characterization. In recent years, molecular studies have also aimed at finding solutions for problems during cultivation process, identifying significant plant diseases and investigating disease resistance and tolerance conditions in cultivars/genotypes. There is also increasing number of studies on development of primers in hazelnut. The methods used in molecular studies show variation according to the study purpose. The use of molecular markers and genetic mapping in hazelnut caused many progress in the field such as population genetics, gene identification genomes, genetic relations, germplasm characterization, variety and type identification. Molecular markers contribute better planning of breeding programs by allowing to save time and money particularly in analyses of characters which may be time-consuming and expensive. Hence, continuation of studies on development of appropriate primers for molecular analysis will be useful.

REFERENCES

- Arciuolo, R., Santos, C., Soares, C., Castello, G., Spigolon, N., Chiusa, G., Lima, N. & Baattilani, P. (2020). Molecular Characterization of Diaporthe Species Associated With Hazelnut Defects. *Front. Plant Sci.* 11:611655. https://doi.org/10.3389/fpls.2020.611655
- Bassil, N.V., & Azarenko, A.N. (2001). RAPD markers for selfincompatibility in *Corylus avellana* L. *Acta Hortic*. 556, 537-543. 10.17660/ActaHortic.2001.556.78
- Bassil, N.V., Botta, R., & Mehlenbacher, S.A. (2003). Microsatellite markers of the European hazelnut. *Hortsci.*, *38*(*5*), 740-741.
- Bassil, N.V., R. Botta, & S.A. Mehlenbacher. (2005a). Microsatellite markers in hazelnut: Isolation, characterization, and cross-species amplification. Journal of the American Society for Horticultural Science 130:543-549. https://doi.org/10.21273/JASHS.130.4.543.
- Bassil, N.V., S.A. Mehlenbacher, & R. Botta. (2005b). Additional microsatellite markers of the european hazelnut. *Acta Hort*. 686:105– 110. https://doi.org/10.17660/ActaHortic.2005.686.13
- Beltramo, C., Marinoni, D. T., Perrona, I., & Botta, R. (2012). Isolation of a gene encoding for a class III peroxidase in female flower of Corylus avellana L. *Mol Biol Rep.*, 39:4997–5008. https://doi.org/10.1007/s11033-011-1296-y
- Beltramo, C., Boccacci, P., Prando, S., Botta, R., & Portis, E. (2014).
 Development of a genetic linkage map in hazelnut for the detection of QTLs. Proc. VIII th International Congress on Hazelnut, *Acta Hortic*. *1052*, 99-104. https://doi.org/ 10.17660/ActaHortic.2014.1052.12
- Beltramo, C., Valentini, N., Portis, E., Marinoni, D. T., Boccacci, P., Prando, M. A., & Botta, R. (2016). Genetic mapping and QTL analysis in European hazelnut (Corylus avellana L.). *Mol Breeding*, 36:27. https://doi.org/10.1007/s11032-016-0450-6

- Benedé, S., Blazquez, A. B., Chiang, D., Tordesillas, L., & Berin, M.C. (2016).
 The rise of food allergy: Environmental factors and emerging treatments. *EBioMedicine*, 7, 27-34. https://doi.org/10.1016/j.ebiom.2016.04.012
- Bhattarai, G. & Mehlenbacher, S. A. (2017). In silico development and characterization of tri-nucleotide simple sequence repeat markers in hazelnut (*Corylus avellana* L.). *PLOS One* 12:e0178061, https://doi.org/10.1371/journal.pone.0178061
- Bhattarai, G., & Mehlenbacher, S. A. (2018). Discovery, Characterization, and Linkage Mapping of Simple Sequence Repeat Markers In Hazelnut. J. Amer. Soc. Hort. Sci. 143(5):347–362. https://doi.org/10.21273/JASHS04461-18
- Blanc, F., Bernard, H., Ah-Leung, S., Przybylski-Nicaise, L., Skov, P. S., Purohit, A., de Blay, F., Ballmer-Weber, B., Fritsche, P., Rivas, M. F., Reig, I., Sinaniotis, A., Vassilopoulou, E., Hoffmann-Sommergruber, K., Vieths, S., Rigby, N., Mills, C., & Adel-Patient, K. (2015). Further studies on the biological activity of hazelnut allergens. *Clinical and Translational Allergy*, 5:26. https://doi.org/10.1186/s13601-015-0066-7
- Boccacci, P., Akkak, A., Bassil, N.V., Mehlenbacher, S.A., & Botta, R. (2005).
 Characterization and evaluation of microsatellite loci in European hazelnut and their transferability to other *Corylus* species. *Molecular Ecology Notes*, 5, 934-937 https://doi.org/10.1111/j.1471-8286.2005.01121.x
- Boccacci, P., Akkak, A., & Botta, R. (2006). DNA typing and genetic relations among European hazelnut cultivars using microsatellite markers. *Genome*, 49, 598-611. https://doi.org/10.1139/g06-017
- Boccacci, P., Botta, R., & Rovira, M. (2008). Genetic diversity of hazelnut germplasm in northeastern Spain. *Hortsci.* 43(3), 667-672. https://doi.org/10.21273/HORTSCI.43.3.667

- Boccacci, P., & Botta, R. (2010). Microsatellite variability and genetic structure in hazelnut cultivars from different growing regions. *Sci Hort. 124*, 128-133. https://doi.org/10.1016/j.scienta.2009.12.015
- Boccacci, P., Aramini, M., Valentini, N., Bacchetta, L., Rovira, M., Drogoudi,
 P., Silva, A.P., Solar, A., Calizzano, F., Erdoğan, V., et al. (2013).
 Molecular and morphological diversity of *on-farm*hazelnut (*Corylus avellana* L.) landraces from southern Europe and their role in the origin and diffusion of cultivated germplasm. *Tree Genetics and Genomes*. 9(6), 1465-1480. https://doi.org/10.1007/s11295-013-0651-7
- Boccacci, P., C. Beltramo, M. A., Sandoval Prando, A., Lembo, C., Sartor, S.A., Mehlenbacher, R. Botta, & D. Torello Marinoni. (2015). In silico mining, characterization and cross-species transferability of EST-SSR markers for European hazelnut (*Corylus avellana* L.). *Mol. Breed.* 35:21. https://doi.org/10.1007/s11032-015-0195-7
- Bryant, D.W., Fox, S.E., Rowley, E.R., Priest, H.D., Shen, R., Wang, W.K., & Mockler, T.C. (2010). Discovery of SNP Markers in Expressed Genes of Hazelnut. Proc. IS on Molecular Markers in Horticulture Acta Hortic. 859, 289-294. https://doi.org/10.17660/ActaHortic.2010.859.33
- Chen, H., Mehlenbacher, S.A., & Smith, D.C. (2005). AFLP markers linked to eastern filbert blight resistance from OSU 408.040 Hazelnut. *Journal of the American Society for Horticultural Science*, 130(3), 412-417. https://doi.org/10.21273/JASHS.130.3.412
- Costa, J., & Mafra, I. (2016). Hazelnut Allergens: Molecular Characterization, Detection, and Clinical Relevance. Critical Reviews in Food Science and Nutrition, 56 (15): 2579-2605. https://doi.org/10.1080/10408398.2013.826173
- Costa, J., Mafra, I., Carrapatoso, I., & Oliveira, M.B.P.P. (2016). Hazelnut allergens: Molecular characterisation, detection and clinical relevance.

Crit. Rev. Food Sci. Nutr., 56, 2579-2605. https://doi.org/10.1080/10408398.2013.826173

- Colburn, B.C., S.A. Mehlenbacher, & V.R. Sathuvalli. (2017). Development and mapping of microsatellite markers from transcriptome sequences of European hazelnut (Corylus avellana L.) and use for germplasm characterization. *Mol. Breed.* 37:16. https://doi.org/10.1007/s11032-016-0616-2
- Davis, J.W., & Mehlenbacher, S.A. (1997). Identification of RAPD markers linked to eastern filbert blight resistance in hazelnut. Fourth Int. Symposium on Hazelnut, Acta Hortic. 445, 553-556. https://doi.org/10.17660/ActaHortic.1997.445.71
- Demir, T. (2014). Molecular characterization of Turkish hazelnut cultivars and accessions. J. Anim. *Plant Sci.*, 24(3), 820-828. https://doi.org/ https://hdl.handle.net/20.500.12619/67378.
- Dhondalay, G.K., Rael, E., Acharya, S., Zhang, W., Sampath, V., Galli, S.J., Tibshirani, R., Boyd, S.D., Maecker, H., Nadeau, K.C., et al. (2018). Food allergy and omics. J. Allergy Clin. *Immunol.*, 141, 20-29. https://doi.org/10.1016/j.jaci.2017.11.007.
- Diogo, E. L. F., Santos, J. M., & Phillips, A. J. L. (2010). Phylogeny, morphology and pathogenicity of Diaporthe and Phomopsis species on almond in Portugal. *Fungal Divers.*, 44, 107–115. https://doi.org/10.1007/s13225-010-0057-x
- Dissanayake, A., Camporesi, E., Hyde, K. D., Zhang, W., Yan, J. Y., & Li, X.
 H. (2017a). Molecular phylogenetic analysis reveals seven new
 Diaporthe species from Italy. *Mycosphere*, 8, 853–877. https://doi.org/10.5943/mycosphere/8/5/4
- Dissanayake, A. J., Phillips, A. J. L., Hyde, K. D., Yan, J. Y., & Li, X. H. (2017b). The current status of species in Diaporthe. Mycosphere, 8, 1106–1156. https://doi.org/10.5943/mycosphere/8/5/5

- Ellegren, H. (2004). Microsatellites: Simple sequences with complex evolution. Nat. Rev. Genet. 5:435-445. https://doi.org/10.1038/nrg1348
- Erdoğan, V. (1999). Genetic relationships among hazelnut (Corylus) Species.
 Phd Thesis, Oregon State University, USA. https://doi.org/10.1038/nrg1348
- Erdoğan, V., Mehlenbacher, S. A., Köksal, A. İ., & Kurt, H. (2005). Incompatibility Alleles Expressed in Pollen of Turkish Hazelnut Cultivars. *Turkish Journal of Biology*, 29(2):111-116.
- Erdoğan, V., Köksal, A., & Aygün, A. (2010). Assessment of genetic relationships among Turkish Hazelnut (*Corylus avellana* L.) Cultivars by RAPD markers. Roman. *Biotechnol. Lett.*, 15, 5591-5601.
- Ertunç, F., Canik, D., Gospodaryk, A., Budzanivska, I.G., & Polischuk, V.P. (2011). Molecular characterization of *Apple mosaic virus* in Turkish and Ukrainian isolates. *J. Agric. Sci.*, *17*, 95-104. https://doi.org/10.1501/tarimbil_0000001161
- Fao, 2022. https://www.fao.org/faostat/en/#data/QCL. Erişim Tarihi: 25.01.2024
- Ferreira, J. J., Garcia-Gonzalez, C., Tous, J., & Rovira, M. (2010). Genetic diversity revealed by morphological traits and ISSR markers in hazelnut germplasm from northern Spain. *Plant Breeding*, 129, 435-441. https://doi.org/10.1111/j.1439-0523.2009.01702.x
- Ferreira, M. C., Vieira, M. de L. A., Zani, C. L., Alves, T. M. de A., Sales, P., Murta, S. M. F., et al. (2015). Molecular phylogeny, diversity, symbiosis and discover of bioactive compounds of endophytic fungi associated with the medicinal Amazonian plant Carapa guianensis Aublet (Meliaceae). *Biochem. Syst. Ecol.*, 59, 36-44. https://doi.org/10.1016/j.bse.2014.12.017
- Fiore, M. C., Marchese, A., Mauceri, A., Digangi, I., & Scialabba, A. (2022). Diversity Assessment and DNA-Based Fingerprinting of Sicilian

Hazelnut (*Corylus avellana* L.) Germplasm. *Plants*, 11(5): 631. https://doi.org/10.3390/plants11050631

- Gantner, M., & Okon, S. (2009). Assessment of genetic relationships between hazelnut cultivars using random amplified polymorphic DNA (RAPD) markers. Acta Horticulturae, 845, 579-584. https://doi.org/10.17660/ActaHortic.2009.845.91
- Galderisi, U., Cipollara, M., Bernardo, G., Masi, L., Galano, G., & Cascino,
 A. (1999). Identification of hazelnut cultivars by RAPD analysis. *Plant Cell Reports*, 18,652-655. https://doi.org/10.1007/s002990050637
- Gao, Y., Liu, F., Duan, W., Crous, P. W., & Cai, L. (2017). Diaporthe is paraphyletic. *IMA Fungus*, 8, 153-187. https://doi.org/10.5598/imafungus.2017.08.01.11
- Gomes, R. R., Glienke, C., Videira, S. I. R., Lombard, L., Groenewald, J. Z., & Crous, P. W. (2013). Diaporthe: a genus of endophytic, saprobic and plant pathogenic fungi. *Persoonia*, 31, 1-41. https://doi.org/10.3767/003158513X666844
- Gökırmak, T., Mehlenbacher, S.A., & Bassil, N.V. (2005). Investigation of genetic diversity among European hazelnut cultivars using SSR markers. Proc. VI th Intl. Congress on Hazelnut. Acta Hortic, 686, 141-148. https://doi.org/10.17660/ActaHortic.2005.686.18
- Gökırmak, T., Mehlenbacher, S.A., & Bassil, N.V. (2009). Characterization of European hazelnut (*Corylus avellena*) cultivars using SSR markers. *Gent Resour Crop Evol.*, 56:147-172. https://doi.org/10.1007/s10722-008-9352-8
- Guarnaccia, V., & Crous, P. W. (2017). Emerging citrus diseases in Europe caused by species of Diaporthe. *IMA Fungus*, 8, 317-334. https://doi.org/10.5598/imafungus.2017.08.02.07

- Guarnaccia, V., Vitale, A., Cirvilleri, G., Aiello, D., Susca, A., Epifani, F., et al. (2016). Characterisation and pathogenicity of fungal species associated with branch cankers and stem-end rot of avocado in Italy. *Eur. J. Plant Pathol.*, 146, 963-976. https://doi.org/10.1007/s10658-016-0973-z
- Guerrero, J., Galdames, R. E., & Perez, S. (2019). First report of Diaporthe foeniculina causing black tip and necrotic spot on hazelnut kernel in Chile. *Plant Dis.*, 104:975. https://doi.org/10.1094/PDIS-06-19-1166-PDN
- Gürcan, K. & S.A. Mehlenbacher. (2010a). Development of microsatellite marker loci for european hazelnut (Corylus avellana L.) from ISSR fragments. Mol. Breed., 26:551-559. https://doi.org/10.1007/s11032-010-9464-7
- Gürcan, K. & S.A. Mehlenbacher. (2010b). Transferability of microsatellite markers in the Betulaceae. J. Amer. Soc. Hort. Sci. 135:159-173. https://doi.org/10.21273/JASHS.135.2.159
- Gürcan, K., S.A. Mehlenbacher, R. Botta, & P. Boccacci. (2010a). Development, characterization, segregation, and mapping of microsatellite markers for European hazelnut (Corylus avellana L.) from enriched genomic libraries and usefulness in genetic diversity studies. *Tree Genet. Genomes*, 6:513-531. https://doi.org/10.1007/s11295-010-0269-y
- Gürcan, K., Mehlenbacher, S. A., & Erdoğan, V. (2010b). Genetic diversity in hazelnut (*Corylus avellana* L.) cultivars from Black Sea countries assessed using SSR markers. *Plant Breed.*, 129, 422-434 https://doi.org/10.1111/j.1439-0523.2009.01753.x
- Hampson, C. R., Coleman, G. D., & Azarenko, A. N. (1996). Does the genome of Corylus avellana L. contain sequences homologous to the self-

incompatibility gene of Brassica? *Theor Appl Genet.*, 93(5-6):759-64. https://doi.org/10.1007/BF00224073

- Hearne, C.M., S. Ghosh, & J.A. Todd. (1992). Microsatellites for linkage analysis of genetic traits. *Trends Genet.*, 8:288-294. https://doi.org/10.1016/0168-9525(92)90256-4
- Helmstetter, A. J., Oztolan–Erol, N., Lucas, S. J., & Buggs, R. J. A. (2020).
 Genetic diversity and domestication of hazelnut (*Corylus avellana* L.) in Turkey. *Plants People Planet*, 2, 326-339. https://doi.org/10.1002/ppp3.10078
- Heslop-Harrison, Y., Heslop-Harrison, J. S., & Heslop-Harrison, J. (1986). Germination Of Corylus Avellana L. (Hazel) Pollen: Hydration And The Function Of The Oncus. *Acta Botanica Neerlandica*, 35(3):265-284. https://doi.org/10.1111/j.1438-8677.1986.tb01289.x
- Hou, S., Zhao, T., Yang, Z., Liang, L., Ma, W., Wang, G., & Ma, Q. (2022).
 Stigmatic Transcriptome Analysis of Self-Incompatible and Compatible
 Pollination in *Corylus heterophylla* Fisch. × *Corylus avellana* L. *Front. Plant Sci.*, 13:800768. https://doi.org/10.3389/fpls.2022.800768
- Ives, C., Sathuvalli, V.R., Colburn, B.C., & Mehlenbacher, S.A. (2014). Mapping the incompatibility and style color loci in two hazelnut progenies. *HortSci.*, 49(3), 250-253. https://doi.org/10.21273/HORTSCI.49.3.250
- Kafkas, S. & Doğan, Y. (2009). Genetic characterization of hazelnut cultivars from Turkey using molecular markers. Hortsci. 44(6), 1557-1561. https://doi.org/10.21273/HORTSCI.44.6.1557
- Karakaya, O., Yaman, M., Balta, F., Yılmaz, M., & Balta, M. F. (2023). Assessment of genetic diversity revealed by morphological traits and ISSR markers in hazelnut germplasm (Corylus *avellana* L.) from Eastern Black Sea Region, Turkey. *Genetic Resources and Crop Evolution*, 70: 525-537. https://doi.org/10.1007/s10722-022-01444-0

- Koma, G. K., Şekerli, M., Snelling, J. W., & Mehlenbacher, S. A. (2021). New Sources of Eastern Filbert Blight Resistance and Simple Sequence Repeat Markers on Linkage Group 6 in Hazelnut (Corylus avellana L.). *Front. Plant Sci.*, 12:68412. https://doi.org/10.3389/fpls.2021.684122
- Leinemann, L., Wilfried, S., Bernhard, H., Oleksandra, K., Wolfgang, A., Barbara, F., Bolko, H., Ralf, K., Martin, R., & Reiner, F. (2013). Genetic variation of chloroplast and nuclear markers in natural populations of hazelnut (*Corylus avellana* L.) in Germany. *Plant Systematics & Evolution.*, 299(2), 369-378. https://doi.org/10.1007/s00606-012-0727-0
- Li, Q., Zhao, T., Liang, L., Hou, S., Wang, G., & Ma, Q. (2020). Molecular cloning and expression analysis of hybrid hazelnut (*Corylus heterophylla* × *Corylus avellana*) ChaSRK1/2 genes and their homologs from other cultivars and species. Gene, 756:144917. https://doi.org/10.1016/j.gene.2020.144917
- Liu, J., Wei, H., Zhang, X., He, H., Cheng, Y., & Wang, D. (2021). Chromosome-Level Genome Assembly and HazelOmics Database Construction Provides Insights Into Unsaturated Fatty Acid Synthesis and Cold Resistance in Hazelnut (*Corylusheterophylla*). *Front. Plant Sci.*, 12:766548. https://doi.org/10.3389/fpls.2021.766548
- Lunde, C.F., Mechlenbacher, S.A. & Smith, D.C. (2000). Survey of hazelnut cultivars for response to Eastern filbert blight inoculation. *HortSci.*, 35 (4), 729-731. https://doi.org/10.21273/HORTSCI.35.4.729
- Luongo, L., Santori, A., Riccioni, L., & Belisario, A. (2011). Phomopsis sp. associated with post-harvest fruit rot of kiwifruit in Italy. *J. Plant Pathol.* 93, 205-210. https://doi.org/10.4454/jpp.v93i1.293
- Martins, S., Siva, A.P., Santos, A.A., & Carnide, V. (2009). Diversity in hazelnut using RAPD and ISSR markers. *Acta Hortic.*, 845, 145-150 https://doi.org/10.17660/ActaHortic.2009.845.17

- Martins, S., Rovira, M., Silva, A.P., & Carnida, V. (2012). Incompatibility alleles in Portuguese hazelnut landraces. International Scholary Research Network, ISRN Agronomy, 1-6 pp. https://doi.org/10.5402/2012/154723
- Mehlenbacher, S. A. (1991). Hazelnuts (Corylus). Acta Horticulturae, 290: Genetic Resources of Temperate Fruit and Nut Crops. https://doi.org/10.17660/ActaHortic.1991.290.18
- Mehlenbacher, S. A. (1997). Revised dominance hierarchy for S-alleles in Corylus avellana L. *Theor. Appl. Genet.*, 94, 360-366. https://doi.org/10.1007/s001220050424
- Mehlenbacher, S. A. (2014). Incompatibility alleles of hazelnut cultivars. Acta Horticulturae 1052(1052):107-116. https://doi.org/10.17660/ActaHortic.2014.1052.13
- Mehlenbacher, S.A., Brown, R.N., Davis, J.W., Chen, H., Bassil, N.V., Smith, D.C., & Kubisiak, T.L. (2004). RAPD markers linked to eastern filbert blight resistance in Corylus avellana. *Theor Appl Genet.*, 108 (4), 651-6 https://doi.org/10.1007/s00122-003-1476-9
- Mehlenbacher, S.A., Brown, R.N., Nouhra, E.R., Gökırmak, T., Bassil, N.V.,
 & Kubisiak, T.L. (2006). A genetic linkage map for hazelnut based on
 RAPD and SSR markers. *Genome*, 49, 122-133. https://doi.org/10.1139/g05-091
- Mehlenbacher, S. A. & D.C. Smith. (2009). 'Red Dragon' ornamental hazelnut. *HortScience*, 44:843-844. https://doi.org/10.21273/HORTSCI.44.3.843
- Miaja, M.L., Vallania, R., Me, C., Akkak, A. Nassi, O., & Lepori, G. (2001). Varietal characterization in hazelnut by RAPD markers. Proc. V Int. Congress on Hazelnut Ed: S. A. Mehlenbacher Acta Hortic., 556, 247-250. https://doi.org/10.17660/ActaHortic.2001.556.35
- Mohammadzedeh, M., Fattahi, R., Zamani, Z., & Khadivi-Khub, K. (2014). Genetic identity and relationships of hazelnut (*Corylus avellana* L.) landraces as revealed by morphological characteristics and molecular

markers. *Sci Hort.*, *167*, 17-26 https://doi.org/10.1016/j.scienta.2013.12.025

- Mostert, L., Crous, P. W., Kang, J. C., & Phillips, A. J. L. (2001). Species of Phomopsis and a Libertella sp. occurring on grapevines with specific reference to South Africa: morphological, cultural, molecular and pathological characterization. *Mycologia*, 93, 146-167. https://doi.org/10.2307/3761612
- Nas, M.N., Mutlu, N., & Read, P.E. (2004). Random amplified DNA (RAPD) analysis of long-term cultured hybrid hazelnut. *Hortsci.*, 39, 1079-1082. https://doi.org/10.21273/HORTSCI.39.5.1079
- Nitride, C., Mamone, G., Picariello, G., Mills, C., Nocerino, R., Canani, R.B., & Ferranti, P. (2013). Proteomic and immunological characterization of a new food allergen from hazelnut (Corylus avellana). *J. Proteom*, 86, 16-26. https://doi.org/10.1016/j.jprot.2013.05.001
- Oztolan-Erol, N., Helmstetter, A. J., İnan, A., Buggs, R. J. A., & Lucas, S. J. (2021). Unraveling Genetic Diversity Amongst European Hazelnut (*Corylus avellana* L.) Varieties in Turkey. *Front. Plant Sci.*, 12:661274. https://doi.org/10.3389/fpls.2021.661274
- Öztürk, S. C., Öztürk, S., Çelik, I., Stampar, F., Veberic, R., Doğanlar, S., et al. (2017a). Molecular genetic diversity and association mapping of nut and kernel traits in Slovenian hazelnut (*Corylus avellana*) germplasm. *Tree Genet. Genomes* 13:16. https://doi.org/10.1007/s11295-016-1098-4
- Öztürk, S. C., Balık, H. Ý, Balık, S. K., Kızılcı, G., Duyar, Ö, Doğanlar, S., et al. (2017b). Molecular genetic diversity of the Turkish national hazelnut collection and selection of a core set. *Tree Genet. Genomes*, 13:113. https://doi.org/10.1007/s11295-017-1195-z
- Parida, S.K., S.K. Kalia, S. Kaul, V. Dalal, G. Hemaprabha, A. Selvi, A. Pandit,
 A. Singh, K. Gaikwad, T.R. Sharma, P. Shankar, S. Nagendra, K. Singh,
 & T. Mohapatra. (2009). Informative genomic microsatellite markers for

efficient genotyping applications in sugarcane. *Theor. Appl. Genet.*, 118:327-338. https://doi.org/10.1007/s00122-008-0902-4

- Petersmith, B.C. (2013). DNA markers and characterization of novel sources of eastern filbert blight resistance in European hazelnut (*Corylus avellana* L.). Oregon State University, Master of Science in Horticulture, MSc Thesis.
- Pomper, K.W., Azarenko, A.N., Bassil, N., Davis, J.W., & Mehlenbacher, S.A. (1998). Identification of random amplified polymorphic DNA markers for self-incompatibility alleles in *Corylus avellana* L. *Theor. Appl. Genet.*, 97, 479-487.https://doi.org/10.1007/s001220050920
- Pscheidt, J. W., Heckert, S., Wiseman, M., & Jones, L. (2019). Fungi associated with and influence of moisture on development of kernel mold of hazelnut. *Plant Dis.* 103, 922-928. https://doi.org/10.1094/PDIS-09-18-1520-RE
- Rehner, S. A., & Uecker, F. A. (1994). Nuclear ribosomal internal transcribed spacer phylogeny and host diversity in the coelomycete Phomopsis. *Can. J. Bot.*, 72, 1666-1674. https://doi.org/10.1139/b94-204
- Ribeiro, M., Costa, J., Mafra, I., Cabo, S., Silva, A. P., Gonçalves, B., Hillion,
 M., Hebraud, M., & Igrejas, G. (2020). Natural Variation of Hazelnut
 Allergenicity: Is There Any Potential for Selecting Hypoallergenic
 Varieties? *Nutrients*, 12: 2100, 1-20.
 https://doi.org/10.3390/nu12072100
- Rigola, D., Pe, M.E., Fabrizio, C., Me, G., & Sari-Gorla, M. (1998). *CaMADS1*, a MADS box gene expressed in the carpel of hazelnut. *Plant Mol. Biol.* 38, 1147-1160. https://doi.org/10.1023/A:1006022524708
- Santino, A., Paolis, A.D., Gallo, A., Quarta, A., Casey, R., & Mita, G. (2003). Biochemical and molecular characterization of hazelnut (*Corylus avellana*) seed lipoxygenases. *Eur. J. Biochem.*, 270, 4365-4375. 10.1046/j.1432-1033.2003.03831.x

- Santos, J. M., Correia, V. G., & Phillips, A. J. L. (2010). Primers for matingtype diagnosis in Diaporthe and Phomopsis: their use in teleomorph induction in vitro and biological species definition. *Fungal Biol.* 114,255-270. https://doi.org/10.1016/j.funbio.2010.01.007
- Santos, J. M., & Phillips, A. (2009). Resolving the complex of Diaporthe (Phomopsis) species occurring on Foeniculum vulgare in Portugal.
 Fungal Divers. 34, 111-125. https://doi.org/10.3767/003158511X603719
- Sathuvalli, V.R., & Mehlenbacher, S.A. (2010). Fine Mapping of Eastern Filbert Blight Resistance in Hazelnut with SCAR and SSCP Markers Developed from BAC End Sequences. Proc. IS on Molecular Markers in Horticulture, Acta Hortic., 859, 395-400 https://dx.doi.org/10.17660/actahortic.2010.859.48
- Sathuvalli, V.R., Chen, H., Mehlenbacher, S.A., & Smith, D.C. (2011). DNA markers linked to eastern filbert blight resistance in Ratoli hazelnut. *Tree Genetics and genomes.*, 7, 337-345. https://dx.doi.org/10.1007/s11295-010-0335-5
- Sathuvalli, V.R. & Mehlenbacher, S.A. (2013). De novo sequencing of hazelnut bacterial artificial chromosomes (BACs) using multiplex Illumina sequencing and targeted marker development for eastern filbert blight resistance. Tree Genet. Genomes, 9:1109-1118. https://doi.org/10.1007/s11295-013-0626
- Testolin, R. & Cipriani, G. (2010). Molecular markers for germplasm identification and characterization. Acta Hort. 859:59-72. https://doi.org/10.17660/ActaHortic.2010.859.5
- Teviotdale, B. L., Themis, J. M. & Pscheidt, J. W. (2002). Compendium of Nut Crop Diseases in Temperate Zones. St. Paul, MN: APS Press.

- Thomidis, T., Exadaktylou, E., & Chen, S. F. (2013). Diaporthe neotheicola, anew threat for kiwifruit in Greece. *Crop Prot.*, 47, 35-40. https://doi.org/10.1016/j.cropro.2012.12.024
- Thompson, M. M. (1979). Genetics of incompatibility in Corylus avellana L. *Theor. Appl. Genet.* 54:113-116. https://doi.org/10.1007/BF01159464
- Torello Marinoni, D., Valentini, N., Portis, E., Acquadro, A., Beltramo, C., Mehlenbacher, S. A., et al. (2018). High density SNP mapping and QTL analysis for time of leaf budburst in *Corylus avellana* L. *PLoS One*, 13:e0195408. https://doi.org/10.1371/journal.pone.0195408
- Udayanga, D., Liu, X., McKenzie, E. H. C., Chukeatirote, E., Bahkali, A. H. A., & Hyde, K. D. (2011). The genus Phomopsis: biology, applications, species concepts and names of common phytopathogens. *Fungal Divers.*, 50, 189-225. https://doi.org/10.1007/s13225-011-0126-9
- Udayanga, D., Liu, X., Crous, P. W., McKenzie, E. H. C., Chukeatirote, E., & Hyde, K. D. (2012a). A multi-locus phylogenetic evaluation of Diaporthe (Phomopsis). *Fungal Divers.*, 56, 157-171. https://doi.org/10.1007/s13225-012-0190-9
- Udayanga, D., Liu, X., Crous, P. W., McKenzie, E. H. C., Chukeatirote, E., & Hyde, K. D. (2012b). Multi-locus phylogeny reveals three new species of Diaporthe from Thailand. *Cryptogam. Mycol.*, 33, 295-309. https://doi.org/10.7872/crym.v33.iss3.2012.295
- Udayanga, D., Castlebury, L. A., Rossman, A. Y., Chukeatirote, E., & Hyde, K. D. (2014a). Insights into the genus Diaporthe: phylogenetic species delimitation in the D. eresspecies complex. *Fungal Divers.*, 67, 203-229.https://doi.org/10.1007/s13225-014-0297-2
- Udayanga, D., Castlebury, L. A., Rossman, A. Y., & Hyde, K. D. (2014b). Species limits in Diaporthe: molecular re-assessment of D. citri, D. cytosporella, D. foeniculina and D. rudis. *Persoonia*, 32, 83-101. https://doi.org/10.3767/003158514X679984

- Uecker, F. A. (1988). A World List of Phomopsis Names with Notes on Nomenclature, Morphology and Biology. Contributions from the U.S. National Fungus Collection. Mycologia Memoir, 13th Edn. Berlin:Cramer Publisher.
- Valentini, N., Marinomi, D., Me,G., & Botta, R. (2001). Evaluation of 'Tonda Gentile Delle Langhe' Clones. Proc. V Int. Congress on Hazelnut. Acta Hortic., 556, 209-218 https://dx.doi.org/10.17660/actahortic.2001.556.30
- Valentini, N., Portis, E., Bolta, R., Acquadro, A., Pavese, V., Cavalet, E., & Torello Marinoni, G. D. (2021). Mapping the Genetic Regions Responsible for Key Phenology-Related Traits in the European Hazelnut. *Front. Plant Sci.*, 12:749394. https://doi.org/10.3389/fpls.2021.749394
- Vicol A., Botu I., Botu M., & Giorgota A. (2009). Preliminary Study Of Incompatibility Alleles Expressed In Pollen Of Romanian Hazelnut Cultivars. *Bulletin Uasvm Horticulture*, 66(1): 480-483. https://www.jstor.org/stable/26525282
- Vitale, S., Santori, A., Wajnberg, E., Castagnone-Sereno, P., Luango, L., & Belisario, A. (2011). Morphological and Molecular Analysis of Fusarium lateritium the cause of GrayNecrosis of Hazelnut Fruit in Italy. *Phytopathology.*, 101(6), 679-686. https://doi.org/10.1094/PHYTO-04-10-0120
- Wei, H., Cheng, Y., Sun, Y., Zhang, X., He, H., & Liu, J. (2021). Genome-Wide Identification of the ARF Gene Family and ARF3 Target Genes Regulating Ovary Initiation in Hazel via ChIP Sequencing. *Front. Plant Sci.* 12:715820. https://doi.org/10.3389/fpls.2021.715820
- Whitcher, I.N., & J. Wen. (2001). Phylogeny and biogeography of Corylus (Betulaceae): inferences from ITS Sequences. *Syst. Bot.*, 26 (2): 283-298. https://doi.org/10.1043/0363-6445-26.2.283

- Wiman, N. G., Webber, J. B., Wiseman, M., & Merlet, L. (2019). Identity and pathogenicity of some fungi associated with hazelnut (*Corylus avellana* L.) trunk cankers in Oregon. *PLoS One*, 14:e0223500. https://doi.org/10.1371/journal.pone.0223500
- Yu, W., Freeland, D.M.H., & Nadeau, K.C. (2016). Food allergy: Immune mechanisms, diagnosis and immunotherapy. *Nat. Rev. Immunol*, 16:751-765. https://doi.org/10.1038/nri.2016.111
- Yılmaz, M. (2009). Pomological, morphological and molecular characterisation of some hazelnut varieties and genotypes University of Cukurova, Department of Horticulture, Institute of Basic and Applied Sciences, Ph. D. Thesis.
- Zong, J. W., Zhao, T. T., Ma, Q. H., Liang, L. S., & Wang, G. X. (2015). Assessment of Genetic Diversity and Population Genetic Structure of *Corylus mandshurica* in China Using SSR Markers. *PLoS One.* 10(9),1-12. https://doi.org/10.1371/journal.pone.0137528

CHAPTER 3

SYNTHETIC BIOLOGY AND MICROBIOME ENGINEERING IN AGRICULTURE

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1. Introduction

The amount of carbon in the soil and the amount and variability of soil microorganisms are key indicators of soil health and fertility (Lal, 2014). It is known that the number of microorganisms is low in soils lacking plant roots. Industrialised agricultural techniques often focus only on productivity and not on soil health. Farm practices that leave the soil bare and exposed and chemical fertilisers applied to increase yields lead to loss of soil carbon and soil microbial diversity. Currently, one-third of the world's land surface is degraded to varying degrees and 24 billion tonnes of fertile soil is lost each year, posing a significant threat to ecosystems and agricultural processes (Coban et al., 2022). Furthermore, the United Nations estimates that the world population will be 9.8 billion people by 2050 (https:// population.un.org/wpp/). To meet the increasing demand for food, feed, fibre and bioenergy, agricultural productivity needs to increase by an estimated 70% (Global Agricultural Productivity Initiative: https://globalagriculturalproductivity.org/). The practice of restorative agriculture recognises soil health as a major concern as well as crop productivity. Restorative agriculture practices such as microbial fertiliser and compost spreading add carbon to the soil, but more importantly they also promote an increase in the number and diversity of bacteria and fungi. The most valuable microbiotas in terms of their impact on soil improvement can be divided into three categories: Plant growth-promoting bacteria (PGPRs), nitrogen-fixing bacteria and arbuscular mycorrhizal fungi together with ectomycorrhizal fungi (Coban et al., 2022). These microbiotas are used to create combinations of microbial fertilisers.

Microorganisms introduced into the plant rhizosphere as microbial fertiliser have been shown to lead significant changes in the functioning, composition and diversity of the soil microbiome, leading to important effects even for subsequent plant generations (Delgado-Baquerizo et al., 2016). Some microorganism used in microbial fertiliser composition can interact with the

plant to stimulate plant-mediated "control" of the microbiome by changing root exudation patterns (Gu et al., 2016), activating additional plant defence mechanisms (Timofeeva et al., 2023) or producing plant-derived antimicrobials (Stringlis et al., 2018). Meanwhile, although some rhizobacteria can colonise in plants, they are unable to transport fixed nitrogen to plants (Ryu et al., 2020). The complexity of plant-microbiome interaction and the very high microbial diversity within the microbiome are among the major challenges to be solved before PGPBs can be directly added to plants as biofertilisers and their efficacy can be enhanced (Han and Yoshikuni, 2022). The obstacles to the full utilisation of the superior properties of PGPBs in agriculture can be removed by synthetic biology applications with synthetic biology, these beneficial microorganisms can be provided with extra abilities and huge steps can be taken in the field of soil improvement and sustainable agriculture.

2. Synthetic Biology

Synthetic biology is an interdisciplinary field that applies engineering principles to existing biological systems to solve various problems and translates them into biological systems that will serve useful purposes. Synthetic biology applications also offer hope for sustainable agriculture. In the last two decades, synthetic biology has evolved into a mature engineering field that enables the development of desired properties that exceed their natural capabilities in a wide range of organisms, including filamentous fungi (Cameron et al., 2014). The Synthetic Biology Research Centre (SynBERC) was established in the USA in 2006. SynBERC was established with the participation of more than one university and thus the foundations of synthetic biology were laid. At the same time, the United States Department of Agriculture started to support related studies in the synthetic biology field (Wang et al., 2022). The first synthetic bacterial genome was developed in 2008 (Gibson et al. 2008). In 2014, Synlogic, one of the first synthetic biology companies, focused on the production of therapeutic first-class synthetic biology

drugs. In 2018, China started to develop projects on the application of synthetic biology to agriculture. The development of synthetic biology has led to new research and new developments in many industrial fields from agriculture to health.

With the development of next-generation sequencing technologies, it has been found that the diversity of the microbiome in roots varies depending on the plant species. The plant co-evolves with its root microbiome (Thesis et al., 2016). To date, plant health protection studies have focused on only a few pathogens and a limited number of beneficial microorganisms (e.g., *Pseudomonas fluorescens*). New molecular techniques have shown that plant microbiome studies should focus not only on combating pathogens but also on improving plant nutrient uptake, and combating biotic and abiotic stresses. In addition, in microbiological studies, not only single strains but also the entire plant microbiota as a community should be investigated. In these studies, new technologies need to be developed continuously.

To create model microbiota-strain collections, *Arabidopsis thaliana* as a model plant was used by Bai et al. (2015). In their study, leaf and root samples of *A. thaliana* plants grown on different soil types and in various environmental sites were collected. Using classic methods, most of the bacterial communities that reproducibly colonise these plants under natural conditions were cultured. A representative microbial collection of 206 root-derived and 224 leaf-derived isolates was established and draft genome sequences were obtained (Bai et al., 2015). The collection of their genomes as well as microbial strains will allow for functional evaluation. This approach allows microbiome selection based on host phenotype.

Genomics identifies all the genes that encode the structural and functional functions of any organism and studies the control of the interaction of these genes with each other and with the environment. The aim of functional genomics is to understand their importance for the organism and to learn the functions of genes by examining the gene expressions of at the genome level in terms of time, form, and quantity. Furthermore, functional genomics is increasingly enabling the discovery of new genes responsible for PGPB activities, including biocontrol, biofertilisation, and bio-stimulation (Ke et al., 2021). Synthetic biology can transfer genes discovered through functional genomics studies to strains that can stably coexist with plants, and so enable these strains to play the same role as PGPRs for microbial fertiliser use (Wang et al., 2022).

Phyto-microbiomes can be designed either top-down or bottom-up. In the bottom-up approach, microorganisms related to specific plant species, strains or organs are derivated from environmental microbiomes (Rodrigues et al., 2018; Toju et al., 2018). Once genetically engineered to carry the desired traits, these core microorganisms are reassembled as synthetic microbial communities (SynComs) (Vorholt et al., 2017). Plants are then inoculated with engineered strains that can strongly recolonise their hosts. In the top-down approach, horizontal gene transfer (HGT) is used for in situ introduction of needed properties into a wide range of hosts. One top-down strategy is to incorporate mobile genetic elements that transfer and integrate exogenous genes into a random subpopulation of microbiomes to enable holistic examination of PGP properties. The development of bacteriophage systems to engineer or remove certain specific species in populations is another top-down strategy, which allow their role to be researched (Ke et al., 2021).

Broad applications of synthetic biology in agriculture include the development of next-generation microbial fertilisers, designing microorganisms that can transform agricultural wastes, developing microbial machines to act as biocontrol agents or play a role in bio-stimulation. It can also provide soil revitalisation by reducing the use of chemical fertilisers, and so contribute to the storage of atmospheric carbon in the soil and solving of the climate crisis (Dodge, 2018).

2.1 PGPR and Synthetic Biology

Plants and their microbiomes are co-evolving holobionts. This association modulates or enhances the environmental adaptation, fitness, competitiveness, abiotic stress tolerance, phytopathogen resistance, health and productivity of plants in agricultural or natural ecosystems (Chiu and Gilbert, 2015; Cesaro et al., 2021; Berg et al., 2021). Such microorganisms can live inside or outside their hosts and colonise abundantly in the rhizosphere or root surface of plants (Bonfante et al., 2010; Turner et al., 2013).

PGPB can synthesise a broad range of compounds such as indole-3acetic acid (IAA), 1-aminocyclopropane-1-carboxylate (ACC) deaminase, extracellular polymeric substances (EPS), antioxidants, and volatile organic compounds (VOC) (Yamaner et al., 2016; Ayuso-Calles et al., 2021). They are used in biofertilizer formulations in agriculture. In addition, it has been reported that they have important effects in preventing economic losses due to stress factors such as drought and salinity caused by the climate crisis (Yamaner, 2021).

The first microorganisms used as biofertilizers were diazotrophic bacteria that can form nodules on the roots of legumes. The first commercial biofertilizer was a diazotrophic formulation called "Nitragin". A patent was granted for this formulation in 1896.

Many biofertilizer formulations on the market shelves contain *Bacillus*, *Brevundimonas*, *Bradyrhizobium*, *Burkholderia*, *Delftia*, *Enterobacter*, *Gluconoacetobacter*, Arthrobacter, *Azosprillum*, *Azatobacter*, *Clostridium*, *Herbaspirillum*, *Klebsiella*, *Paenibacillus*, *Rhiseudobacillus*, *Serratia* species. However, the most commonly used bacteria for biofertilizer formulation are *Azospirillum*, *Bacillus* and *Pseudomonas* (Gómez-Godínez et al., 2023).

Long-term ecological study data have shown that continuous addition of chemical fertilisers to the soil and failure to select microbial combinations appropriate to the plant phenotype have negative effects on plant microbiome diversity. It is important to introduce PGPBs into the soil by rearranging the colonisation properties of plant roots, phytohormone production mechanisms at the gene level and creating plant species-specific microbiomes to remove the barriers to sustainable agriculture. Such studies are important to increase the plant growth-promoting properties of PGPBs or to provide these properties to microorganisms that can be found in abundance in every environment. For example, there are studies on genotype-phenotype mapping using CRISPR technique. These studies were carried out on *E. coli* and yeasts. According to a recent research, *E. coli* was engineered to synthesise IAA, an important plant growth regulator. The resulting strain produced 744 mg/L IAA from 20 g/L glucose in 24 hours (Guo et al., 2019).

Cupriavidus pinatubonensis JMP134 is a neutral rhizobacterium that does not promote plant growth and is also a plant-associated rhizobacterium. This bacterium contains a plasmid designed to produce IAA from L-tryptophan based on quorum sensing (QS) signalling. This genetically engineered bacterium was now able to produce sufficient inducer to activate IAA gene expression and synchronise the entire population to produce IAA. Inoculation of *A. thaliana* with *C. pinatubonensis* strain significantly increased root growth (Zuniga et al., 2018).

In recent research, *Yarrowia lipolytica* was engineered to reproduce gibberellins (GA) GA3, GA4 and GA7, which play a role in salt tolerance and plant development in many higher plants such as soybean, sugarcane and maize (Kildegaard et al., 2021).

Microorganisms can be used as an alternative to expensive and environmentally damaging phosphate fertilisers. In their study, Shulse et al. (2019) achieved heterologous expression of the phytase gene in Proteobacteria. This result shows that microbiome engineering can be used to produce plantassociated strains with phosphate solubilisation capabilities. Microorganisms reprogrammed at the genetic level have the potential to be used in functional natural product discovery.

2.2 Nitrogen-fixing bacteria and synthetic biology

Chemical nitrogen fertilisers, which have been used on agricultural land for many years, degrade soil and water quality, reduce soil biodiversity and increase gas emissions due to the formation of nitrous oxide, a greenhouse gas. Scientific studies have identified biological nitrogen fixation, a function of certain prokaryotes and archaea, as an alternative source of nitrogen with lower production costs and higher utilisation efficiency (Reed et al., 2011). Nitrogen fixation was first characterised in the Rhizobiales (Mus et al., 2016). The ability to fix nitrogen has been detected in many bacterial genera, including Azospirillum, Azotobacter, Burkholderia, Gluconacetobacter, Rhizobium, Sinorhizobium. Azorhizobium. Allorhizobium. Mesorhizobium. Bradyrhizobium, Herbaspirillum, Klebsiella, Paenibacillus and Pseudomonas (Santi et al., 2013). Nitrogen-fixing bacteria can associate with plant roots symbiotically or as endophytes, or they can be free-living nitrogen fixers. These bacteria form special symbioses with plant roots. In order for this symbiotic relationship to occur, each plant requires its own specific bacteria. In addition, for the formation of a sufficient number of nodules and an effective fixation, there must be bacteria specific to that plant in the environment where the plant will grow. If the same plant has been grown in an area before, the bacteria belonging to the plant is already present in that area; but if the plant is to be grown for the first time, it is necessary to introduce the bacteria by inoculation.

Lately, applications of synthetic biology have been used to rearrange the regulation of genes encoding the nitrogenase enzyme complex, providing greater control over nitrogenase expression in various species (Temme et al., 2012). Bloch et al. (2020) used gene editing to improve nitrogen fixation of *Kosakonia sacchari*, a naturally occurring diazotroph proteobacterium related to maize roots that can be applied as a seed coating, reducing fertiliser

applications and increasing yield. While nitrogen fixation of the wild-type strain was observed to be suppressed under conditions with bio-available nitrogen, such as fertilised field experiments and greenhouse, the remodelled strains were able to fix high levels of nitrogen even in the presence of exogenous nitrogen in the rhizosphere of maize in the field and greenhouse.

KV137 (*Klebsiella variicola*137), a synthetic biology product developed for farmers, increased nitrogen fixation activity 122-fold in nitrogen-rich environments. This product has created a fertiliser that will provide the nitrogen needed in maize production. KV137 developed by Pivot Bio company can produce the nitrogen needed by interacting with maize roots. The wild-type *K*. *variicola*137 genome isolated from agricultural soils has been reshaped by synthetic biology (Temme et al, 2020). This bacterium is the active ingredient in PROVEN liquid fertiliser, which reduces the need for chemical fertiliser by 25 lbs per acre and increases yields by up to 5.8 bushels (Temme, 2019). Using this product, the fact that chemicals used in liquid nitrogen fertiliser mix into groundwater with rain is also eliminated. In addition, it has reduced the emission of strong greenhouse gases. In 2021, it was used on 3 million acres of corn.

In addition, a subsidiary called Joyn Bio was formed from the partnership between Ginkgo Bioworks, the synthetic biology company, and Leaps, the investment arm of the life sciences holding Bayer, and the first investments were made in October 2017. Joyn Bio is working to design a microorganism that will allow maize, rice and wheat farmers to reduce nitrogen fertiliser use in half while continuing the same crop yields. They have reported that Ginkgo and Bayer will continue their partnership to develop biological solutions in carbon sequestration, nitrogen optimisation and next generation crop protection. Ginkgo and Bayer are two companies that will continue to work on sustainable food production and safety by using the power of synthetic biology. They also aim to reach large masses by establishing new partnerships in the development of microbial products for agriculture (https://www.ginkgobioworks.com/2022/10/18/ag-biologics-division-bayer-joyn/).

2.3 Mycorrhizae and synthetic biology

Mycorrhiza, which refers to the symbiotic co-operation between plant roots and fungi, are divided into two classes. If the fungal cells form a diffuse layer on the outside of the root, it is called ectomycorrhiza, and if the fungal mycelia penetrate into the plant root tissue, it is called endomycorrhiza. Ectomycorrhiza is mostly found in forest trees, especially in conifers. Mycorrhizal fungi are rarely found in areas far from roots in nature. Therefore, most of them can be considered as obligate symbionts. Despite the close relationship between the fungus and the root, a single pine species can form mycorrhizal associations with more than 40 fungal species.

About 95 % of the plant communities on Earth (about 240 000 plant species) form arbuscular mycorrhizal associations with endomycorrhizas (Koide and Lu, 1992; Bonfante and Perotto, 1995). Nevertheless, we have still very limited information of the underground fungal network. Because of lack of information, the Society for the Protection of Underground Networks (SPUN) has initiated some studies to research and map mycorrhizal fungi worldwide (Popkin, 2022). Mycorrhizal fungi, which form mutualistic relations with plants, naturally sinks billions of tonnes of fixed atmospheric carbon per year in the fungal network in underground (Dodge, 2018). Mycorrhizal fungal networks offer a unique and unexplored opportunity to create enormous carbon sinks globally.

However, when compared to yeasts and bacteria, the synthetic biology background is considerably less developed in filamentous fungi, especially in cap-forming species. These gaps in technological development in filamentous fungi are due to biological factors such as, low efficiency of genetic transformation, slow growth rate and secretion of undesirable enzymes (Meyer et al., 2016).

3. Conclusion

Developments in synthetic biology are exciting. Rapid developments in areas such as DNA synthesis and process automation, biological data science, computer-aided design and machine learning indicate that the capabilities of living beings that can be designed with synthetic biology will be beyond imagination. Realising this development, private companies have started to make investments rapidly. Unfortunately, this super technology can be used for different purposes by malicious people. Detailed strategic plans on biosafety issues should be developed by a community of synthetic biologists, economists, public health protectors, data security units and politicians. Advances in synthetic biology are essential for the sustainability of modern agriculture, as in many other fields.

References

- Ayuso-Calles M., Flores-Felix JD., Rivas R. 2021. Overview of the Role of Rhizobacteria in Plant Salt Stress Tolerance. Agronomy 2021, 11, 1759. https://doi.org/10.3390/agronomy11091759
- Bai Y., M€uller DB., et al. 2015. Functional overlap of the *Arabidopsis* leaf and root microbiota. Nature 528, 364–369.
- Berg G., Kusstatscher P., Abdelfattah A., Cernava T., Smalla K. 2021. Microbiome modulation—toward a better understanding of plant microbiome response to microbial inoculants. Front. Microbiol. 12, 650610. doi: 10.3389/fmicb.2021.650610
- Bloch S.E., Ryu M-H., Ozaydin B., Broglie R. 2020. Harnessing atmospheric nitrogen for cereal crop production. Current Opinion in Biotechnology, 62:181–188.
- Bonfante P., Perotto, S. 1995. Tansley Review No.82. Strategies of arbuscular mycorrhizal fungi when infecting hostplants. New Phytologist 130,3-21.
- Bonfante P., Genre A. 2010. Mechanisms underlying beneficial plant—Fungus interactions in mycorrhizal symbiosis. Nat. Commun., 1, 48.
- Cameron DE., Bashor CJ., Collins JJ. 2014. A brief history of synthetic biology, Nat. Rev. Microbiol. 12 (2014) 381–390, https://doi.org/10.1038/nrmicro3239.
- Cesaro P., Gamalero E., Zhang J., Pivato B. 2021. the plant holobiont volume i: microbiota as part of the holobiont; challenges for agriculture. Front. Plant Sci. 12, 1–3. doi: 10.3389/fpls.2021.799168
- Chiu L., Gilbert SF. 2015. The birth of the holobiont: multi-species birthing through mutual scaffolding and niche construction. Biosemiotics 82, 191–210. doi: 10.1007/s12304-015-9232-5
- Coban O, De Deyn GB, van der Ploeg M. 2022. Soil microbiota as gamechangers in restoration of degraded lands. Science. 2022;375, eabe0725.

- Delgado-Baquerizo M., Maestre FT., et al. 2016. Microbial diversity drives multifunctionality in terrestrial ecosystems. Nat. Commun. 2016, 7, 10541.
- Dodge E. 2018. Chapter 16 carbon deposits—using soil and blockchains to achieve netzero emissions, in: A. Marke (Ed.), Transforming Climate Finance and Green Investment with Blockchains, Academic Press, pp. 217–228, https://doi.org/10.1016/B978-0-12-814447-3.00016-1.
- Gibson, D.G., et al., 2008. Complete chemical synthesis, assembly, and cloning of a mycoplasma genitalium genome. Science, 319 (5867), 1215–1220.
- Gómez-Godínez LJ. Aguirre-Noyola JL. et al. 2023. A Look at Plant-Growth-Promoting Bacteria. Plants, 12, 1668. https://doi.org/10.3390/plants12081668
- Gu Y., Wei Z., Wang X., et al. 2016. Pathogen Invasion Indirectly Changes The Composition Of Soil Microbiome Via Shifts İn Root Exudation Profile. Biol. Fertil. Soils, 52, 997–1005.
- Guo D, Kong S, Chu X, Li X, Pan H. 2019. De novo biosynthesis of indole-3acetic acid in engineered escherichia coli. J Agric Food Chem, 67(29):8186–90. https://doi.org/10.1021/acs.jafc.9b02048.
- Han S-W, Yoshikuni Y. Microbiome engineering for sustainable agriculture: using synthetic biology to enhance nitrogen metabolism in plantassociated microbes. Curr Opin Microbiol. 2022;68, 102172.
- https://www.ginkgobioworks.com/2022/10/18/ag-biologics-division-bayerjoyn/
- https://globalagriculturalproductivity.org/
- https:// population.un.org/wpp/
- Ke J., Wang B., Yoshikuni Y. 2021. Microbiome Engineering: Synthetic Biology of Plant-Associated Microbiomes in Sustainable Agriculture. Trends in Biotechnology, March , Vol. 39, No. 3 245-261.

- Kildegaard KR, Arnesen JA, et al. 2021. Tailored biosynthesis of gibberellin plant hormones in yeast. Metab Eng, 66:1–11. https://doi.org/10.1016/j. ymben.2021.03.010
- Koide RT, Lu X. 1992. Mycorrhizal infection of wild oats: parental effects on offspring nutrient dynamics, growth and reproduction. in "mycorrhizas in ecosystems" (Eds. Read, D.J., Lewis, D.H., Fitter, A.H. and Alexander, I.J.) Cab International, Wallingford, U.K. Pp. 55-58.
- Lal R. 2014. Societal value of soil carbon, Journal of Soil and Water Conservation, 69(6).
- Meyer V., Andersen M.R., et al. 2016. Current challenges of research on filamentous fungi in relation to human welfare and a sustainable bioeconomy: a white paper, Fungal Biol. Biotechnol. 3; 6, https://doi.org/10.1186/s40694-016-0024-8.
- Mus F, Crook MB, et al. 2016. Symbiotic nitrogen fixation and the challenges to its extension to nonlegumes. Appl Environ Microbiol 2016, 82:3698-3710.
- Popkin G. 2022. A fungal safari, Science 377;142–147, https://doi.org/10.1126/ science.add7606.
- Reed SC, Cleveland CC, Townsend AR. 2011. Functional ecology of freeliving nitrogen fixation: a contemporary perspective. Ann Rev Ecol Evol Syst 2011, 42:489-512.
- Rodrigues RR., Rodgers NC. et al. 2018. COREMIC: a web-tool to search for a niche associated CORE MICrobiome. PeerJ 6, e4395
- Ryu M-H, Zhang J, Toth T, et al. 2020. Control of nitrogen fixation in bacteria that associate with cereals. Nat Microbiol. 5:314–330.
- Santi C., Bogusz D., Franche C. 2013. Biological nitrogen fixation in nonlegume plants. Ann Bot, 111:743-767.

- Shulse CN, Chovatia M, et al. 2019. Engineered root bacteria release plantavailable phosphate from phytate. Appl Environ Microbiol 85:e01210-19. https://doi.org/10.1128/AEM.01210-19.
- Stringlis IA., Yu K., et al. 2018. MYB72-Dependent Coumarin Exudation Shapes Root Microbiome Assembly to Promote Plant Health. Proc. Natl.Acad. Sci. USA , 115, E5213–E5222.
- Temme K, Zhao D, Voigt C.A. 2012. Refactoring the nitrogen fixation gene cluster from Klebsiella oxytoca. Proc Natl Acad Sci U S A, 109:7085-7090.
- Temme K. 2019. Pivot ProveN Performance Report. (Pivot Bio, 2019).
- Temme K., Tamsir A., Bloch S., Clark R., Tung E. 2020. Methods and Compositions for Improving Plant Traits (ed USPTO) (Pivot Bio, Inc., USA).
- Theis KR., Dheilly NM., et al. 2016. Getting the hologenome concept right: an eco-evolutionary framework for hosts and their microbiomes. mSystems 1, e00028–16.
- Timofeeva AM., Galyamova MR., Sedykh SE. 2023. Plant Growth-Promoting Bacteria of Soil: Designing of Consortia Beneficial for Crop Production. Microorganisms, 11, 2864. https://doi.org/ 10.3390/microorganisms11122864
- Toju H. et al. 2018. Core microbiomes for sustainable agroecosystems. Nat. Plants. 4, 247–257.
- Turner TR., James EK., Poole PS. 2013. The plant microbiome. Genome Biol., 14, 209.
- Vorholt JA et al. 2017. Establishing causality: opportunities of synthetic communities for plant microbiome research. Cell Host Microbe 22, 142– 155.
- Wang L., Zang X., Zhou J. 2022. Synthetic biology: A powerful booster for future agriculture. Advanced Agrochem 1: 7–11.

- Yamaner Ç. 2021. Bitki büyümeyi teşvik edici bakterilerin bitkilerdeki tuz stresini azaltmadaki önemli metabolik Aktiviteleri. 3. International Baku Scientific Research Congress October 15-16 / Baku, AZERBAIJAN / Baku Eurasia University
- Yamaner et al. 2016. Mısır rizosferindeki fosfat çözebilen mikroorganizmaların genetik ve fonksiyonel çeşitliliğinin araştırılması. I. Ulusal Tarımsal Biyoteknoloji Kongresi, Haziran 1-3 / Samsun /Türkiye. Ondokuz Mayıs Üniversitesi.
- Zuniga A, Fuente F, et al. 2018. An engineered device for indoleacetic acid production under quorum sensing signals enables *cupriavidus pinatubonensis* jmp134 to stimulate plant growth. ACS Synth Biol, 7(6):1519–27. https://doi.org/10.1021/acssynbio.8b00002

CHAPTER 4

EVALUATION OF VERMICOMPOST IN TERMS OF SUSTAINABILITY OF AGRICULTURAL MANAGEMENT

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1. Introduction

Soils can be rendered inefficient by agricultural activities applied during agricultural production. Soil conservation is an important parameter in sustainable agriculture. Chemical applications applied to soils for many years cause a decrease in organic matter content. Organic matter supplementation is necessary to minimize the losses that may occur in yield in soils exposed to continuous cultivation and cultivation and to ensure continuity in agriculture. The organic matter content in Turkish soils is decreasing over time and negatively affects the physical, chemical and biological properties of soils (Demirtaş, 2004).The most important factor for sustainable agriculture is the use of organic wastes as regulators in soils. Vermiculture is the transformation of wastes into quality vermicompost (vermicompost fertilizer) in a short period of time (Erşahin, 2007).

Vermicompost is a type of compost formed by worms digesting organic wastes in their bodies. The characteristics of the wastes added during the formation process of vermicompost play an important role in determining the content of vermicompost and it has been reported that in soils with earthworm activities, firstly organic C and then total N amounts show a significant increase (K121lkaya and Hepşen, 2007; Namlı et al., 2014).The end product, vermicompost, is stated as a method that can be applied to reduce fertilizer use by converting waste into organic fertilizer (Blouin et al., 2019). Vermicompost acts to secrete various antibiotics, enzymes and plant growth regulators in the rhizosphere, which is the plant root zone (Maltaş et al., 2017). With these effects, vermicomposts are a very good soil organizer consisting of a finely separated, peat-like material with high porosity, good water holding capacity and microbial activities (Atiyeh et al., 2001).

Based on this information, in this study, vermicompost was evaluated in terms of its usability, crop yield, soil conditioner and environmental effects compared to other alternative organic fertilizer materials in order to ensure the sustainability of agricultural activities.

2. RECYCLING ORGANIC WASTE WITH VERMICOMPOST

Composting is one of the most successful methods of recycling and reintroducing organic wastes back into nature. The vermicomposting method, which has outstanding features compared to traditional composting, has recently gained wide application areas.

Providing organic fertilizer is less costly than chemical fertilizer and organic fertilizer is a recycling material in order to protect the natural balance. It is possible to produce organic fertilizer by using many products such as household wastes, market and market wastes, greenhouse wastes (herbal green parts and fruits), park and garden pruning wastes, grass wastes, etc. These wastes can be fermented for certain periods of time by composting method to produce food for worms. The nutrients produced for worms must provide appropriate pH and temperature values. These values depend on the fermentation time and the content of the materials used for the composting process. The products obtained by the composting process can be used as a food source for earthworms. Earthworms consume the composting products and convert them into vermicompost, which is a richer material and increases the nutrient content of organic waste.

The compost products obtained by the traditionally used composting method can lose approximately 55% of organic matter and 30-50% of nitrogen nutrients (Ketkar, 1993). However, it has been reported that the use of earthworms in compost production from organic wastes, their interaction with other decomposing organisms (Sampedro and Dominguez, 2008) reduces the time spent in the formation phase in maturing composts, provides a more

uniform appearance (Atiyeh et al., 2000), finer structure and large surface area (Shi-wei and Fu-zhen, 1991).

There are differences in the stage of compost production, and in the past years, studies on the effects of compost and vermicompost on plants and soil have increased (Ngo et al., 2011). It can be observed that biological activity increases in agricultural soils where vermicompost is applied. Vermicompost contains a lot of microorganisms, and the application of this material to soils is expected to increase microbial activities important for plants and soils, such as the conversion of plant nutrients, the formation of regulators that provide growth in plants, the formation of resistance in plants or the increase in resistance to diseases (Arancon et al., 2006).

In this study, tobacco stalks and horse manure mixtures were used as organic wastes to obtain food sources for Eisenia fetida earthworms and the possibilities of vermicomposting were investigated. The wastes were subjected to vermicomposting for 3 months and chemical, microbial and biochemical parameters were analyzed in samples taken at 30, 60 and 90 days. Biochemical parameter values decreased, maximum enzyme activity was determined at day 60 and minimum enzyme activity was determined at day 30. The humification index value, which is a stability parameter in vermicompost samples, was determined to be above 5 and this shows that the stabilization process did not occur in the vermicomposting stage. At the end of the 3-month period, it was recommended to prepare more appropriate ratios of composting mixtures prepared with the death of worms and to conduct preliminary trials in subsequent studies (Kayıkçıoğlu et al., 2016).

The amount of organic matter in soils in our country is decreasing day by day due to unconscious agricultural practices. Depending on the decrease in the organic matter in the soil, the water holding capacity of the soils, the airfilled part required in the soil, plant nutrients and microorganism activities in the soil, such as a decrease in plant yield and inefficiency in soils, and the balance in sustainable agriculture is disrupted. In order to ensure the continuity of sustainable agriculture, the use of chemical fertilizers should be reduced, the practices of obtaining maximum yield per unit area from soils due to the increasing population day by day should be reduced and soils should be protected with organic fertilizer applications.



Figure 1. Recycling of organic waste with vermicompost (1)

3. USE OF VERMICOMPOST IN AGRICULTURAL PRODUCTION

Vermicompost is produced by using the fertilizer produced by earthworms and organic wastes together (Garg et al., 2010). Vermicompost aims to increase the activities of earthworms that recycle macro and micro nutrients in nature (Şimşek-Erşahin, 2007). The uptake forms of nutrients are of great importance for plant development. Approximately 97% of the nutrients in vermicompost are in the form that can be taken up by plants and primarily nitrogen, phosphorus and potassium are in the form that can be taken up by plants (Barley, 1961).

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The amount of organic matter in the soil is of great importance for the protection and maintenance of soil fertility and high crop yield (Önal et al., 2003). While the use of organic inputs in agriculture regulates the physical properties of soils, the natural balance is preserved by recycling wastes (Nazli et al., 2016).



Figure 2. Use of vermicompost (2)

This study was carried out to determine the effects of biochar and vermicompost mixtures on some biological properties of the soil during the incubation phase by applying different doses of biochar and vermicompost to the soil and it was determined that biochar and vermicompost mixtures had positive effects. As a result of the studies, it was determined that the biological properties of the soil increased in 100% biochar and 100% vermicompost, urease enzyme activity increased in 100% biochar and 100% biochar during the 60-day incubation period, the amount of organic matter in the soil increased in 100% biochar during the 40-day incubation period and arylsulfatase enzyme activity increased in 50% biochar + 50% vermicompost during the 60-day incubation period. Due to the high C and C/N ratio in biochar, it was suggested

that it was inadequate to be used alone and that vermicompost material with a narrow C/N ratio should be used together (Y1lmaz and Kurt, 2018).

Vermicompost was applied at different rates and its effects on yield and nutrients in maize plants were investigated. The application rates were 0%, 10%, 20%, 20%, 30%, 30%, 40% and 50%. The amount of dry matter of green parts of maize plants was determined. It was determined that the amount of dry matter increased as vermicompost rates increased in maize plants. The maximum amount of dry matter in maize plant was determined as 3.98 g/plant in 40% vermicompost application. The maximum amount of phosphorus (0.80%), nitrogen (4.24%) and potassium (5.75%) were determined in 30% vermicompost application. According to the results obtained from the research, it was determined that the amount of macro plant nutrients increased as the amount of vermicompost application increased, while the amount of micro nutrients decreased and it was recommended to use vermicompost for sustainable agriculture (Durukan et al., 2020).

The effects of certain ratios and mixtures of vermicompost and mycorrhizae on the growth and mineral concentration of pepper plants were investigated. Nutrient concentration, wet and dry weight ratio of pepper plants were determined. As a result of these applications, it was determined that they had a positive effect on nutrient concentration, wet and dry weight ratios in pepper plants (Küçükyumuk et al., 2014).

When vermicompost was applied to maize plant, it was stated that it provided an increase in the above-ground parts. According to the results obtained from the research, it had a positive effect on the amount of N, P, K, Zn, Ca, Mg nutrients, while it had a negative effect on the amount of Fe, Mn and Cu (Durukan, 2020).

In this study, vermicompost was applied to tomato, pepper and strawberry plants, and according to the results obtained, it was stated that it increased the market value of strawberry fruit to a great extent, and increased the development and leaf area in the shoot part for tomato and pepper plants (Arancon et al., 2003).

The effects of different doses of vermicompost on yield and factors affecting yield in potato plants were examined. According to the results obtained from productivity analyses and measurements of green parts of potato plants, the highest values among the application doses were obtained from 12 tons/da vermicompost application (Yourtchi et al., 2013).

The effects of vermicompost and mycorrhizae in cowpea cultivation in calcareous soils were determined as the amount of vermicompost increased depending on the application doses, the above-ground wet and dry weights of the plants and the wet and dry weights in the root area increased (Şahin and Ataklı, 2021).



Figure 3. Images of matured vermicompost

4. RECOMMENDED VERMICOMPOST APPLICATION AMOUNTS IN AGRICULTURAL ACTIVITIES

Different proportions of vermicompost specific to the plant to be cultivated in crop production are recommended. Different rates of vermicompost are recommended depending on the needs of the plant and vermicompost amounts are recommended for maximum yield.

PLANT GENUS	AMOUNT	UNIT
Cereals and field crops	100-200	kg/da
Greenhouse and field vegetables	100-150	kg/da
Fruit trees	0.5-4	kg/ağaç
Flowering garden plants	300-400	g/m ²
Grass	300-400	g/m ²
Flowering potted plants	300-400	g/saksı
Potted plants without flowers	150-200	g/m ²

Table 1. Recommended amounts of vermicompost in agricultural applications (3)

In agricultural activities, different fertilization rates are recommended for different plant species depending on the amount of nutrients in vermicompost. As given in Table 1, application doses are recommended according to their units. For example, vermicompost should be applied in different units and rates for fruit trees and flowerless potted plants (Table 1).

PARAMETER	TRADITIONAL	VERMICOMPOST
	COMPOST	
pH	7.80	6.80
EC(mmhos/cm)	3.60	11.70
Total Kjeldal Nitrogen(%)	0.80	1.94
Nitrate Nitrogen(ppm)	156.50	902.20
Phosphate(%)	0.35	0.47
Potassium(%)	0.48	0.70
Calcium(%)	2.27	4.40
Sodium(%)	< 0.01	0.02
Magnesium(%)	0.57	0.46
Iron(ppm)	11.690	7.563
Zinc(ppm)	128	278
Manganese(ppm)	414	475
Copper(ppm)	17	27
Borom(ppm)	25	34
Aluminum(ppm)	7.380	7,012

 Table 2. Chemical contents of conventional compost and vermicompost (5)

When the chemical contents of traditional compost and vermicompost fertilizers given in Table 2 are compared, nitrate (ppm) content was measured as 156.50, 902.20 respectively and the nitrate content in vermicompost fertilizer

was higher. Most of the parameters analyzed to determine the chemical contents were found to be higher in vermicompost fertilizer.

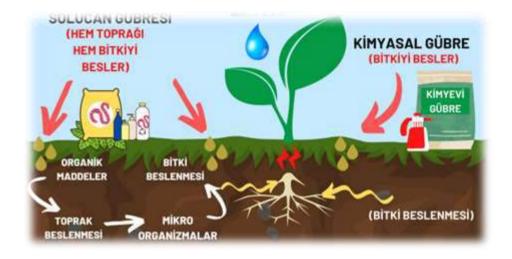


Figure 4. Use of vermicompost (vermicompost) for crop growth and soil quality(4),

5. VERMICOMPOST PRODUCTION FACILITIES



Figure 5. Images from inside the vermicompost production facility





Figure 6. Worm beds with continuous flow and casing systems

5.1 Vermicompost food prepared for vermicompost production





Figure 7. Images from food preparation for vermicompost production



Figure 8. Images from the preparation of food for worms in preparation for analysis

6. CONCLUSION

Ensuring the continuity of sustainability in agriculture is directly linked to the practices applied to soils. As a result of chemical fertilization and spraying applied to soils for many years, low yields and deterioration in soil quality occur. In this research, examining the effects of vermicompost on soil and plants contributes to sustainable agriculture. In addition to the effects on soil and plants with the use of organic wastes in the formation phase of vermicompost, the effects on the environment in the long term and its potential to be used as an alternative to existing organic fertilizers were examined. One of the most important effects of vermicompost that makes it an alternative fertilizer instead of other fertilizers is the protection of the environment by recycling wastes.

The organic matter content in the soils of our country is low and it is becoming difficult to prevent this decrease day by day. In scientific studies, the decrease in the amount of organic matter and the applications made to prevent this decrease are not at the desired level in the soils of our country. Depending on the increase in scientific researches and the increase in experience, it will be a guide in the agricultural production to be carried out in the following years and at the same time, the confidence of the users towards vermicompost will increase.

As a result of these applications, organic wastes are used and returned to nature, protecting the natural balance, having positive effects at the economic level, sustainable agriculture on soils, quality and healthy plant production, etc. It benefits the environment with many positive effects such as. Reducing chemical inputs in agricultural practices and recycling organic wastes are of great importance for the continuity of sustainable agriculture and at the same time, it protects the natural balance by saving agricultural inputs.

7. References

- Demirtaş, I.E. 2004. Kentsel katı atık kompostunun tarımda kullanımı. Derim, 21(2): 27-34.
- Erşahin Y, 2007. Vermikompost Ürünlerinin Eldesi ve Tarımsal Üretimde Kullanım Alternatifleri. Gaziosmanpaşa Üniversitesi Ziraat Fakültesi Dergisi 24(2): 99-107.
- Blouin, M., Barrere, J., Meyer, N., Lartigue, S., Barot, S., Mathieu, J., 2019.Vermicompost Significantly Affects Plant Growth: A Meta-Analysis.Agronomy for Sustainable Development, 39(4): 34.
- Atiyeh, R.M., Edwards, C.A., Subler, S., Metzger, J.D., 2001. Pig manure As A Component of A Horticultural Bedding Plant Medium: Effect on Physico Chemical Properties and Plant Growth. Bioresource Technology, 78(1): 11–20.
- Maltaş, A.Ş., Tavalı, İ.E., Uz, İ., Kaplan, M., 2017. Kırmızı Baş Lahana (Brassica oleracea var. capitata f. rubra) Yetiştiriciliğinde Vermikompost Uygulaması. Mediterranean Agricultural Sciences, 30(2):155-161.
- Kızılkaya R, Hepşen Ş, 2007. Microbiological properties in earthworm Lumbricus terrestris L. cast and surrounding soil amended with various organic wastes. Communication in Soil Science and Plant Analyses 38: 2861-2876.
- Namlı A, Akça O, Perçimli C, Beşe S, Gür Ş, Arıkan H, Eser İ, İzci E, Gümüşay E, Tunca G, Khálau IJ, Mutafçılar Z, Demirtaş Ö, 2014. Evsel ve endüstriyel arıtma çamurlarının solucanlar (Eisenia fetida) ile kompostlanması. Toprak Bilimi ve Bitki Besleme Dergisi 2(2): 46-56.
- Şimşek-Erşahin, Y., 2007. Vermikompost Ürünlerinin Eldesi ve Tarımsal Üretimde Kullanım Alternatifleri. GOÜ. Ziraat Fakültesi Dergisi, 24 (2), 99-107.

- Garg, V.K., Gupta, R. and Yadav, A., 2010. Vermicomposting Technology for Solid Waste Management. http://www.environmental-expert.com Erişim Tarihi: 03.12.2013
- Barley, K. P. 1961. Plant nutrition levels of vermicast. Advances in Agronomy. 13, 251.
- Kayıkçıoğlu, H. H., Okur, N. ve Bayız, O. 2016. Toprak Solucanları ile Kompostlaştırılmış Tutun Atıklarının Vermikompost Olarak Değerinin Belirlenmesi, Bu çalışma, E.Ü.B.A.P tarafından desteklenen 2011-ZRF-030 no'lu projeden hazırlanmıştır, Ege Üniversitesi, Ziraat Fakültesi, Toprak Bilimi ve Bitki Besleme Bölümü, 35100, İzmir/Türkiye, Sorumlu Yazar: husnu.kayikcioglu@gmail.com
- Durukan, H., Saraç, H. ve Demirbaş, A. 2019. Farklı Dozlarda Vermikompost Uygulamasının Mısır Bitkisinin Verimine ve Besin Elementleri Alımına Etkisi, Ziraat Fakültesi Dergisi Türkiye 13. Ulusal, I. Uluslararası Tarla Bitkileri Kongresi Özel Sayısı:45-51, 2020 ISSN 1304-9984, Araştırma Makalesi Sivas Cumhuriyet Üniversitesi, Sivas Meslek Yüksekokulu, Bitkisel ve Hayvansal Üretim Bölümü, Sivas, Türkiye. Sorumlu yazar: hasandurukan@cumhuriyet.edu.tr
- Yılmaz, F. I. ve Kurt, S. 2018. Biyokömür ve vermikompost uygulamalarının toprağın bazı biyolojik özellikleri üzerine etkisi, Ordu Üniversitesi, Ziraat Fakültesi, Toprak Bilimi ve Bitki Besleme Bölümü, Ordu Toprak Bilimi ve Bitki Besleme Dergisi, 6(2) 143-150.
- Küçükyumuk, Z., Gültekin, M., ErdaL, İ. 2014. Vermikompost ve Mikorizanın Biber Bitkisinin Gelişimi ile Mineral Beslenmesi Üzerine Etkisi.
 Süleyman Demirel Üniversitesi Ziraat Fakültesi Toprak Bilimi ve Bitki Besleme Bölümü, Isparta Sorumlu yazar: zelihakucukyumuk@sdu.edu.tr. Süleyman Demirel Üniversitesi Ziraat Fakültesi Dergisi 9 (1):51-58, 2014 ISSN 1304-9984, Araştırma Makalesi.

- Durukan H., Saraç H. & Demirbaş A. (2020). Farklı Dozlarda Vermikompost Uygulamasının Mısır Bitkisinin Verimine ve Besin Elementleri Alımına Etkisi. Ziraat Fakültesi Dergisi, Türkiye 13. Ulusal, I. Uluslararası Tarla Bitkileri Kongresi Özel Sayısı:45-51.
- Arancon N.Q., Edwards C.A., Bierman P., Metzger J.D., Lee S. & Welch C. (2003). Effects of vermicomposts on growth and marketable fruits of field-grown tomatoes, peppers and stawberries. Pedobiologia 47: 731-735.
- Yourtchi M.S., Hadi M.H.S. & Darzi M.T. (2013). Effect of nitrogen fertilizer and vermicompost on vegetative growth, yield and NPK uptake by tuber of potato (Agria CV.). International Journal of Agriculture and Crop Sciences, 5(18), 2033-2040.
- Şahin, S. ve Ataklı, S., B. (2021). Kireçli Toprakta Börülcenin Fide Gelişimi Üzerine Vermikompost ve Mikoriza Uygulamalarının Etkileri, Türk Tarım–Gıda Bilim ve Teknoloji Dergisi, 9 (sp): 2659 - 2662, sezer.sahin@gop.edu.tr, seda.bice@gop.edu.tr.
- (3)https://www.google.com/url?sa=i&url=https%3A%2F%2Fm.facebook.co m%2Fsosg25%2F&psig=AOvVaw2zA1O6QCJGAxNxwbRujvXY& ust=1702455250554000&source=images&cd=vfe&opi=89978449&v ed=0CBAQjRxqFwoTCIi7vqW6iYMDFQAAAAAAAAAAAAAA

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iYMDFQAAAAAdAAAABAD

(5)https://www.google.com/url?sa=i&url=https%3A%2F%2Fwww.agroenerji .com%2FSolucanGubresi.aspx&psig=AOvVaw1foUP_xBN6qDvDN d4V-

YIz&ust=1702466271358000&source=images&cd=vfe&opi=899784 49&ved=0CBAQjRxqFwoTCJj92p_jiYMDFQAAAAAAAAAAAA D

CHAPTER 5

THE USE OF GEOSTATISTICS IN SOIL SCIENCE AND AN OVERVIEW OF STUDIES

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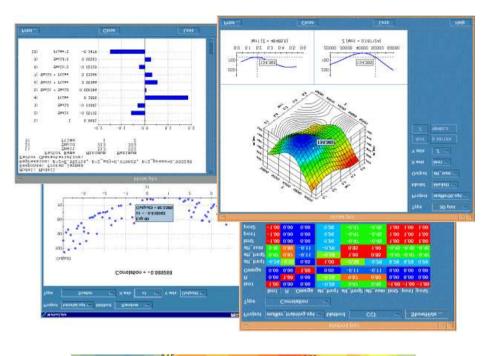
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1. Introduction

As the world's population is increasing over time, it is aimed to obtain maximum yield in the products to be obtained per unit area in agricultural production. Accordingly, it is possible to determine the local variability in the lands and as a result of the applications to be made specific to the region, it is possible to provide the required amount of applications. With the practices carried out by determining the local variability in agricultural areas, both sustainable agriculture and sensitive agriculture can protect the soils to be transferred to future generations. There are geostatistical methods used to determine spatial variability in soil. Among these methods, methods such as ordinary kriging, inverse distance weighing and Co-Kriging methods are used to estimate soil properties. Among the methods used to estimate soil properties, the prediction model that most supports the accuracy of the predictions is used and this varies for each parameter and each soil.

Soil properties change over short and long distances as a result of genetic structure and anthropogenic influences (Webster and Oliver, 2007). The fact that the physical and chemical properties of soils in agricultural cultivation vary with distance shows a significant variation in land area in crop production even in only one plot area (Cambardella et al., 1994). Accordingly, it is stated that determining the spatial variability in soils is beneficial in farming for productivity, conservation of nutrients in the soil and management of other agricultural practices (Mali et al., 2016).

The local variability determined in the soils maps the study area by creating predictions in the points that are not sampled according to the data at the sampled points and it is possible to make applications based on this map. With the integration of the maps obtained into precision agricultural tools, it is possible to make region-specific applications.



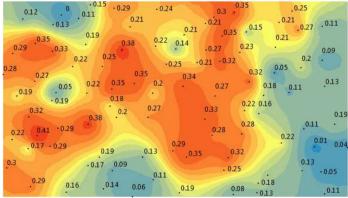


Figure 1. Images from data processing during geostatistical analysis (1)

2. Geostatistics in soil science

Since the soil has a heterogeneous structure, applying the same agricultural practices to every area of the land causes negative results in terms of sustainable agriculture. After determining the regional variability of the soil, it is important to apply agricultural practices according to these variabilities in order to reduce the situations that negatively affect sustainable agriculture such as soil degradation and excessive agricultural input application. Accordingly, the use of geostatistics to determine the spatial variability of soils is becoming increasingly widespread. Depending on the soil properties that show spatial dependence with soil samples taken from certain distances from the land, maps representing the area can be obtained by making predictions in areas where soil sampling is not performed.

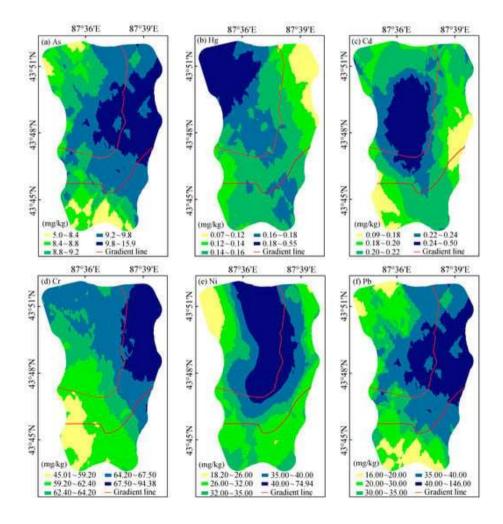


Figure 2. Geostatistical maps showing variability in a terrain with given coordinates (2)

3. Studies using Ordinary kriging method in soil science

Determination and mapping of spatial variability of soil properties is of great importance for sustainable production in areas of intensive agriculture. In this study, physical and chemical properties of soil samples taken from a part of the Tigris Basin were determined. Distance dependent variability of the obtained soil properties was determined using models and the area was mapped based on the models. Soil samples were divided into grids between Divarbakır and Siirt provinces and a total of 175 samples were taken from 0-20 cm depth. While determining the spatial variability, some soil samples were taken at certain distances and a total of 33 soil samples were taken with a distance of 250, 750, 1750 meters between the points located at the corners of two consecutive grids in order to estimate more accurately. The soil samples were analyzed for texture, organic matter, lime, pH, EC, available P and K, their variability depending on the distance was determined and estimations were made at the points where sampling was not done and maps representing the area were obtained accordingly. According to the results obtained from the study, the lowest variability depending on the distance was determined as pH and the highest variability was determined as P. With the maps obtained as a result of the analyzes, it is possible to make applications according to the variability in the field and at the same time, it will provide maximum efficiency from the inputs depending on the application made at the required level (Budak et al., 2018).

Determination and mapping of the distance-dependent variability of soil properties in agricultural lands enables sustainable agricultural management and determination of change in time and space. In this study, useful phosphorus was determined using two geostatistical methods and maps were obtained. The geostatistical methods used were ordinary kriging and sequential Gaussian simulation. A total of 800 degraded soil samples were taken from 0-30 and 30-60 cm depths from Tokat Kazova plain by random sampling. The average useful P obtained from the study was 14.18 mg/kg in the topsoil and 8.87 mg/kg in the subsoil. The useful phosphorus obtained by Ordinary kriging method was found to be low in 11.56% of the topsoil, sufficient in 82.16% and excessive in 6.28% of the topsoil. According to the results obtained by sequential Gaussian simulation method, 9.98% of the topsoil was found to be low, 86.27% of the topsoil was found to be sufficient and 3.75% of the topsoil was found to be excessive. In the lower soil samples taken from 30-60 cm, there are more differences in the results obtained according to the methods used in the upper soil samples. The maps obtained at the end of the study will contribute to the decision makers about the amount of fertilizer to be used and to protect the environment while applying phosphorus (Akbaş, 2012).

In this study, chemical soil quality was evaluated by using factor and geostatistical analysis methods in vertisol soils taken from Bafra Delta Plain. In the data set, chemical properties were determined in the soils taken from 0-30 cm depth and 80.16% of the total variance was explained. In the soils taken from 30-60 cm depth, 5 factors explained 77.77% of the total variance. With the mapping showing the distribution in the area, it was determined that it is important to guide the applications to be made for the sustainability of soil quality (Sağlam and Dengiz, 2013).

In this study, using ordinary kriging and universal kriging methods in geostatistical methods, total alpha and total beta activities in natural spring waters in Artvin province were estimated at unsampled points and their distributions in the region were determined. Maps showing the distribution of alpha and beta activities were produced. Water samples were taken from 120 stations to represent the study area and 50% of the randomly selected samples were used to create the model, while the remaining 50% of the samples were used for verification in the data obtained as a result of the model. The highest measurement value for alpha activity was 0.771 Bq/L and the lowest

measurement value was 0.005 Bq/L. For beta activity, the highest value was 0.808 Bq/L and the lowest measurement value was 0.0013 Bq/L. Among the methods used, the universal kriging method gave better spatial distribution and produced maps that better represent the distribution of alpha and beta activities in mapping (Yeşilkanat et al., 2014).

Within the scope of this study area, the spatial distributions of the properties of soil samples taken from 0-30 cm depth from 53 different points of agricultural lands in Siverek district of Sanliurfa province were determined with variograms and coefficient of variation. The properties of the soils formed on basaltic parent material and their spatial distributions were examined by geostatistical method. Among the samples examined in the study area, based on the coefficient of variation values, it was determined that pH was the most homogeneous parameter and silt was the most heterogeneous parameter. According to the results obtained from the study, it was determined that the distance between the sampling points should be kept lower and the number of sampling should be increased for some parameters of the soils formed on the basaltic parent material. Exponential and Gaussian models, which are models used for different parameters of the study area soils, were compared and the spatial distributions of clay, silt, organic matter, pH, EC were defined with the exponential model, and sand content was defined with the Gaussian model. Using a different model was determined by the RMSE (Root Mean Square Error) value. The model with a lower RMSE value is closer to the real values. When the soil maps created were examined, it was determined that EC and clay content decreased towards the north, and organic matter content was higher in the lands located in the west of the land. Good soil management can be achieved with geostatistical methods (Aygür, 2020).

In a study conducted by Yetgin (2004), the spatial variability of the physical properties of soils was analyzed by geostatistical methods. According to the research results, it was determined that the soil properties that showed

the highest variability were lime and saturated hydraulic conductivity. Additionally, as a result of the study, it was determined that the kriging method can be used safely to interpolate the soil properties in the study area.

In a study conducted by Ağca (2015), the spatial distribution of heavy metal contents in soils around Iskenderun was determined. According to the research results, it has been determined that the most suitable semivariable models for heavy metals are exponential and spherical models. In addition, some of the heavy metals showed moderate autocorrelation, while others showed strong autocorrelation.

4. Studies using Co-Kriging method in soil science

In this study to determine the applicability of the Cokriging method for field capacity estimation, the study was conducted in 8 plots of 32 km2 in a region between Ankara and Konya. A total of 135 surface soil samples (0-20 cm) were taken by grid method at 500 m intervals in the study area. The lowest mean absolute error and mean squared error were obtained in the estimation of field capacity with clay auxiliary variable by Cokriging method. However, the fact that the mean of the field capacity values (33.38) and the mean of the data obtained with cokriging co-estimation (33.20) were close to each other indicates that the method can be used in long and costly analyzes (Tuncay et al., 2017).

The spatial distribution of rainfall was tried to be determined by using TRMM (Tropical Rainfall Measuring Mission) satellite rainfall product 3B43 and Co-Kriging (CK), a geostatistical method using height as an auxiliary variable. According to the spatial precipitation estimation performance results determined by TRMM and CK methods, the CK method gave a lower error value (Taş, 2018).

In this study, multivariate geostatistical methods were used, land topography (elevation) was used as the second parameter for the estimation of precipitation parameter and better results were obtained compared to univariate geostatistical methods (Khorrami and Gündüz, 2019).



Figure 3. An example of Google Earth images of coordinate points with geostatistical methods

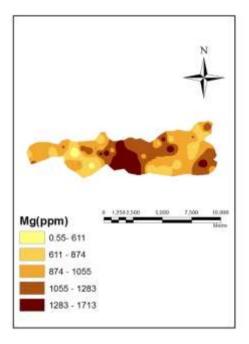


Figure 4. Soil Mg content map obtained by IDW (Inverse Distance Weighting) method, one of the geostatistical methods (Kaplan, 2020)

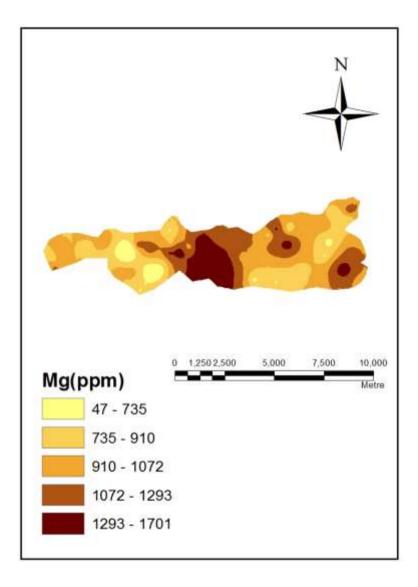


Figure 5. Soil Mg content map obtained by kriging method from geostatistical methods (Kaplan, 2020)

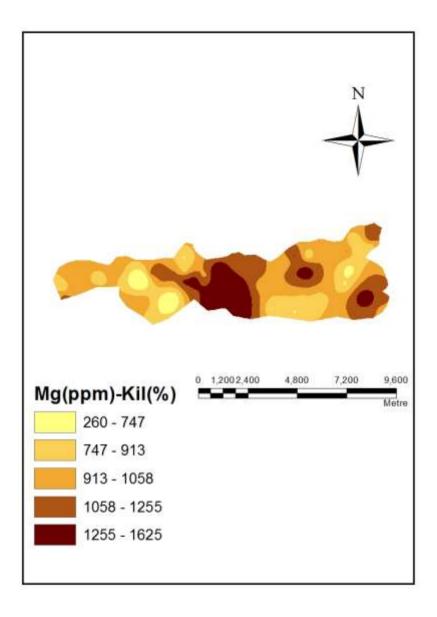


Figure 6. Soil Mg content map obtained by Co-kriging method from geostatistical methods (Kaplan, 2020)

While estimating soil properties with geostatistical methods, maps showing regional variability were obtained according to different methods used. While only the Mg parameter was used in the laboratory results while creating prediction maps with IDW and kriging, clay was used as the second parameter for the Mg parameter in the Co-kriging method.

In this study, soils consisting of basalt parent material were sampled from a depth of 0-30 cm in Siverek district of Sanlıurfa province and soil properties were determined by geostatistical methods. Among the geostatistical methods, IDW (Inverse Distance Weighting), Ordinary Kriging (OK), Co-Kriging (COK) methods were used and geostatistical analyzes were carried out using the ArcMap program. The accuracy of the predictions of the different methods used was determined using the root mean square error (RMSE; Root Mean Square Error). According to the results obtained from the study; The lowest RMSE value (0.18) for the pH parameter is the Ordinary Kriging (OK) method; The lowest RMSE value (2.43) for the sand (%) parameter is the Co-Kriging (COK) method; lowest RMSE value for exchangeable Ca (ppm) parameter (2002) Co-Kriging (COK) method; The lowest RMSE value for the variable K (ppm) parameter is (178) Co-Kriging (COK) method; Co-Kriging (COK) method gave the lowest RMSE value (70.9) for the exchangeable Na (ppm) parameter. When geostatistical methods are examined, in general, the Inverse Distance Weighting (IDW) method gave the highest RMSE result. The Co-Kriging (COK) method gave the lowest RMSE result. Regional variations were determined by obtaining maps representing the study area (Kaplan, 2020).

5. Conclusion

The use of technology in agriculture is recommended as it saves time, chemical use and manpower in agricultural applications. By using univariate and multivariate geostatistical methods, the variability in the land can be estimated and mapped. Accordingly, it is possible to see the variability in the land on the map and to make agricultural practices according to the density on the map. While carrying out agricultural activities, it is of great importance to utilize precision agriculture technology to contribute to sustainable agriculture and to prevent soil degradation. Geostatistics, which is used as precision agriculture technology, enables mapping according to regional differences of soils. Mapping provides the benefit of making region-specific agricultural practices according to the variations in the land. With agricultural practices based on these variations, savings are created for many inputs, while at the same time agricultural lands are protected against degradation. Precision agriculture practices based on environmental protection and optimum yield should be introduced to farmers and made widespread.

References

- Kaplan, F., 2020. toprak özelliklerinin tahmininde jeoistatistiksel ve spektroradyometrik yöntemlerin karşılaştırılması, harran üniversitesi, fen bilimleri enstitüsü, yüksek lisans tezi, toprak bilimi ve bitki besleme anabilim dalı, 1-63.
- Webster, R., Oliver, M.A., 2007. Geostatistics for Environmental Scientisits. Second Edition. John Wiley and Sons Limited, Chichester, England.
- Cambardella, C.A., Moorman, T.B., Parkin, T.B., Karlen, D.L., Novak, J.M., Turco, R.F., Konopka, A.E., 1994. Field-scale variability of soil properties in central Iowa soils. Soil Science Society of America Journal, 58(5): 1501-1511.
- Mali, S.S., Naik, S.K., Bhatt, B.P., 2016. Spatial variability in soil properties of Mango Orchards in Eastern Plateau and Hill Region of India. Vegetos-An International Journal of Plant Research, 29(3): 74-79.
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- Ege Üniv. Ziraat Fak. Derg., 2013, 50 (2): 181-190 ISSN 1018 8851 Kimyasal Toprak Kalite Göstergelerinin Faktör ve Jeoistatistik Analiz Yöntemleriyle Değerlendirilmesi The evaluating with factor and geostatistics analyses methods of soil chemical quality indices Alınış (Received): 18.01.2013 Kabul tarihi (Accepted): 29.03.2013 Mustafa SAĞLAM Orhan DENGİZ Ondokuz Mayıs Üniversitesi Ziraat Fakültesi, Toprak Bilimi ve Bitki Besleme Bölümü, 55139, Samsun. eposta: mustafa.saglam@omu.edu.tr Araştırma Makalesi (Research Article)
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- (1)https://www.google.com/url?sa=i&url=https%3A%2F%2Fjeogenc.net%2F madencilikte-jeoistatistik-nedir-neden

jeoistatistik.html&psig=AOvVaw1dNz00q9N8jC944A249qA2&ust=17

02458790282000&source=images&cd=vfe&opi=89978449&ved=0CB AQjRxqFwoTCOis783HiYMDFQAAAAAdAAAABAD

(2)https://www.google.com/url?sa=i&url=https%3A%2F%2Fwww.mdpi.com %2F2071-

1050%2F15%2F15%2F11842&psig=AOvVaw1njEbBdurMxydN7w9 HgAsU&ust=1702462411588000&source=images&cd=vfe&opi=8997 8449&ved=0CA8QjRxqFwoTCKCZt_XUiYMDFQAAAAAAAAAA BB5

- Ağca N (2015) Spatial Distribution of Available Heavy Metal Contents in Soils around an Industrial Area in Southern Turkey. Arab. J. Geosci., 8 : 1111– 1123.
- Osman A, 2020. Estimation and mapping of the properties of soils formed on basaltic parent material with different geostatistical methods. Harran university, institute of sciences, master's thesis, department of soil science and plant nutrition, Şanlıurfa.
- Kaplan F, 2020. Comparison of geostatistical and spectroradiometric methods in the estimation of soil properties, Harran University, Institute of Science, master's thesis, soil science and plant nutrition department, 1-63.
- Yetgin B (2004) Toprak fiziksel özelliklerinin uzaysal değişkenliğinin jeoistatistik yöntemlerle analizi. Yüksek Lisans Tezi, Gaziosmanpaşa Üniversitesi Fen Bil. Ens., Toprak ABD, 94 s.

CHAPTER 6

NITRATE TOXICATION IN RUMINANTS

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INTRODUCTION

Nitrate can be found in forage crops and water. It can reach levels that cause toxic effects in some plants. Plants used in animal nutrition with high nitrate levels are oats, beets, Soybean, Sunflower, Barley, Sweet clover, Rye, potato, sudangrass, Wheat, Corn(Hall, 2018) triticale, and carrots. In addition, susceptibility to nitrate toxicity varies in different animal species and physiological periods. In this section, detailed information will be given about the susceptibility to nitrate toxicity in different animal species and physiological periods, the mechanism of nitrate toxicity, clinical symptoms, diagnosis, and treatment.

1. Susceptibility of animals to nitrate intoxication

Animals' susceptibility to nitrate toxicity varies depending on their species and physiological periods.

1.1 Susceptibility of animals to nitrate intoxication by species

Susceptibility to nitrate toxicity in farm animals varies among species. Ruminant animals such as cattle, sheep, and goats, and monogastric animals such as horses and pigs are more sensitive to nitrate toxicity than other species(Bruning-Fann & Kaneene, 1993; Oruç & Ceylan, 2001a). The susceptibility of these animal species to nitrate toxicity also differs from each other. The animals most sensitive to nitrate toxicity are ruminants. The reason for this situation is that nitrate is reduced to toxic nitrite as a result of fermentation in the rumen of ruminants. Monogastric animals are less susceptible to nitrate toxicity compared to ruminants (Bruning-Fann & Kaneene, 1993). The reason for this situation is the absence of rumen in monogastric animals. Under normal conditions, nitrate cannot be converted to nitrite by microbial fermentation in the stomachs of monogastric animals. However, when the pH of the stomach increases and a suitable environment is created for microorganisms to live, nitrate can be converted to nitrite and nitrate toxicity may occur. This situation is mostly seen in newborn animals and humans whose stomachs cannot function fully. Methemoglobinemia, which occurs as a result of nitrate toxicity in humans, is called blue baby syndrome(Beyhun & Güler, 2002).

While monogastric animals and pre ruminant calves excrete nitrate from the body through urine, adult ruminants excrete a very small portion of nitrate through urine(McGuirk & Semrad, 2005). In adult ruminants, nitrate in the blood returns to the rumen through saliva and is metabolized in the rumen(Bolan & Kemp, 2003). In mono-gastric animals, especially horses and rabbits, nitrate is converted to nitrite due to microbial fermentation in the large intestines. However, this process is not as extensive as in ruminants, and absorption from the large intestine is at low levels. For this reason, nitrate toxicity in horses is rarely reported.(Hintz & Thompson, 1998; Oruc et al., 2010). In other mono-gastric animals, nitrate toxicity occurs mostly due to the consumption of foods with high nitrite levels that are not of plant origin(Worth et al., 1997). Susceptibility to nitrate toxicity varies among ruminants. Goats consume mostly the leaves of plants. The nitrate level in the leaves of plants is lower than in the roots and stems. For this reason, nitrate toxicity in goats is rare compared to other ruminants. Since sheep also eat the stem of the plant, their risk of toxicity is higher than goats. Sheep are less susceptible to nitrate toxicity compared to cattle. The main reason for this is that cattle consume large amounts of feed in a short time compared to sheep(Hall, 2018). As a result of cattle consuming large amounts of feed in a short time, a large amount of nitrate is suddenly taken into the rumen and the risk of toxicity will increase. In a study, the same amount of TMR was given to sheep as 1, 2, and 8 meals, and blood methemoglobin levels were measured. While the methemoglobin level in sheep fed 8 meals was around 2-3% throughout the day, the methemoglobin level increased up to 20-25% after feeding hours in 2-meal feeding, and in 1-meal feeding, the methemoglobin level increased to around 25-30% in the hours following feeding(Nolan et al., 2016). As can be seen from this study, consuming large amounts of feed in one meal increases the risk of toxicity.

1.2 Susceptibility of animals to nitrate intoxication according to physiological periods

Depending on the physiological periods of animals, their susceptibility to nitrate toxicity varies. The fetus has a high sensitivity to nitrate toxicity. Since the stomach in newborns does not function fully, the pH of the stomach is above normal. Microorganisms can live in this environment and convert nitrate to nitrite. This may cause nitrate toxicity. Susceptibility to nitrate toxicity decreases with age. Susceptibility to nitrate toxicity is increased in pregnant animals compared to non-pregnant animals(SULAK & AYDIN, 2005).

2. Mechanism of nitrate intoxication in ruminants

The main cause of nitrate toxicity is the ingestion of excessive amounts of nitrate or nitrite without an adaptation period. In ruminants, this situation is generally seen as a result of consuming large amounts of nitrate-rich forages or pastures without acclimatization, drinking nitrate-contaminated water, or consuming nitrate fertilizers. When nitrate taken in large amounts reaches the rumen, it is quickly reduced by bacteria to the more toxic nitrite by the enzyme nitrate reductase(Hall, 2018). Nitrite is very slowly reduced to ammonia using the enzyme nitrite reductase. The main reason for the rapid reduction of nitrate to nitrite and the slow reduction of nitrite to ammonia is the difference in the number of enzymes in the rumen. In a study conducted by Allison and Reddy[(Allison & Reddy, 1984), before getting used to nitrate diets, the amount of nitrate reductase in the rumen was 25 mmol/ml, while the amount of nitrite reductase was 4.5 mmol/ml. As can be understood from this, in animals that are not adapted to nitrate rations, the amount of nitrate reductase enzyme is higher than the amount of nitrite reductase enzyme, nitrite accumulates in the rumen and is absorbed through the rumen wall and passes into the blood. In the blood,

nitrate and nitrite serve as precursors of nitric oxide. However, excessive amounts of nitrite oxidize the +2-valent iron (ferrous (Fe⁺²)) in the structure of hemoglobin in the erythrocyte and turn it into +3-valent iron (ferric (Fe⁺³)). This condition is called Methemoglobinemia. In the case of methemoglobinemia, hemoglobin's affinity for oxygen decreases and it cannot carry oxygen. Tissues remain deprived of oxygen(Darling & Roughton, 1942). So, cyanosis occurs. Methemoglobin is converted to hemoglobin in erythrocytes in two ways. The first pathway is nicotine adenine dinucleotide-dependent methemoglobin reductase (NADH-Meth reductase), also known as cytochrome-b5 reductase(Anderson et al., 1988). The second pathway is NADPH-meth reductase. While the first pathway can continue physiologically, the second pathway requires either a cofactor or an electron acceptor such as methylene blue to work(Cortazzo & Lichtman, 2014). Methemoglobin concentration in the blood peaks approximately 2.5 hours after feeding (Nolan et al., 2016). In a study by Van't Klooster et al. (van't Klooster et al., 1990), methemoglobin reached its peak concentration 1.5 hours after intravenous nitrite administration. In a study by Schneider Yeary(Schneider & Yeary, 1975), he found that the half-life of Methemoglobin in sheep was 1.5 hours. Methemoglobin level in the body is considered normal if it is less than 3% (Eder et al., 1949). In a study by Oruç and Ceylan(Oruç & Ceylan, 2001a), the average methemoglobin concentration in cattle was found to be $2.64\pm0.21\%$. When the methemoglobin level reaches 30-40%, clinical symptoms begin to appear. When it reaches around 70-80%, death occurs(Burrows, 1980). Nitrate and nitrite are not the only reason for methemoglobin formation. Studies have reported that some local anesthetics such as lidocaine and benzocaine(Cortazzo Lichtman, 2014; Spielman et al., 1984), antibiotics such as & sulfamethoxazole, antineoplastic agents such as flutamide, drugs used in the treatment of malaria such as primaquine, and industrial products such as Amino phenol cause methemoglobinemia(Cortazzo & Lichtman, 2014). Nitrate

toxicity in ruminant feeding generally results from roughage, pasture, or water. While the nitrate level in water is desired to be below 100 (Puls, 1994). it has a toxic effect on ruminants at 1000 ppm and above(Hall, 2018). Nitrate in water is more toxic than in roughage. The main reason for this is that the nitrate in water turns into nitrite much faster than roughage. While a nitrate level of less than 0.5% in DM is considered safe in roughage, it has been reported that 1% and above nitrate in DM has a lethal effect on ruminants. However, in some publications, it has been reported that nitrate content of 2260 ppm and above in roughage is toxic(Oruç & Ceylan, 2001b). Pregnant animals should not be given roughage containing 2000 ppm or more nitrate. Even if feeds containing lower amounts of nitrate are given to animals that have been starved for a long time, toxicity may occur(Hall, 2018).

3. Examples of nitrate toxicity due to different causes

In 1986, it was reported that 18 heifers fed with wild hay containing 6% potassium nitrate died(Howard, 1993). In 1988, 16 beef cattle were found dead in a pasture in Canada. As a result of the investigations, it was found that the waters were contaminated with nitrate fertilizers.(Yong et al., 1990). 6 of the cattle fed with oat hay died shortly after feeding. In the analysis, it was found that oat hay contains 18000 ppm nitrate and 15500 ppm nitrite(Smith, 1991). On a farm in the USA, 3 of the dry cows fed hay in the evening died in the morning. In the examinations, it was found that there was 31000 ppm nitrate in the straw(Brown et al., 1990). It has been reported that amaurosis occurred in the calves of cattle fed with feeds with high nitrate levels in Isparta(ŞANLI, 1983). Four adult alpacas fed oat straw died because of nitrate poisoning. In the analysis, it was found that oat straw contained 1950 ppm nitrate(McKenzie et al., 2009). Three cattle died after consuming the plant called Chenopodium album, also known as goosefoot. In the analysis, it was found that the goosefoot plant contains 2500 ppm nitrate nitrogen(Ozmen et al., 2003). 9 horses died

between February and April on a horse farm in Bursa. Research has found that alfalfa hay and some forages contain high levels of nitrate(Oruc et al., 2010).

4. Clinical symptoms in nitrate toxicity

As a result of nitrate toxicity, methemoglobinemia occurs and oxygen cannot be transported to the tissues. Symptoms also occur due to this. Symptoms worsen when methemoglobin concentration increases. Under normal conditions, methemoglobin concentration is around 2%(Oruç & Ceylan, 2001b). Clinical symptoms are not obvious when the methemoglobin concentration is below 10%. At low doses, there is a decrease in efficiency without clinical symptoms, and when the methemoglobin concentration increases, cyanosis occurs due to the inability to carry oxygen to the cells, and a blue-violet discoloration of the skin occurs. In addition, there is an increase in breathing, increased heart rate, and tremors, and when the severity of toxicity increases, pregnant animals experience abortion(Hall, 2018) When the methemoglobin concentration reaches 80%, death may occur(Lee & Beauchemin, 2014). Although not always seen, increased salivation, vomiting, and diarrhea may occur due to nitrate toxicity(Robson, 2007).

5. Diagnosis

There are no specific symptoms of nitrate toxicity. It can be confused with many diseases. Acute nitrate toxicity: It can be confused with hypomagnesemia, ammonia toxicity, cyanide poisoning, insecticide, and herbicide toxicity. Darkening of the blood due to methemoglobinemia can be confused with septicemia(Hall, 2018). For definitive diagnosis, nitrate analysis should be performed in the feed and water of living animals, and in addition, blood methemoglobin concentration should be measured. To diagnose nitrate toxicity in dead animals, nitrate analysis should be done in feed and water, and additionally, nitrate analysis should be done by taking samples from the ocular fluid the best place to sample for nitrate analysis in deceased animals is the ocular fluid. Because nitrate in the ocular fluid can remain stable for a day at room temperature and can remain stable for a week at +4°C(Boermans, 1990). It is not recommended to take and analyze the rumen content of dead animals to diagnose nitrate toxicity. Because even though the animal is dead, fermentation in the rumen continues for a while and the nitrate and nitrite concentrations change. For diagnosis, blood and plasma samples can be taken, and nitrate analysis can be performed. However, the recommended location is the ocular fluid. Nitrate concentration in ocular fluid is 35% less than serum nitrate concentration(Boermans, 1990). Diphenylamine blue test is commonly used to diagnose nitrate in samples taken(Burrows, 1980). In an animal without nitrate toxicity, the ocular fluid nitrate concentration is less than 5 ppm. Nitrate concentration in the ocular fluid is between 10-20 ppm, indicating excessive nitrate exposure. If the concentration of nitrate in the ocular fluid is more than 20 ppm, it is considered diagnostic of poisoning(Burrows & Tyrl, 2001). In cases of abortion and stillbirth, the interpretation of the sample taken from the ocular fluid of the offspring is different. Because the normal nitrate concentration in fetal ocular fluid is around 20 ppm. To be diagnosed with nitrate toxicity, there must be 30 ppm or more nitrate in the fetal ocular fluid(Hall, 2018; Johnson et al., 1994). It is not realistic to perform nitrite analysis in samples taken from living tissues to diagnose nitrate toxicity. This is because nitrite has a short half-life(Hall, 2018).

6. Treatment

Treatment should first begin by placing the sick animal in a stress-free environment. 1-2% methylene blue solution should be calculated as 4-15 mg/kg per animal body weight and given intravenously. Practically, 2% methylene blue solution can be administered intravenously as 20 ml per hundred kilograms of live weight. In severe cases, treatment can be repeated with low doses. The half-life of methylene blue is 90 minutes. However, it accumulates in some tissues. In treated animals, tissues and urine turn dark green. Treated animals must wait at least 180 days for slaughter. Tolonium chloride can be used as an alternative to methylene blue for treatment. However, the therapeutic index of tolonium chloride is narrow.

6.1 Use of vitamin A, vitamin C, and selenium for therapeutic purposes

In nitrate and nitrite toxicity, methemoglobinemia occurs and oxygen transport to the tissues is prevented. Methylene blue is used for treatment purposes(Ginimuge & Jyothi, 2010). However, in studies conducted on humans, it has been reported that the use of methylene blue may be dangerous in individuals deficient in the glucose 6 phosphate dehydrogenase enzyme(Gregg & Prchal, 2018). Antioxidants and vitamins can be tried as an alternative treatment(Mansouri & Lurie, 1993). Vitamin C has been reported to levels(Prchal Gregg, reduce methemoglobin & 2005). However, methemoglobin levels may increase depending on the dose of vitamin In an in vitro study on alternative treatments to nitrate toxicity in ruminants, it was reported that vitamin A and vitamin B1 had no effect, but vitamin C and vitamin E reduced methemoglobin levels(Atyabi et al., 2012). It has been reported that there may be an increase or decrease in methemoglobin levels in both vitamin E and vitamin C, depending on the application dose. It has been reported that there is a decrease in methemoglobin levels when vitamin C and vitamin E are used together(Atyabi et al., 2012). As a result, it has been understood that vitamin C can be used as an alternative treatment for nitrate toxicity. It has been reported that the optimum dose in ruminants under in vitro conditions is 5 mmol/L. However, in vivo studies need to be conducted before using it in treatment.

REFERENCES

- Allison, M., & Reddy, C. A. (1984). Adaptations of gastrointestinal bacteria in response to changes in dietary oxalate and nitrate.
- Anderson, S. T., Hajduczek, J., & Barker, S. J. (1988). Benzocaine-induced methemoglobinemia in an adult: accuracy of pulse oximetry with methemoglobinemia. Anesthesia & Analgesia, 67(11), 1099-1101.
- Atyabi, N., Yasini, S. P., Jalali, S. M., & Shaygan, H. (2012). Antioxidant effect of different vitamins on methemoglobin production: An in vitro study. Veterinary Research Forum,
- Beyhun, E., & Güler, Ç. (2002). İçme suyunda nitrat ve sağlık. Hacettepe Tıp Dergisi, 33(2), 99-102.
- Boermans, H. (1990). Diagnosis of nitrate toxicosis in cattle, using biological fluids and a rapid ion chromatographic method. American journal of veterinary research, 51(3), 491-495.
- Bolan, N. S., & Kemp, P. (2003). A review of factors affecting and prevention of pasture-induced nitrate toxicity in grazing animals. Proceedings of the New Zealand Grassland Association,
- Brown, C., Burrows, G., & Edwards, W. (1990). Nitrate intoxication. Veterinary and human toxicology, 32(5), 481-482.
- Bruning-Fann, C. S., & Kaneene, J. (1993). The effects of nitrate, nitrite and Nnitroso compounds on human health: a review. Veterinary and human toxicology, 35(6), 521-538.
- Burrows, G. (1980). Nitrate intoxication. Journal of the American Veterinary Medical Association, 177(1), 82-83.
- Burrows, G., & Tyrl, R. (2001). Toxic plants of North America. Iowa State Univ. Press, Ames. Toxic plants of North America. Iowa State Univ. Press, Ames., -.

- Cortazzo, J. A., & Lichtman, A. D. (2014). Methemoglobinemia: a review and recommendations for management. Journal of cardiothoracic and vascular anesthesia, 28(4), 1043-1047.
- Darling, R. C., & Roughton, F. (1942). The effect of methemoglobin on the equilibrium between oxygen and hemoglobin. American Journal of Physiology-Legacy Content, 137(1), 56-68.
- Eder, H. A., Finch, C., & McKee, R. W. (1949). Congenital methemoglobinemia. A clinical and biochemical study of a case. The Journal of Clinical Investigation, 28(2), 265-272.
- Ginimuge, P. R., & Jyothi, S. (2010). Methylene blue: revisited. Journal of Anaesthesiology Clinical Pharmacology, 26(4), 517-520.
- Gregg, X. T., & Prchal, J. T. (2018). Red blood cell enzymopathies. In Hematology (pp. 616-625). Elsevier.
- Hall, J. O. (2018). Nitrate-and nitrite-accumulating plants. In Veterinary Toxicology (pp. 941-946). Elsevier.
- Hintz, H., & Thompson, L. (1998). Nitrate toxicosis in horses. Equine practice (USA).
- Howard, J. L. (1993). Current veterinary therapy 3: food animal practice. WB Saunders Company.
- Johnson, J., Grotelueschen, D., & Knott, M. (1994). Evaluation of bovine perinatal nitrate accumulation in western Nebraska. Veterinary and human toxicology, 36(5), 467-471.
- Lee, C., & Beauchemin, K. A. (2014). A review of feeding supplementary nitrate to ruminant animals: nitrate toxicity, methane emissions, and production performance. Canadian Journal of Animal Science, 94(4), 557-570.
- Mansouri, A., & Lurie, A. A. (1993). Methemoglobinemia. American journal of hematology, 42(1), 7-12.

- McGuirk, S. M., & Semrad, S. D. (2005). Toxicologic emergencies in cattle. Veterinary Clinics: Food Animal Practice, 21(3), 729-749.
- McKenzie, R., Gordon, A., Burren, B., Gibson, J., & Gardner, M. (2009). Alpaca plant poisonings: nitrate-nitrite and possible cyanide. Australian veterinary journal, 87(3), 113-115.
- Nolan, J. V., Godwin, I., de Raphélis-Soissan, V., & Hegarty, R. (2016). Managing the rumen to limit the incidence and severity of nitrite poisoning in nitrate-supplemented ruminants. Animal Production Science, 56(8), 1317-1329.
- Oruc, H. H., Akkoc, A., Uzunoglu, I., & Kennerman, E. (2010). Nitrate poisoning in horses associated with ingestion of forage and alfalfa. Journal of Equine Veterinary Science, 30(3), 159-162.
- Oruç, H. H., & Ceylan, S. (2001a). Bursa yöresinde sığırların yemlerinde, içme sularında ve rumen içeriğinde nitrat, nit-rit ve kanda methemoglobin düzeylerinin araştırılması. Uludağ Üniv. Vet. Fak. Dergisi, 20, 25-32.
- Oruç, H. H., & Ceylan, S. (2001b). Bursa yöresinde sığırların yemlerinde, içme sularında ve rumen içeriğinde nitrat, nitrit ve kanda methemoglobin düzeylerinin araştırılması. Uludağ Üniv. Vet. Fak. Dergisi, 20, 25-32.
- Ozmen, O., Mor, F., & Ayhan, U. (2003). Nitrate poisoning in cattle fed Chenopodium album hay. Veterinary and human toxicology, 45(2), 83-84.
- Prchal, J. T., & Gregg, X. T. (2005). Red cell enzymes. ASH Education Program Book, 2005(1), 19-23.
- Puls, R. (1994). Mineral levels in animal health. Diagnostic data. Sherpa international, Clearbrook. British Columbia, Canada, 48.
- Robson, S. (2007). Nitrate and nitrite poisoning in livestock. Profitable and Sustainable Primary Industries, 415, 1-4.

- Schneider, N., & Yeary, R. (1975). Nitrite and nitrate pharmacokinetics in the dog, sheep, and pony. American journal of veterinary research, 36(7), 941-947.
- Smith, R. (1991). Nitrite intoxication from large round bales. Veterinary and human toxicology.
- Spielman, F. J., Anderson, J. A., & Terry, W. C. (1984). Benzocaine-induced methemoglobinemia during general anesthesia. Journal of oral and maxillofacial surgery, 42(11), 740-743.
- SULAK, M., & AYDIN, İ. (2005). Yem bitkilerinde nitrat birikmesi. Anadolu Tarım Bilimleri Dergisi, 20(2), 106-109.
- ŞANLI, Y. (1983). ISPARTA YÖRESİNDE DOGMUŞ BUZAĞILARDA GÖRÜLEN AMOROZİS OLGULARı iLE GEBE İNEKLERDE KARŞıLAŞıLAN KRONİK NİTRAT ZEHİRLENMELERİ ARASINDAKİ ILIŞKİLERİN İNCELENMESİ. Ankara Üniversitesi Veteriner Fakültesi Dergisi, 30(04).
- van't Klooster, A. T., Taverne, M., Malestein, A., & Akkersdijk, E. (1990). On the pathogenesis of abortion in acute nitrite toxicosis of pregnant dairy cows. Theriogenology, 33(5), 1075-1089.
- Worth, A., Ainsworth, S., Brocklehurst, P., & Collet, M. (1997). Nitrite poisoning in cats and dogs fed a commercial pet food. New Zealand Veterinary Journal, 45(5), 193-195.
- Yong, C., Brandow, R. A., & Howlett, P. (1990). Saskatchewan. An unusual cause of nitrate poisoning in cattle. The Canadian Veterinary Journal, 31(2), 118.

CHAPTER 7

ASTRAGALUS GENUS and IT'S FORAGE POTENTIAL

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INTRODUCTION

The *Astragalus* genus has a narrative that permeates both human culture and the natural world; it is more than simply a chapter in a book on botany. "*Astragalus*" seems archaic, doesn't it? It means "ankle bone" in Greek (Castillon et al., 2023). It's kind of amusing, really, how these seeds gained their name—they resemble tiny bones. How do these seeds in their dried pods sound? similar to dice (Barneby, 1964). It begs the question of whether people in the past used them for a brief game of chance.

However, *Astragalus* is not simply a fascinating name. The Fabaceae is a large family with more than 3000 species (Frodin 2004). They may be found all over the world, including Europe and North America, the arid regions of Central Asia, and the temperate zones of the Northern Hemisphere (Castillon et al., 2023). They seem to have a talent for appearing almost anyplace, showcasing their adaptability and toughness (Amiri et al., 2020).

These plants assume a variety of shapes and forms that are both a botanist's nightmare and dream come true. Sorting them all out is a tremendous difficulty due to their variation in appearance. For years, taxonomists have been baffled by the many classification schemes that have been tried. They have divided them into groups, separated them into categories, and tried everything. Then, a whole new level of mystery was added when molecular studies emerged and completely caught everyone off guard (Zarre and Azani, 2013).

It's not all science, however, *Astragalus* is deeply ingrained in both the economy and culture. They have been used historically for a variety of purposes, including food, medicinal, and even ornamentation (Teng et al., 2011). Not to be overlooked is gum tragacanth, a true gem derived from these plants. Iran is the biggest supplier and is essentially the place to go for it (Anderson, 1998).

The knowledge about *Astragalus* is a little disorganized despite all the tales and applications. There is still so much to discover, particularly in the

areas of pharmacology and phytochemistry (Zarshenas et al., 2013; Amiri et al., 2020). This reviews aims to address that by attempting to integrate all of these disparate pieces of information. The objective? to establish a strong base that will serve as a jumping off point for further research projects and maybe even some business endeavors.

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Discovering the world of *Astragalus* species is like to investigating the ultimate adapters inside the realm of plants. These hardy cookies really have a talent for thriving in semi-arid to desert environments. Imagine them prospering in areas where the annual rainfall gauge rarely reaches 300–400 mm, and enduring harsh frosts (Davis, 1970). They are sun seekers who enjoy the grandeur of sunny locations and have a preference for sandy or well-drained loam soils (Erkovan et al., 2015). However, they are not interested in just any soil; they are particularly drawn to the kind of thick, calcareous clay that are prevalent in the driest regions of the earth.

Moreover, these plants don't care what elevation they grow in. They can reach heights of up to 2500 meters and are perfectly content in deteriorated settings. They tend to thrive at elevations of 1500 to 2500 meters above sea level, where they may plant their roots in the calcareous clay soils that they like (Davis, 1970). They grow in pH ranges of 6.5 to 8.0, with neutral soils being most happiest (Vasquez et al., 2010). They are cool about soil pH moist soles? These plants are all about living a dry lifestyle, and they appear to really get going when they receive a little CO_2 boost, particularly outside in the sun (Vasquez et al., 2010).

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To put it technically, *Astragalus* species are heliophytes, or real sun lovers, yet they don't always have sunny days. If you put them in the shadow, they won't be the happy campers, and their population will decline in less sunny areas. It is also unrealistic to expect them to recover from their roots in the event of a calamity like a fire or a haircut (Koc et al., 1994; Erkovan et al., 2015).

FORAGE POTENTIAL and EVALUATION

We start our study of the *Astragalus* genus by going into a region where more than 3,000 species of small bushes and plants do well. This area is under the broad wing of the Fabaceae family. These plants, which are also called milkvetches, can be found in a wide range of places on many continents, from the green areas of North America to the historic areas of Europe and Asia (Castillon et al., 2023).

Astragalus has been used in traditional medicine for a long time because it is good for you in many ways, especially in the complex system of Chinese plant treatments. The genus is useful for more than just human health. It is also useful in rural fields because it feeds animals like the cows and ovines that live on our farms. This two-in-one function links the health and farming areas and shows how adaptable the genus is (Benchadi et al., 2013).

But there is a word of caution in *Astragalus*' story. Even though it has a lot of different kinds, some species have parts that are more or less dangerous, which could put animals that eat them in danger (Gökkuş et al., 2001). Because of this, people who take care of animals must be careful. To learn about the safety of the local *Astragalus* species and to protect their charges, they should talk to farm development agencies or plant experts.

Because this genus grows in grasslands, *Astragalus* stands out as a nutritional powerhouse. Its leaf contains a wide range of fiber and protein that grazing animals need. This abundance comes with a risk, though, because the group has some species that animals shouldn't eat because they are bad for their health. There is a paradox here that shows how important it is to correctly identify species and use skilled grazing methods, like rotational grazing, to help these plants grow again and protect the animals that eat them (Shu et al., 2010).

Astragalus species, which are good at living in dry and semi-arid places, are seen as models of resilience in places where water is scarce because they can handle drought. Some types of *Astragalus* also have a mutually beneficial connection with bacteria that fix nitrogen. These bacteria use the nodules on their roots as crucibles for nitrogen synthesis, which is good for the soil and the environment as a whole (Erkovan and Koc, 2022).

Although its worth comes mostly from its use in traditional medicine, *Astragalus* makes important contributions to the rural world. Some growers

grow *Astragalus* for its health benefits as well as its use as feed. *Astragalus* group, which has many different species, is where traditional medicine and herding come together. The need to be careful with its many health and nutritional benefits is matched by the fact that these plants can live in harmony with the places they live and the people they touch (McKenna et al., 2002).

There is a multitude of *Astragalus* species in their natural habitats. There are numerous species that are capable of withstanding extreme temperatures, droughts, and irregular soil throughout the world (Davis, 1970; Erkovan et al., 2015). Limited selections of these species meet the criteria for forage plants, and they can manifest in two forms: annual or perennial, with or without thorns. Animals as undesirable intruders regard the majority of *Astragalus* species found in the rangelands of our country. Animal grazing may be permitted on a number of plants found in our rangelands (Serin et al., 2005), contingent upon the condition of their secondary metabolites. As an illustration, among other comparable herbaceous plants, small ruminants (e.g., sheep, goats) favor *Astragalus angustifolius* Lam., A. *fraxinifolius* DC., A. *idea* Sirj., A. *onobrychis* L., A. *schizopterus* Boiss., A. *squalidus* Boiss. and Noe. (Syn. A. amoenus Fenzl.) and A. *trojanus* Stev. (Serin et al., 2005) (Figure 1).



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Figure 1: Some species of Astragalus with nutritional value in pastures.

Apart from the aforementioned species, certain prickly plants that are not fed are subjected to alternative treatment and are employed as animal fodder in regions of Anatolia characterized by harsh winters. *Astragalus angustifolius* Lam., A. *baibutensis* Bunge, A. *brachypterus* Fisch., A. *cadmicus* Boiss., A. *compactus* Lam., A. *mongholicus* Bunge (Syn. A. *microcephalus* Fisch.) and A. *propinquus* Schischkin. are species that are utilized more locally and regionally (Figure 2).

In specific regions, specific species of *Astragalus* that are harvested from the surface of the soil are processed into fodder and fed to livestock during the winter. In some regions, root-bearing plants are dug up, submerged in water for the winter, then harvested and fed to animals. Soaking the prickly leaves of the plant in water during the wintertime renders them suitable for animal feed (Kargıoğlu et al., 2008; Özüdoğru et al., 2011; Amiri et al., 2020). The leaflet at the tip of the leaf stalk is in the shape of a thorn. Therefore, it is difficult for animals to graze on it, but goats prefer it (Figure 2).



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https://antropocene.it/en/2023/01/09/astragalus-propinquus-2/

Figure 2. *Astragalus* species used in animal feeding with locally different applications.

Although the *Astragalus* genus contains approximately 3000 species, only *Astragalus cicer* is viable as a nutrition source (Açıkgöz, 2021). This perennial plant is suitable as sustenance due to its extended lifecycle, resistance to frigid temperatures, and absence of bloading. This plant occurs naturally between the Caucasus and southern Spain. Under ideal conditions—rich soil and a favorable climate—it could attain a height of three meters. One meter is the maximum depth of its roots. Clustering of plants is possible due to the production of rhizomes. Occasionally, the cluster may attain a dimension of 120 cm (Figure 3).



https://en.wikipedia.org/wiki/Astragalus_cicer#/media/File:Astragalus_cicer1.jpg

Figure 3. The species of Astragalus used as a forage plant.

The flattened, coarse stems sag toward the earth. Depending on the conditions, the dry matter yield can range from 500 to 1000 kg/ha, which is significantly lower than that of alfalfa. The average protein content is 20% and the rate of digestion is 70% (Açıkgöz, 2021). In addition to being a forage crop, it can be utilized in grazing systems because it does not induce bloating in animals. Due to the fact that the majority of its growth takes place during the summer, it is presently utilized more frequently in pasture regions. Despite this, it is a species that is not cultivated frequently in our country.

Astragalus species contain toxic compounds that can cause poisoning when consumed above a certain level. Therefore, consumption in its green state is not advisable. *Astragalus* species are among the plants that accumulate selenium, which promotes growth (Gökkuş and Saygı, 2004). While selenium is a necessary nutrient for *Astragalus* species, it can be toxic for animals that consume selenium-rich plants excessively (Altın et al., 2021). However, when dried, its toxicity diminishes (Gökkuş et al., 2011). Among the A. cicer, it is a species that does not exhibit toxic properties.

In conclusion, species of *Astragalus*, despite not being widely used as forage or fodder, are valuable for preventing erosion in areas at risk. However, some species can still be utilized by animals to a limited extent. These species are more important for ecosystem vitality, presence, and continuity. Therefore, they are species that need to be extensively studied for various purposes.

REFERENCES

- Açıkgöz, E. 2021. Yem bitkileri. Tarım ve Orman Bakanlığı, Bitkisel Üretim Genel Müdürlüğü, Ankara.
- Altın, M., Gökkuş, A., Koç, A. 2021. Çayır mera Islahı. Palme Yayınevi, Ankara.
- Amiri, M.S., Jonarchi, M.R., Nadaf, M., Nasseh, Y. 2020. Etnobotancal knowledge of Astragalus spp.: The world's largest genus of vascular plants. Avicenna J. Phytomed, 10(2), 128-142.
- Anderson, D.M.W. 1989. Evidence for the safety of gum tragacanth (Asiatic Astragalus spp.) and modern criteria for the evaluation of food additives. Food Addit Contam, 6, 1-12.
- Barneby, R.C. 1964. Atlas of North American *Astragalus*. Memoirs of the New York Botanical Garden 13, 1–1188.
- Benchadi, W., Haba, H., Lavaud, C., Harakat, D., Benkhaled, M. 2013. Secondary metabolites of Astragalus cruciatus Link. and their hemotaxonomic significance. Records of Natural Products. 7(2), 105-113.
- Castillon, E.E., Quintanilla, J.A.V., Delgado-Salinas, A., Rebman, J.P. 2023.The genus Astragalus (Leguminosae: Papillonideae: Galegeae) in Mexico. Phytotaxa, 586(1), 1-162.
- Davis, P.H. 1970, Flora of Turkey and the East Aegean Islands. Edinburgh University Press, Edinburgh.
- Erkovan, S., Güllap, M.K., Erkovan, H.İ., Koç, A. 2015. A review of *Astragalus eriocephalus* Wild. synonym *Astracantha eriocephale* (Wild.) Podlech an invasive species for rangelands. Ecology & Safety, 9, 268-274.
- Erkovan, S., Koc, A. 2022. Competition-productivity relationship between some common grasses and forbs plant species in high altitude rangelands. Journal of Agricultural Sciences, 28(2), 287-295.

- Gökkuş, A., Koç, A., Comaklı, B. 2001. Çayır-mera uygulama kılavuzu. Atatürk Üniversitesi Ziraat Fakültesi Yayınları, Erzurum.
- Gökkuş, A., Saygı, Ç. 2004. Mera yönetiminde önemli bir bitki:Geven. Ziraat Mühendisliği, Temmuz-Aralık, 343: 8-11.
- Kargıoğlu, M., Cenkci, S., Serteser, A., Evliyaoğlu, N., Konuk, M., Kök, M.Ş., Bağcı, Y. 2008. An Ethnobotanical Survey of Inner-West Anatolia, Turkey. Hum Ecol. 36:763–777.
- Koç, A., Çomaklı, B. Gökkuş, A., Tahtacıoğlu, L. 1994. Azot ve fosforla gübreleme ile korumanın Güzelyurt Köyü (Erzurum) merasının bitki örtüsüne etkileri. Tarla Bitkileri Kongresi Çayır-Mera Yem Bitk. Bildirileri, İzmir, Türkiye, s. 78-82.
- McKenna, D.J., Hughes, K., Jones, K. 2002. Astragalus. Int J Integr Med. 4, 40-46.
- Özüdoğru, B., Akaydın, G., Erik, S., Yeşilada, E. 2011. Inferences from an ethnobotanical field expedition in the selected locations of Sivas and Yozgat provinces (Turkey). Journal of Ethnopharmacology, 137, 85–98.
- Serin, Y., Zengin, H., Tan, M., Koç, A., Erkovan, H.I., Avcıoğlu, R., Soya, H., Geren, H., Gemici, Y., Kendir, H., Sancak, C., Özaslan Parlak, A., Öztekin, M. 2005. Çayır ve mera bitkileri klavuzu. T.C. Tarım ve Köyişleri Bakanlığı Yayınları, Ankara.
- Shu, H.Q., Langran, X., Podlech, D. 2010. Astragalus L., sp.: Flora of China, 10, 328- 453.
- Teng, Y., Guo, H., Liang, Z., Shu, Z., Li, Z., Wu, W. 2011. Ethnobotanical survey of medicinal plants and their utilization in Shaanxi Province, China. J Med Plants Res, 5, 1762- 1778.
- Vasquez, E.A., James, J.J., Monaco, T.A., Cummings, D.C. 2010, Invasive plants on rangelands: a global threat. Rangelands, 32, 3-5.

- Zarre S, Azani, N. 2013. Perspectives in taxonomy and phylogeny of the genus *Astragalus* (Fabaceae): a review. PBioSci, 3, 1-6.
- Zarshenas, M.M., Arabzadeh, A., Tafti, M.A., Kordafshari, G., Zargaran, A., Mohagheghzadeh, A. 2013. Application of herbal exudates in traditional Persian medicine. Galen Med. Journal, 1, 78-83.

CHAPTER 8

ASTRAGALUS GENUS and IT'S FORAGE POTENTIAL

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INTRODUCTION

The *Astragalus* genus has a narrative that permeates both human culture and the natural world; it is more than simply a chapter in a book on botany. "*Astragalus*" seems archaic, doesn't it? It means "ankle bone" in Greek (Castillon et al., 2023). It's kind of amusing, really, how these seeds gained their name—they resemble tiny bones. How do these seeds in their dried pods sound? similar to dice (Barneby, 1964). It begs the question of whether people in the past used them for a brief game of chance.

However, *Astragalus* is not simply a fascinating name. The Fabaceae is a large family with more than 3000 species (Frodin 2004). They may be found all over the world, including Europe and North America, the arid regions of Central Asia, and the temperate zones of the Northern Hemisphere (Castillon et al., 2023). They seem to have a talent for appearing almost anyplace, showcasing their adaptability and toughness (Amiri et al., 2020).

These plants assume a variety of shapes and forms that are both a botanist's nightmare and dream come true. Sorting them all out is a tremendous difficulty due to their variation in appearance. For years, taxonomists have been baffled by the many classification schemes that have been tried. They have divided them into groups, separated them into categories, and tried everything. Then, a whole new level of mystery was added when molecular studies emerged and completely caught everyone off guard (Zarre and Azani, 2013).

It's not all science, however, *Astragalus* is deeply ingrained in both the economy and culture. They have been used historically for a variety of purposes, including food, medicinal, and even ornamentation (Teng et al., 2011). Not to be overlooked is gum tragacanth, a true gem derived from these plants. Iran is the biggest supplier and is essentially the place to go for it (Anderson, 1998).

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areas of pharmacology and phytochemistry (Zarshenas et al., 2013; Amiri et al., 2020). This reviews aims to address that by attempting to integrate all of these disparate pieces of information. The objective? to establish a strong base that will serve as a jumping off point for further research projects and maybe even some business endeavors.

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FORAGE POTENTIAL and EVALUATION

We start our study of the *Astragalus* genus by going into a region where more than 3,000 species of small bushes and plants do well. This area is under the broad wing of the Fabaceae family. These plants, which are also called milkvetches, can be found in a wide range of places on many continents, from the green areas of North America to the historic areas of Europe and Asia (Castillon et al., 2023).

Astragalus has been used in traditional medicine for a long time because it is good for you in many ways, especially in the complex system of Chinese plant treatments. The genus is useful for more than just human health. It is also useful in rural fields because it feeds animals like the cows and ovines that live on our farms. This two-in-one function links the health and farming areas and shows how adaptable the genus is (Benchadi et al., 2013).

But there is a word of caution in *Astragalus*' story. Even though it has a lot of different kinds, some species have parts that are more or less dangerous, which could put animals that eat them in danger (Gökkuş et al., 2001). Because of this, people who take care of animals must be careful. To learn about the safety of the local *Astragalus* species and to protect their charges, they should talk to farm development agencies or plant experts.

Because this genus grows in grasslands, *Astragalus* stands out as a nutritional powerhouse. Its leaf contains a wide range of fiber and protein that grazing animals need. This abundance comes with a risk, though, because the group has some species that animals shouldn't eat because they are bad for their health. There is a paradox here that shows how important it is to correctly identify species and use skilled grazing methods, like rotational grazing, to help these plants grow again and protect the animals that eat them (Shu et al., 2010).

Astragalus species, which are good at living in dry and semi-arid places, are seen as models of resilience in places where water is scarce because they can handle drought. Some types of *Astragalus* also have a mutually beneficial connection with bacteria that fix nitrogen. These bacteria use the nodules on their roots as crucibles for nitrogen synthesis, which is good for the soil and the environment as a whole (Erkovan and Koc, 2022).

Although its worth comes mostly from its use in traditional medicine, *Astragalus* makes important contributions to the rural world. Some growers

grow *Astragalus* for its health benefits as well as its use as feed. *Astragalus* group, which has many different species, is where traditional medicine and herding come together. The need to be careful with its many health and nutritional benefits is matched by the fact that these plants can live in harmony with the places they live and the people they touch (McKenna et al., 2002).

There is a multitude of *Astragalus* species in their natural habitats. There are numerous species that are capable of withstanding extreme temperatures, droughts, and irregular soil throughout the world (Davis, 1970; Erkovan et al., 2015). Limited selections of these species meet the criteria for forage plants, and they can manifest in two forms: annual or perennial, with or without thorns. Animals as undesirable intruders regard the majority of *Astragalus* species found in the rangelands of our country. Animal grazing may be permitted on a number of plants found in our rangelands (Serin et al., 2005), contingent upon the condition of their secondary metabolites. As an illustration, among other comparable herbaceous plants, small ruminants (e.g., sheep, goats) favor *Astragalus angustifolius* Lam., A. *fraxinifolius* DC., A. *idea* Sirj., A. *onobrychis* L., A. *schizopterus* Boiss., A. *squalidus* Boiss. and Noe. (Syn. A. amoenus Fenzl.) and A. *trojanus* Stev. (Serin et al., 2005) (Figure 1).



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https://botany.cz/cs/astragalus-amoenus/



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Figure 1: Some species of Astragalus with nutritional value in pastures.

Apart from the aforementioned species, certain prickly plants that are not fed are subjected to alternative treatment and are employed as animal fodder in regions of Anatolia characterized by harsh winters. *Astragalus angustifolius* Lam., A. *baibutensis* Bunge, A. *brachypterus* Fisch., A. *cadmicus* Boiss., A. *compactus* Lam., A. *mongholicus* Bunge (Syn. A. *microcephalus* Fisch.) and A. *propinquus* Schischkin. are species that are utilized more locally and regionally (Figure 2).

In specific regions, specific species of *Astragalus* that are harvested from the surface of the soil are processed into fodder and fed to livestock during the winter. In some regions, root-bearing plants are dug up, submerged in water for the winter, then harvested and fed to animals. Soaking the prickly leaves of the plant in water during the wintertime renders them suitable for animal feed (Kargıoğlu et al., 2008; Özüdoğru et al., 2011; Amiri et al., 2020). The leaflet at the tip of the leaf stalk is in the shape of a thorn. Therefore, it is difficult for animals to graze on it, but goats prefer it (Figure 2).



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https://twitter.com/antalyafestival/status/1303299523104694273



http://www.vanherbaryum.yyu.edu.tr/flora/famgenustur/fab/astragalus/comp/index.ht m



https://m.blog.naver.com/PostView.naver?isHttpsRedirect=true&blogId=jdchol&log No=220116554088&view=img_6



https://antropocene.it/en/2023/01/09/astragalus-propinquus-2/ Figure 2. *Astragalus* species used in animal feeding with locally different applications.

Although the *Astragalus* genus contains approximately 3000 species, only *Astragalus cicer* is viable as a nutrition source (Açıkgöz, 2021). This perennial plant is suitable as sustenance due to its extended lifecycle, resistance to frigid temperatures, and absence of bloading. This plant occurs naturally between the Caucasus and southern Spain. Under ideal conditions—rich soil and a favorable climate—it could attain a height of three meters. One meter is the maximum depth of its roots. Clustering of plants is possible due to the production of rhizomes. Occasionally, the cluster may attain a dimension of 120 cm (Figure 3).



https://en.wikipedia.org/wiki/Astragalus_cicer#/media/File:Astragalus_cicer1.jpg

Figure 3. The species of Astragalus used as a forage plant.

The flattened, coarse stems sag toward the earth. Depending on the conditions, the dry matter yield can range from 500 to 1000 kg/ha, which is significantly lower than that of alfalfa. The average protein content is 20% and the rate of digestion is 70% (Açıkgöz, 2021). In addition to being a forage crop, it can be utilized in grazing systems because it does not induce bloating in animals. Due to the fact that the majority of its growth takes place during the summer, it is presently utilized more frequently in pasture regions. Despite this, it is a species that is not cultivated frequently in our country.

Astragalus species contain toxic compounds that can cause poisoning when consumed above a certain level. Therefore, consumption in its green state is not advisable. *Astragalus* species are among the plants that accumulate selenium, which promotes growth (Gökkuş and Saygı, 2004). While selenium is a necessary nutrient for *Astragalus* species, it can be toxic for animals that consume selenium-rich plants excessively (Altın et al., 2021). However, when dried, its toxicity diminishes (Gökkuş et al., 2011). Among the A. cicer, it is a species that does not exhibit toxic properties.

In conclusion, species of *Astragalus*, despite not being widely used as forage or fodder, are valuable for preventing erosion in areas at risk. However, some species can still be utilized by animals to a limited extent. These species are more important for ecosystem vitality, presence, and continuity. Therefore, they are species that need to be extensively studied for various purposes.

REFERENCES

- Açıkgöz, E. 2021. Yem bitkileri. Tarım ve Orman Bakanlığı, Bitkisel Üretim Genel Müdürlüğü, Ankara.
- Altın, M., Gökkuş, A., Koç, A. 2021. Çayır mera Islahı. Palme Yayınevi, Ankara.
- Amiri, M.S., Jonarchi, M.R., Nadaf, M., Nasseh, Y. 2020. Etnobotancal knowledge of *Astragalus* spp.: The world's largest genus of vascular plants. Avicenna J. Phytomed, 10(2), 128-142.
- Anderson, D.M.W. 1989. Evidence for the safety of gum tragacanth (Asiatic *Astragalus* spp.) and modern criteria for the evaluation of food additives.Food Addit Contam, 6, 1-12.
- Barneby, R.C. 1964. Atlas of North American Astragalus. Memoirs of the New York Botanical Garden 13, 1–1188.
- Benchadi, W., Haba, H., Lavaud, C., Harakat, D., Benkhaled, M. 2013. Secondary metabolites of Astragalus cruciatus Link. and their hemotaxonomic significance. Records of Natural Products. 7(2), 105-113.
- Castillon, E.E., Quintanilla, J.A.V., Delgado-Salinas, A., Rebman, J.P. 2023.The genus *Astragalus* (Leguminosae: Papillonideae: Galegeae) in Mexico. Phytotaxa, 586(1), 1-162.
- Davis, P.H. 1970, Flora of Turkey and the East Aegean Islands. Edinburgh University Press, Edinburgh.
- Erkovan, S., Güllap, M.K., Erkovan, H.İ., Koç, A. 2015. A review of *Astragalus eriocephalus* Wild. synonym *Astracantha eriocephale* (Wild.) Podlech an invasive species for rangelands. Ecology & Safety, 9, 268-274.
- Erkovan, S., Koc, A. 2022. Competition-productivity relationship between some common grasses and forbs plant species in high altitude rangelands. Journal of Agricultural Sciences, 28(2), 287-295.

- Gökkuş, A., Koç, A., Comaklı, B. 2001. Çayır-mera uygulama kılavuzu. Atatürk Üniversitesi Ziraat Fakültesi Yayınları, Erzurum.
- Gökkuş, A., Saygı, Ç. 2004. Mera yönetiminde önemli bir bitki:Geven. Ziraat Mühendisliği, Temmuz-Aralık, 343: 8-11.
- Kargıoğlu, M., Cenkci, S., Serteser, A., Evliyaoğlu, N., Konuk, M., Kök, M.Ş., Bağcı, Y. 2008. An Ethnobotanical Survey of Inner-West Anatolia, Turkey. Hum Ecol. 36:763–777.
- Koç, A., Çomaklı, B. Gökkuş, A., Tahtacıoğlu, L. 1994. Azot ve fosforla gübreleme ile korumanın Güzelyurt Köyü (Erzurum) merasının bitki örtüsüne etkileri. Tarla Bitkileri Kongresi Çayır-Mera Yem Bitk. Bildirileri, İzmir, Türkiye, s. 78-82.
- McKenna, D.J., Hughes, K., Jones, K. 2002. Astragalus. Int J Integr Med. 4, 40-46.
- Özüdoğru, B., Akaydın, G., Erik, S., Yeşilada, E. 2011. Inferences from an ethnobotanical field expedition in the selected locations of Sivas and Yozgat provinces (Turkey). Journal of Ethnopharmacology, 137, 85–98.
- Serin, Y., Zengin, H., Tan, M., Koç, A., Erkovan, H.I., Avcıoğlu, R., Soya, H., Geren, H., Gemici, Y., Kendir, H., Sancak, C., Özaslan Parlak, A., Öztekin, M. 2005. Çayır ve mera bitkileri klavuzu. T.C. Tarım ve Köyişleri Bakanlığı Yayınları, Ankara.
- Shu, H.Q., Langran, X., Podlech, D. 2010. Astragalus L., sp.: Flora of China, 10, 328-453.
- Teng, Y., Guo, H., Liang, Z., Shu, Z., Li, Z., Wu, W. 2011. Ethnobotanical survey of medicinal plants and their utilization in Shaanxi Province, China. J Med Plants Res, 5, 1762-1778.
- Vasquez, E.A., James, J.J., Monaco, T.A., Cummings, D.C. 2010, Invasive plants on rangelands: a global threat. Rangelands, 32, 3-5.
- Zarre S, Azani, N. 2013. Perspectives in taxonomy and phylogeny of the genus *Astragalus* (Fabaceae): a review. PBioSci, 3, 1-6.

Zarshenas, M.M., Arabzadeh, A., Tafti, M.A., Kordafshari, G., Zargaran, A., Mohagheghzadeh, A. 2013. Application of herbal exudates in traditional Persian medicine. Galen Med. Journal, 1, 78-83.

CHAPTER 9

LAMB RAISING PROCESS AND ITS IMPORTANCE

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1. INTRODUCTION

They are ensuring that lambs survive after birth and develop until weaning age is very important for livestock farms that produce both milk and meat (Caroprese, 2008). In farms where sheep milk is sold at high prices, increasing the amount of milk marketed has made it necessary to artificially grow some milk (Cristobal Carballo et al. 2019). In sheep farms that produce milk and meat together to increase the profits of sheep-goat farms and to meet market and consumer-based expectations regarding animal welfare, keeping lambs whose mothers have died or whose mother's milk is not sufficient until the weaning or marketing age is a difficult and tiring task for the farm. In New Zealand, one of the countries where sheep farming is intensive, pasture-based sheep production systems offer a lower-cost production system to produce highquality food for human consumption (Mc et al. 2019). Globally, dairy sheep and many mutton production systems use highly intensive feed inputs, with flock sizes of less than five hundred head in most of these production systems. Therefore, widely applied artificial rearing systems have been developed to accommodate existing herd management systems, rather than where animals are grazed largely on pasture. The viability of the lamb is closely related to the condition of the ewe in different physiological periods as well as in the reproductive cycle. In herd management, it can be affected at different levels by the practical breeding work to be done on the animals (Binns et al. 2002). In other words, it is recommended that ewes with poor body condition be given additional feeding before mating, planned mating to prevent unnecessary harsh environmental conditions at birth, appropriate shelter with good hygiene during lambing, and monitoring and support of weak lambs. All of these are practices to increase the survival of lambs (Tibbo et al. 2001, 2003; Holmøy and Waage, 2015).

Reducing feed and labour costs without compromising the growth, health and welfare of lambs before and after weaning contributes significantly to the profitability of lamb-rearing systems. In many countries, research conducted by the public for this purpose is also supported and the results are put into practice (Everett-Hincks and Dodds, 2008). In countries where sheep meat is prominent, such as Australia and New Zealand, many programs are being carried out to develop and support dairy sheep farming (Fridlund et al. 2013). With the help of these programs, stakeholders are provided with information regarding production systems, various lamb-rearing systems using natural and artificial breeding, and the aforementioned methods. Lamb rearing programs affect the cost of raising female and male lambs and enable several different rearing systems to be implemented in commercial environments. For example, in New Zealand, due to the seasonal structure of the current production systems in sheep breeding, it is aimed to establish small or largescale breeding farms in which 3-4 lambs can be raised by one producer each year in a short time. This situation conflicts with some sheep production systems. There are also opportunities to practice early weaning in naturally reared lambs on a smaller pasture (>250 sheep) to increase commercial milk production before the lambs' postnatal survival and development decline (Fthenakis et al. 2012). In the studies to be carried out on this subject, the rapid resolution of possible problems encountered during the process of raising the offspring will increase the survival rate of the lambs and will reduce the costs of raising the offspring proportionally, albeit relatively. In this article; is to provide an overview of the best practice information available for successful lamb farming. Although the paper is especially based on dairy sheep production, the information produced will also be useful for producing sheep meat, especially for systems for raising lambs whose dams have died or are orphaned.

2. WHICH RAISING METHOD SHOULD BE USED IN THE SHEEP FARM?

In sheep farming, deciding what the production system should be is very difficult due to many factors. For example, decisions to be made for a newly established dairy sheep farm may affect the working system even in some works performed without an additional task, such as raising lambs. The breeding method to be applied is not an approach suitable for everyone, and sometimes the capital of the farm is not enough (McHugh et al. 2016). The economic pros and cons of cost, stress of the work, and risk also play an important role in the decision. Another important issue is that the physical infrastructure and workforce requirements of the farm should also be taken into consideration (Figure 1).



Figure 1. Points to consider when deciding on the lamb-rearing method Source: McCoard et al. 2019; Stevens et al. 2017

If the farm is based solely on milk production;

a. How much milk income will be lost if natural rearing is implemented? What will be the benefit: cost value in case of milk loss due to natural rearing and if this milk is sold commercially? Could switching to artificial rearing be more profitable for the farm?

b. What is the cost to the farm of giving up natural growth?

c. How long should the lambs stay with the mother? The current industry-rearing model for reducing teat damage is nearly 30 days, but the optimum duration in pasture-based systems has not yet been determined.

d. How meaningful is it to reduce density in a lamb breeding facility by marketing only single-born or all-male lambs to leading breeders? This approach is an option to reduce stress and/or alleviate the incidence of disease in farms.

e. How keen is the sheep producer to avoid the potential stress associated with raising lambs? There may also be breeders who want to give up dairy farming and engage in dairy sheep breeding and therefore have little experience.

f. Do lambs reared with their dams get a better start in life and have better lifelong performance? If so, how much importance is given to this?

g. What are the advantages and disadvantages of a mixed system, in which single and triplets are reared artificially, while twins and the rest are raised naturally and weaned after a suckling period of about 30 days? This practice reduces the number of lambs to be raised and ensures that fewer sheep are accustomed to the milking platform earlier.

If the farm is combined efficiency-oriented, it may be answers to some questions. These questions are respectively;

• What is the economic value of weaned lambs?

• Does the breeder have the capacity to graze and market the lambs, or do they need to be sold to the slaughterhouse?

• What is the cost of ewes that are milked early to avoid drying out after losing their lambs?

• Finally, how much does the consumer value natural farming?

Since there are great differences between the production systems of farms and consumer demands, there are different approaches to this issue. An example of the decision tree approach is given with the idea that it may help in deciding which production and breeding system to use on the farm (Figure 2). This study example may provide some information on how to decide on the right augmentation system for the purposes (Gebretensay et al. 2019).

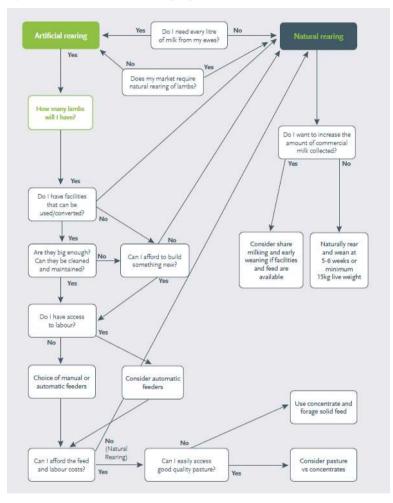


Figure 2. Decision tree to be applied in raising offspring Source: McCoard et al. 2020

2.1. Determination of Pregnancy

Lamb survival is affected by many factors such as farm conditions, breed, nutrition and herd management. For sustainable production, it is very important to obtain the lambs to the marketing age without mortality. In this case, nutrition is the most critical parameter to realize the potential. Pregnancy control is a valuable tool for nutritional management and it is useful to know some of the recommendations mentioned (Genfor et al. 2023). The first of these is that reformed or unproductive animals in the farm must be identified individually to reduce feed and labour inputs. To make annual feed planning, the total number of lambs and therefore the nutritional requirements and all the details of the feed planning, including the number of lambs carried by each ewe, should be determined. Nutrient requirements should be calculated according to the number of fetuses. During the last two months of pregnancy, nutrient requirements are 50% higher in triplet-bearing ewes and 20% higher in twinbearing ewes compared to single-bearing ewes. During the suckling period, the sheep nutritional requirements increase. Separation of birth groups and feeding according to the pregnancy period should be well planned to increase the pregnancy rate and when the sheep will give birth, to use feed resources strategically and efficiently, that is, to provide the right feed to the right animals at the appropriate time.

2.2. Body Condition Scoring

The body condition score is used to evaluate the effectiveness of the herd management system applied in the farm. In addition, scoring is easy and provides important information to the farm owner in making annual feed planning (Taşkın et al. 2005). The general condition of the body, a function of the muscle (red) and fat mass (yellow) of the animal (Figure 3), reflects the level of nutrition and welfare. Inadequate or unbalanced nutrition, physiological conditions and health problems negatively affect the body condition score of the sheep. As a result, the animal's welfare and health decline due to poor body condition scores.



Figure 3. Body condition scoreing and its application in sheep Source: McCoard et al. 2020; https://www.fas.scot/livestock/sheep/condition-scoring-for-sheep/

Efforts should be made to maintain good body condition scores in sheep throughout the year. In other words, the animal should have a body condition score ranging from three to four. Maintaining the nutritional requirements of the sheep and improving the use of feed at the right place, time and animal should be the basis. If live weight in sheep is not determined regularly and changes in live weight cannot be monitored individually over time, BCS is not a good indicator for feeding management in that farm. A small but fat sheep can have a similar live weight as a larger but thin sheep (Tozlu Çelik et al. 2023). Body condition scoring when sheep do not have a small body covering like wool is very risky due to the high margin of error. The best approach is to physically determine the body condition score through palpation. It should be done before mating to determine which ewes need additional feeding before mating (Taşkın, 2014). Low live weight sheep (condition score below 3) should be given additional feed and then their body condition should be re-evaluated. This is only worse when there is insufficient pasture to meet the nutrient requirements of the animals. To increase the body condition and survival of the lambs, a supplementary feed rich in grain will be beneficial.

Body condition scoring (BCS) should be performed to monitor live weight changes in animals and to identify sheep with poor body condition in advance. While this scoring is being done, a feeding plan should also be developed to maintain body condition. Loss of body condition in the late period of pregnancy (last 4-6 weeks) negatively affects the viability and pre-weaning growth rates of lambs, even in ewes in good condition. A one-unit change in body condition score (e.g., a decrease from 4 to 3) during the specified period of pregnancy can increase lamb mortality by 8% in singletons and twins, 15% in triplets, and 20% in quadruplets. In older sheep (> 5 years old) these effects are relatively greater. Therefore, regardless of the body condition of the sheep, the important thing is to develop a new feeding program to prevent further condition loss. Because even for sheep in good condition, what is dangerous is not only the condition score but also the loss of condition. If there are sheep with a condition score between 2-2.5, they should be helped to maintain or improve their condition, preferably by feeding them. To prevent condition losses, good care should be provided to sheep with a condition score of 3 and above (Bates et al. 2022). Since this is a welfare problem for sheep with a condition score of 1, the animals should be intervened and the necessary improvement should be made as soon as possible.

2.3. Pregnancy Nutrition and Herd Management in Sheep

The nutritional requirements of sheep means, in other words, that the animals start lactation with a better body condition after birth. Another important point is that good nutrition of the dams also means that the offspring have a high live weight and survival rate (Goddard et al. 2006). As pregnancy progresses, the mother's nutritional requirements increase, especially in the last six weeks before lambing (Figure 4). In this case, the feed given to the dams should never be reduced for the development of the fetus. A restriction in feeding during the last six weeks of pregnancy, especially in sheep giving birth to multiples, may negatively affect some of the following characteristics;

a. Fetal development: Since the development of offspring is not high in undernourished pregnant sheep, this results in low birth weight in the offspring.

b. **Viability in lambs**: This feature affects the time spent for birth, allows the offspring to stand up and suckle their mother as soon as possible, and delays the formation of the bond between sheep and offspring.

c. **Regulation of body temperature**: Since brown fat tissue is low due to malnutrition, especially in newborn lambs, the mechanism of regulating body temperature is not sufficient. In such a case, the lamb's chance of survival is only 50% with the special fatty tissue that accumulates under the skin during pregnancy.

d. Sheep colostrum and milk production: Inadequate nutrition of the dam during pregnancy negatively affects both the quantity and quality of colostrum.

e. As the number of offspring at birth increases, the nutritional requirements of the mothers also increase.

f.Animal health: Disruptions that may occur due to poor herd management in the farm should be prevented and all necessary vaccinations should be made before lambing.

g. When feeding based on pasture, sheep's relevant vaccinations and vitamin and trace mineral requirements must be met.

h. **Particular attention** should be paid to good nutrition and feeding during pregnancy and lactation period to ensure increased viability and growth of lambs as well as the continued reproductive performance of ewes.

k. Sheep should not be grazed in areas with vegetation of 1500 kg dry matter per hectare, especially in the last one or one and a half weeks of pregnancy, and the ration should contain 15-18% crude protein.

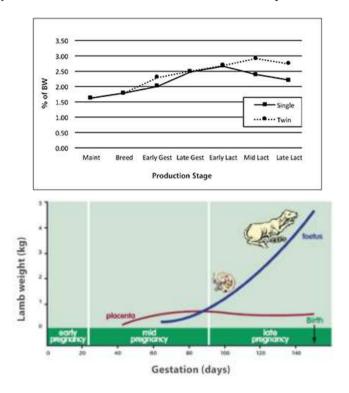


Figure 4. Changes in gestation period, birth type and nutritional needs in sheep Source: https://pubs.nmsu.edu/_circulars/CR685/index.html Source: https://www.rumenco.co.uk/sites/default/files/screenshot_2020-11-05 at 10.06.50.png

2.4. Regulation of Body Temperature in Lambs

Lamb deaths cause significant income loss to farms and ranches. Although lamb deaths occur anywhere from birth to weaning, the largest lamb deaths occur in the first few days of life. This ability to survive is multifaceted, depending on genetics, behaviour, physiology and environment, and is closely linked to the birth weight of lambs. Breers can help minimize lamb losses by optimizing the birth weights of lambs within flocks and breeds. In a sheep population, lamb birth weight to lamb survival is a U-shaped distribution, with the mean somewhere in the middle. Lambs that fall at either end of the Ushaped distribution, i.e. those with abnormally low or high birth weights, have a higher chance of death. Lambs with low birth weight may be exposed to more hunger and adverse environments due to their higher body surface/body mass ratio. In addition, since movement decreases in these lambs, the behaviour of searching for the sheep's udder is relatively insufficient. On the other hand, higher birth weights, in other words, dystocia, are not a very desirable event. This may result in birth injuries, decreased viability of the lamb at birth, and sometimes death. Although it increases the risk of dystocia in high birth weight lambs, multiple births in ewes with high fertility, abnormal birth arrivals in smaller lambs and longer birth time may be the primary problems.

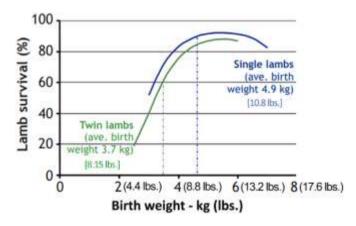


Figure 5. Variation of viability in lambs according to birth weight Source: https://www.zoetis.com.au/livestock-solutions/sheep/maximise-lambproductivity/lamb-survival.aspx

https://extension.sdstate.edu/lamb-birth-weights-relation-lamb-survivability

Normal body temperature in lambs is 39°C. A value of 37-39°C indicates mild to moderate hypothermia, and if the thermometer reads below 37°C, the lamb is said to be significantly hypothermic (McDowall et al. 2011).

Hypothermia treatment should vary depending on the age and body temperature of the affected lamb. Viability in lambs is related to rectal temperature at birth. This requires focusing on the relationship between sexual maturity, postnatal behaviour and body temperature regulation mechanisms (Jensen et al. 2017). It is used to experimentally induce mild hypothermia in lambs after birth, through a controlled water bath test. This test first determines the time it takes for the body temperature to reach 35°C and the time it takes for the body temperature to return to 39°C. Lambs that are slow to stand up and reach the udder for the first time after birth also have weakened resistance to cold (Plush, 2013). Some blood parameters and hormone levels do not correlate with the performance of lambs in a warm water bath used to prevent hypothermia. Older lambs experienced a delay in recovery from cold compared to lambs with lower vigour and maturity, as assessed by the speed at which they performed perinatal behaviours and physiological blood measurements. Although it has been suggested that this may be due to the decreased ability to regulate body temperature without muscle contraction in lambs in the following weeks, unlike newborns, many studies need to be conducted on this subject. In production systems in countries where sheep farming is extensive or pasture-based, most lambs may die within the first 3 days after birth (Everett-Hinks and Dodds, 2008). The most important cause of lamb deaths is hypothermia, especially in adverse weather conditions. A dangerous drop in body temperature that occurs when newborn puppies lose more body heat quickly than they can produce is called hypothermia. Many environmental reasons, such as the inability to produce colostrum in sheep, the mother not adopting her offspring, and low birth weight, negatively affect the viability of lambs (Koyuncu et al. 2018). To summarize, sheep regulate their body temperature in four different ways. These are respectively;

Wool: The thick fleece layer on the sheep's body acts as an insulating layer, keeping it warm in cold weather and helping to prevent heat loss from the body.

Sweat glands: Sheep have sweat glands in their skin that help cool the body by evaporating and removing excess heat.

Respiratory system: Sheep regulate their body temperature through the respiratory system by panting, which helps release heat from their bodies.

Behavior: Sheep do this through behaviours such as looking for shade or cool places to cool off in hot weather, and huddling together to maintain body temperature in cold weather.

2.5. Vitality in Lambs After Birth

In the first days following birth, the mortality rate in lambs can reach 15% and above under poor care and feeding conditions. This issue is not emphasized much by some breeders in sheep farming farms in some countries and regions. However, when the causes of lamb death are examined; It has been determined that factors such as dystocia, hunger and inability to find the mother play a major role (Swalha et al. 2007). Among the factors examined, when the relative effect of dystocia on offspring mortality is examined, high body condition score or excessive fatness in sheep, inappropriate rump width and depth, large or heavy lambs; large-bodied lambs; Weak uterine structure, resulting in lack of contraction power and insufficient amniotic fluid production. This problem can be minimized with herd management decisions and correct selection practices in this direction (Figure 6). In Merino lambs, the coat of hair that covers or covers the body at birth can vary from fine fibres and short, tight curls to a coarse coat dominated by long, coarse protruding halo hairs. It consists of primary fibres and some of the secondary fibres before birth and is replaced by a hair cover similar to that in pigs after birth, through the maturation of the remaining secondary fibres (Ponzoni et al. 1997). Olivier et al. (1994) stated that hairy lambs are practically culled to reduce fibre diameter and that the change in fibre diameter as age increases and the condition called coarse edge (fibre percentage over 30 μ m) (Cloete et al. 2003). It has been reported that hair cover shape at birth has a high degree of heritability, ranging from 0.65 to 0.70 (Schinckel, 1955; Olivier et al. 1994; Kemper et al. 2003). These researchers also reported that the relationship between wool and other properties was moderately positive and negative. Ponzoni et al. (1997) concluded that fleece fibre type at birth is limited as an early selection criterion in Australian Merino sheep, despite the high heritabilities reported in the literature. Kemper et al. (2003) stated that the decrease in fibre diameter would also lead to a decrease in the birth coat score in fine-wool Merinos (Table 1).

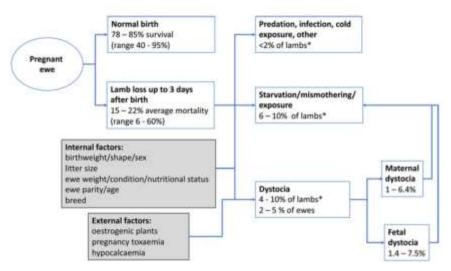


Figure 6. Dystocia in sheep and its causes

Source: Jakobson ve ark. 2020

Hair so	core	at	Explanation	
birth				
1	1		When the lamb is lifted into the air and held up to the light, the lower	
			hairs are not visible. Hair fibres are visible, group-shaped, and have a	
			short wavy structure.	
2	2		There are no underhairs even when the lamb is on the ground. When	
			the lamb is held up in the air and held up to the light, some under hairs	
			can be seen.	
3	3		The few underhairs can be seen when the lamb is on the ground. The	
			dominant hair in the lamb's appearance is short and wavy.	
4	1		The lower hairs are quite visible. It is short, wavy, distinct and easily	
			visible	
5	5		There are many lower bristles. Its short and wavy structure can be	
			noticed among the long hairs.	
6	5		No short hairs. Wavy fibres can be seen. There are only long straight	
			hairs	
7	7		There are no visible wavy hairs. There are only very long hairs. When	
			viewed from the outside, it resembles dog hair.	

Table 1. Scoring of the hair covering the body in newborn lambs

Source: McDowall et al. 2011

Pregnancy is an important period to contribute to the success of lambs in terms of postnatal viability. It is very important to know dystocia better externally or to be able to identify possible animals. For this aim, animals in the herd should be handled individually and treated accordingly (Bancheva et al. 2022). Although feeding is more important in sheep giving birth to twins, the metabolic status of the animals must be determined in case of limited feeding. For this reason, the period when the risk of lamb death is highest is the period close to birth, and therefore, it may be a more appropriate approach to focus on weaning success, which is even more affected by the environment. The features to be discussed by looking at the external or morphological structure of the animals are as follows; It should include some behavioral measurements such as live weight, body measurements, rectal temperature, blood sugar and latency of bleating in the offspring, as well as new measurements such as postnatal movement activity in lambs. Lamb deaths should be minimized with pre-and post-natal practices in the farm (Kandemir et al. 2015).

Lamb mortalities and their causes are briefly summarized below. These are respectively;

a. Not being able to find the mother after birth: Lambs lost during pregnancy and the ewes being left alone at lambing time also play an important role in this.

b. Loss of offspring occurring in the last months of pregnancy: This may be caused by bacterial or viral abortions.

c. **Stillbirths:** Lambs that are stillborn despite completing the gestation period are included in this group.

d. **Deaths occurring in the first 48 hours**: Although lambs are born alive, the babies may die due to poor birth chambers, lack of umbilical cord cleaning, hunger and other factors.

e. **Deaths occurring within two days and later**: Colostrum and its quality, as well as the growth management applied, have an impact on this rate.

The causes of lamb deaths are given in Figure 7.

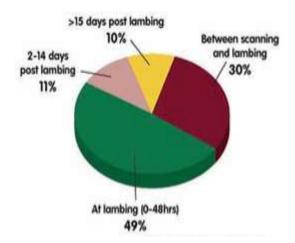


Figure 7. Causes of lamb deaths and their percentage distribution Source: https://www.nadis.org.uk/disease-a-z/sheep/lambing/lambing-part-4-ensuringsurvival-of-newborn-lambs/

The causes of lamb death and contributing factors are briefly summarized in Table 2.

Factors	Basic causes	Suggestions
Low birth	Malnutrition	Pay attention to pregnancy nutrition (especially in
weight	Exposure to	case of twins)
	cold	Sheltered lambing pen
	Wild animal	Precautions to prevent wild animal attacks
	attack	
Dystocia in large	Birth trauma	Pregnancy feeding (especially single-bearing)
lamb		Taking good care of the animal during birth
		sheepherding
		Genetic selection
Lack of energy	Birth trauma	Adequate nutrition during pregnancy
in sheep to	Malnutrition	Placing feeders in lambing pens
complete birth		
Abandoning or	Birth trauma	BCS of ewes at birth
mismothering	Malnutrition	Do not place feeders in birthing areas
		Sheep selecting with good maternal characteristics

Table 2. The causes of lamb death and contributing factors

		Herd size and number of animals to give birth
Unsufficient	Malnutrition	BCS of ewes at lambing
colostrum	Exposure to	Do not place feeders in birthing areas
production	cold	Feeding grain to sheep to increase the amount of
	Wild animal	colostrum
	attack	
Predators	Protecting wild	Control of predators
	animals	
Advers weather	Exposure to	Sheltered lambing pens,
conditions	cold	Designing of paddocks against prevailing winds

Source: Trompt, 2014.

An important reason for possible lamb deaths before and immediately after birth is related to abortion. Although there are many reasons for abortion, this rate should not exceed 2% in the herd. If the abortion rate exceeds 2%, action should be taken as soon as possible to solve the problem, and both the abortion membranes and the discarded lamb should be sent to a laboratory to find out the cause of the problem (Linch and Brien, 2016). Since peri-natal behavior in sheep is difficult to measure, intensive observation of sheep at birth is necessary. Often attempts are made to measure lamb viability through subjective estimates that are not measured directly after birth, but rather at birth. For this purpose, first of all, ear fitting should be done within the first 12-36 hours after birth and birth information should be recorded individually (McCoard et al. 2020). Otherwise, since lamb deaths may increase, it is useful to know more detailed characteristics aimed at estimating viability or lamb survivability (Table 3) (Brien et al. 2010).

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Table 3. The scoring	system is used to	subjectively	define the	strength of the lamb
during ear tagging.				

Score	Description
0	The lamb is still wet. Newborn, no record
1	A constant movement. There is bleating behaviour towards the mother.
	There is a behaviour of reaching the mother in a short time and following
	her.
2	Bleating behaviour is very common. There is constant movement when
	held. Moves towards the host when released
3	There is less mobility. There is movement towards the mother's voice, but
	there is no contact. Bleating can be observed
4	There is less mobility. There is the behaviour of trying to walk. But there
	is no specific purpose. There is no response to the mother's bleating
5	There is less mobility when held. Lying down behaviour is very common
	when a lamb is released.

Kaynak: Brien ve ark. 2010

It is also necessary to pay attention to the behaviour of the lambs during birth and counting. These behaviors; It is the duration of standing, bleating, contacting and following the mother and contacting the udder after numbering. The age of the dam, the type of birth of the lamb, and the behaviour of the dam also affect the postnatal viability or viability of lambs (Everett-Hincks et al. 2005). Unlike behaviours recorded at birth, behaviours resulting from enumeration practice are not affected by birth weight. However, it has been determined that these are affected by the structure of the pasture used during birth. This may reflect differences in the availability of feed in the environment and also the micro-environmental impact that pasture can create (Lynch et al. 2010)—heritability estimates for these behaviours performed after release from enumeration range from 0.09 to 0.16. Once again, it suggests that lamb strength has some genetic component (Matheson et al. 2011). However, there is little point in maintaining the viability of lambs unless a clear relationship to survival can be established.

2.6. Relationships Between Vitality and Viability in Postnatal Lambs

A lamb's ability to stand up and nurse its mother immediately after birth affects its viability. Many studies have demonstrated this relationship between behaviour and survival, highlighting the importance of birth weight and the strength of the parent-offspring relationship at birth. A study showed that a large proportion of lambs that were unable to contact the udder or that took longer than average to do so died (Napolitano et al. 2008). Although the researchers did not show a direct relationship between the duration of standing or suckling attempts in lambs and viability, this was revealed in subsequent studies. A reduction in the latency to perform key behaviours has been shown to lead to increased survival rates (Nieper et al. 2017). It was also determined that lambs that died within three days after birth showed a delay in the time to reach the knees, attempt to stand up, stand and reach the udder, compared to lambs that survived in the same period (Dwyer et al., 2008). Study results reveal that the relationship between postnatal behaviour and viability is not linear but threshold. Lambs that fail to reach this threshold are at greater risk of mortality, and lambs that exceed it survive, but higher survival is not observed in lambs that show more rapid behavioural progression once above this threshold.

While the importance of studies examining the phenotypic relationships between the behaviour and viability of newborn lambs is emphasized, there is limited research on genetic relationships. Published research on perinatal lamb behaviour has not examined correlations between behaviour and survival but has instead compared both maternal and direct breeding values for behavioural traits between death and survive lambs (Cloete et al., 2009). In this analysis, it was determined that deceased lambs exhibited higher reproductive values than surviving lambs in terms of birth time, time from birth to standing, and time from standing to suckling. Indirect estimates of lamb viability have also been shown to be genetically linked to survival. Subjective lamb vitality score has been shown to show a low but positive genetic correlation with survival, although standard errors are high (-0.26) (Brien et al. 2010). To summarize, immediately after birth, before the first feeding, the lamb produces heat by burning brown fat. Brown fat is a specialized fat reservoir used to generate 50% of the total heat generated, supporting adaptation to the cold challenge of the extrauterine environment and helping to prevent hypothermia. During cold exposure, the lamb loses heat rapidly (McCoard et al. 2019). Brown fat stores form in the mid-to-late period of pregnancy and decrease with inadequate nutrition of sheep, so it is important to meet the feed requirements of sheep. The decrease in the amount of brown fat prevents hypothermia, as it shortens this period when newborn lambs use brown fat tissue to maintain body temperature (McCoard et al. 2020).

2.7 Udder Health in Sheep

Udder health is an often overlooked feature in terms of the viability of lambs and their good performance in later life periods. The viability of lambs depends mainly on the colostrum they receive/drink from the mother in the first few days following birth. For this reason, udder health of the mother becomes more important, especially in dairy-type farms (Richmond et al. 2017). Quality colostrum is needed to ensure the survival and development of the lamb. This is closely related to udder health and animal welfare in sheep. Achieving this condition is possible with nutrition, which is an important environmental factor. In other words, the protein content of the feed given to dams should be 16-18%. Because protein insufficient nutrition in advanced pregnancy may cause a decrease or delay in colostrum production. The mucous structure on the teat may need to be removed at birth so that the lamb can access the colostrum. This mucous plug within the teat ducts prevents colostrum from flowing out or bacteria from entering the teat. In case of mastitis or insufficient milk in sheep

for any reason, it may be necessary to give lamb colostrum first and then milk. Mastitis and other problems such as pregnancy toxaemia, nipple lesions and retained fetal membranes can negatively affect milk production (Stevens et al. 2017a). The risk of mastitis is affected by environmental conditions, including infective microorganisms. The most common of these negative factors are; Staphylococcus aureus, Mannheimia haemolytica, Coagulase-negative staphylococci, Pasturella spp., Escherichia coli and Klebsiella. Other environmental factors are the age of the ewe, the structure of the feed given, the number of suckling lambs and udder characteristics. Mastitis can lead to clinical and subclinical conditions that cause excruciating pain in sheep. To improve lambs' access to the teats, ewes that should be avoided should be selected for breeding. In other words, ewes with drooping udder structures resulting from damage to the lateral and/or medial suspensory ligaments, such that the teats hang close to the ground, should be separated as non-breeding. Damage to the udder or teats (bruising, tearing) is more likely to develop mastitis, and it may be difficult for lambs to suckle their dams. Large or have downward-pointing teats may prevent the lamb from reaching the teat (Stewens et al. 2017b).

2.8. Possible Diseases and Problems Encountered in Lambs

Lambs may need a veterinarian after lambing for many reasons (Taşkın et al. 2015). Some common diseases/conditions that are seen in bad condition of a lamb that refuses to suckle from its mother or is not accepted by its mother are given below:

Discharge/Diarrhea – This is the most common problem we see and can be broadly divided into nutritional and infectious. The vast majority are nutritious and usually easy to fix. You need to consult a veterinarian to get the best advice.

Belly and Joint Disease: Bacteria can enter the bloodstream through the wet navel, often causing belly button infection or joint disease/infection. If the

lamb looks lame and moves slowly or holds one limb up, it may have a joint disorder. Both are treated with a course of antibiotics and it is important to address them promptly.

Eyes - eyes should be open and clean. If there is discharge, squinting or discolouration, contact your veterinarian. Problems may include entropion (eyelids turning into eyes and eyelashes scratching the cornea), pinkeye, or conjunctivitis.

Abomasal Bloat – This is the biggest cause of death among artificially reared lambs, usually from 3-4 weeks of age. Acute bloating occurs approximately 1-2 hours after large amounts (>500 ml) of milk replacer are given to lambs, and this is caused by irregular feed consumption. Symptoms of this include acute depression, swollen and tense abdomen, pain (colic) and if the lambs are left untreated death is rapid. This should be taken to a veterinarian as soon as possible as it can be a life-threatening situation. The best way to prevent bloat is to feed little and often, using a whey-based milk replacer or yoghurt milk replacer.

3. SUITABLE ENVIRONMENT and HOUSING for LAMB RAISING

Mammals have a set of behavioural and physiological mechanisms that allow them to adapt to various situations to maintain body temperature between certain values to survive. Lambs are particularly vulnerable to environmental factors that can affect their survival, health and welfare, especially in the first few weeks of life (Figure 8). Richmond et al. Some of the welfare criteria and indicators associated with the "Good Environment" well-being principle as defined by (2017) are given in Table 3. These may be positive animal-based welfare measures of the quality of the environment provided to the lambs during rearing.



Figure 8. Pens with plastic grates (left) and straw mat system (right). Source: https://www.mdpi.com/2073-4395/9/11/694

Welfare Parameters	Welfare Indicator
Well-being/comfort in the	Lying down time
recreation area	Number of beds per unit of time
	Skin pollution
Thermal welfare	Respiratory rate
	Shake
	Rectal temperature
	Hematocrit, plasma protein level in blood or
	urine
ease of movement	Placement frequency
	The slipperiness of the ground
	Aggression and displacement
	Excessive growth of nails

Table 3. Relationship between environment and welfare in lambs

Source - Richmond et al. 2017.

There are general principles that must be taken into consideration for a good lamb-raising facility. First of all, the environment must be clean, dry, warm, well-ventilated and free of airflow and dust. Air quality inside the shelter is very important. The housing conditions of the lambs live should be checked by getting down on your hands and knees, especially in the lambing pen, at lamb level, that is, in the pen. It is necessary to avoid housing or keeping too

many lambs in the pen. Lambs should be grouped by age and/or feeding ability. Group pens require at least 0.5-0.6 m² of space per lamb (DEFRA, 2003). For lambs up to the 12th week, these standards may even be slightly larger. Floor space per lamb may need to increase over time depending on housing duration. It has shown that providing 0.9 m² of space per lamb in the first three weeks of rearing is associated with improved growth and health compared to 0.5 m². Based on experience in commercial breeding units, small groups (10-15 lambs) are recommended for the first 2-3 weeks of rearing to reduce competition at the dairy feeder, support training to feeders and facilitate lamb handling. After animal health checks are carried out, the stocking rate can be increased after 3 weeks.

4. COLOSTRUM FEEDING

Good nutrition is one of the easiest strategies to support the health, production and welfare of lambs. It is important to remember that the growth performance and health of lambs will be affected by many factors other than the amount of milk or milk replacer offered (Novak and Poindron, 2006). Such factors are respectively; These can be listed as race, birth type (single, twin, triplets, etc.), quality of the ingredients used in milk replacer formulations, feeding frequency, animal health problems, settlement frequency, amount and quality of starter feeds (solid feeds) and environment. Nutrition provides the nutrients necessary for growth and development and is an important determinant of the response of the immune system to adverse environmental conditions (Table 4). Malnutrition can have lifelong negative consequences for health, productivity and well-being.

Welfare characteristic	Welfare parameters	Welfare indicators
	Not hungry for a long	Body condition score
	time	Survivability for lambs
Good nutrition	Not thirsty for a long time	Skin patch test
		Blood and urine sample
		Water drinking

Table 4. Welfare measures depending on nutrition

Kaynak: Richmondve ark. 2017

3 critical periods affect the immune system in newborn ruminants in the first 3 months of life: These are respectively; a. Feeding with colostrum, b. Milk feeding, etc. It is weaning. Colostrum is the first milk produced by sheep and contains high levels of nutrients, bioactive substances, energy and immunoglobulins that play an important role in defence against external pathogens (Swalha et al. 2007). Lambs must receive adequate amounts of colostrum in the first few days of life. This is because lambs are born without natural immunity. After all, the ruminant placenta blocks the transfer of immunoglobulins from the dam to the fetus. These immunoglobulins do not occur until the lambs' immune systems are fully functional, which is about 3-4 months old. As the time after lambing increases, the amount of immunoglobulin and nutrient concentration in colostrum decreases relatively, and the lamb's intestine loses its ability to absorb immunoglobulins from colostrum on the first day of life. Therefore, the lamb should consume sufficient colostrum immediately after birth. Inadequate neonatal absorption of immunoglobulins on the first day of life is associated with failure of passive transfer of immunity. This can lead to increased mortality and lasting negative effects on performance. Higher mortality and morbidity rates have been observed in colostrum-deprived lambs (up to 80%) compared to colostrum-fed lambs (up to 20%)

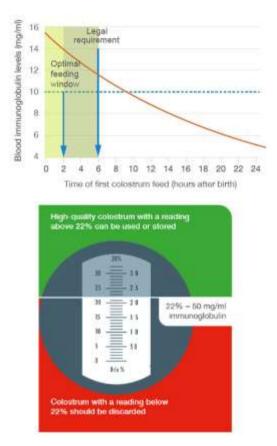


Figure 9. Change in immunoglobulins during colostrum feeding Source: https://ahdb.org.uk/knowledge-library/using-a-brix-refractometer https://projectblue.blob.core.windows.net/media/Default/Dairy/Graph%20showing%20effect% 20of%20time%20of%20first%20colostrum%20feed%20on%20antibody.jpg

Data obtained in research to date show that the transport of immune substances in colostrum is at a good level when lambs stay with their mothers for at least 48 hours. Lambs need to get enough colostrum early (within 6-8 hours after birth). Colostrual milk must be of high quality and must be given for at least 2 days, preferably 3-4 days. If lambs receive colostrum from the mother at birth, feeding with colostrum on day 1 and then a gradual transition from colostrum to normal milk is mandatory (Figure 9). If artificial growth is necessary, colostrum at the rate of 15% of the birth weight is given to the

animals during the day, divided into 5-6 meals for the first few days. The quality of colostrum will decrease significantly within 24 hours, even if stored in the refrigerator. Therefore, to achieve the best results in terms of quality, priority should be given to giving the freshest colostrum to the youngest lambs (Canto et al. 2023). Experience in the dairy sheep industry shows that sows often contain very thick colostrum on day 1 of lactation, which can be difficult for lambs to wean. Therefore, extra care needs to be taken. It is important to remember that competition for teats between multiples or poor maternal ability can affect the ability of lambs to obtain adequate colostrum. It may be necessary to carefully check newborns for signs of hunger and, if necessary, bottle-feed the lambs.

4.1. Which Milk Replacer Should Be Used?

There is a wide variety of lamb milk substitutes commercially available. These vary in quality and price. The high cost of milk proteins leads to the development of cheaper lamb milk replacer formulations. These formulations use whey proteins or vegetable proteins and fats as cheaper alternatives to casein milk proteins and milk fat (Figure 10). Spray-dried milk proteins are recommended for the formulation of milk replacers. Some research trials have suggested that non-dairy protein sources such as soy protein can be used, but high inclusion rates (up to 40%) may suppress growth, and discretionary feeding should be avoided due to sedimentation issues with inclusion rates above 30%. Soybean concentrate or full-fat soy flour. Most published studies evaluating the effects of lamb milk substitute protein sources on lamb performance are over 30 years old and have focused on soybean protein. The inclusion of hydrolyzed wheat protein as a cheaper alternative to soy protein is not common in commercially available milk replacers.



Figure 10. An example of feeding lambs with milk replacer Source: https://www.fwi.co.uk/livestock/livestock-feed-nutrition/6-things-to-consider-whenfeeding-lambs-milk-replacer

There are six important issues to consider when raising lambs with milk replacer. These are as follows:

a. Should you use skim milk or whey-based powders?

Whether the milk replacer is skim or whey-based, high levels of animal protein are important.

b. What should be considered on the product label?

Ingredients are listed by inclusion, with the main ingredient listed at the top. In most lamb milk substitutes, whey is at the top of the list, with skim being the second most dominant ingredient. We can say that "milk protein is necessary for muscle growth; the higher the percentage, the faster the growth time - and fat is the source of energy and gives the lamb the ability to build muscle." Crude fibre should be as low as possible; Ideally, it should not be higher than 0.05%. A percentage higher than 0.15% is indicative of a product containing vegetable protein that is less digestible for young ruminants than animal protein. "It would not be right to feed the lambs with a cheap milk replacer. This is because the lambs will not be able to get enough nutrients and energy, so they will not develop sufficiently and a lower carcass weight will be achieved.

c. What should be the feeding rate from weaning to weaning?

For feeding to be done four times, there should be between 1-1.5 litres of feed per day - the upper limit of 1.5 litres. However, substitute feed should not be given until the lamb is one week old. Feeding a large number of meals (number of meals) during the day reduces the risk of bloat in animals; It is recommended to feed as little but frequently as possible.

4.2. Does Casein-Based Milk Replacer Cause Swelling in the Abomasum?

One of the health problems commonly observed in artificial lamb-rearing systems is abomasal bloating. Anecdotally, claims have been made that casein-heavy milk replacer formulations cause bloating in both small- and large-scale growing systems. The potential cause is thought to be the lack of clot formation. It prevents bloating that occurs in whey or vegetable protein-based formulations (Banchero et al. 2016). In all other studies conducted as well as in the studies described above, no cases of abomasal bloating were observed with any of the milk replacers used. This suggests that factors other than the milk replacer formulation or protein source likely cause abomasal swelling. Factors that will reduce bloat include preventing overfeeding and maintaining good hygiene regarding feed preparation.

4.3. Milk Feeding Equipment

A wide variety of milk feeders are available. The choice of dairy feeding equipment depends on the number of lambs raised, the infrastructure available and the choice of feeding system (Figure 11).



Figure 11. Some mechanisms for determining motility/vitality in artificial incubation in lambs Source: Abecia and Canto, 2023b

Some suggestions on the subject are given below;

a. All feeding equipment should be cleaned regularly to reduce the risk of infection and animal health problems. If a bottle or feeding machine is used, all equipment used to mix and feed milk should be cleaned with detergent after each feeding and rinsed thoroughly before drying.

b. If automatic feeders are used, they should be cleaned and calibrated at least weekly. It is necessary to include the mixing bowl as well as the milk lines leading to the teats in the cleaning process, as milk residue can accumulate in the lines and provide a perfect environment for pathogens. It is recommended to have two sets of milk lines so that the used set can be removed and replaced with a clean set. Among changes, Clean lines can be dried and stored or placed in a bucket (with a lid) containing a sterilizing solution to keep them clean before use. Milk lines should be kept as short as possible.

c. The area around the teats mounted on the feeding panels needs to be kept clean, and the teats themselves need to be cleaned regularly. Nipples should be monitored daily for damage and replaced when necessary.

d. When automatic feeding machines are used, the area around milk feeders often becomes dirty with milk. It may be helpful to place a rubber mat

or net under the feeding area to ensure drainage and easy feeding. Regular cleaning should be done to prevent pathogen formation.

e. Automatic feeders require teats to be mounted inside pens to allow lambs free access to milk. Studies on this subject have found that it is preferable to mount teats in a feeding stall where other lambs cannot push them away from the teat, compared to mounting teats directly on the side of the pens (Figure 12). Competition between lambs when feeding from automatic feeders, especially when young, can reduce feed intake and growth rates, which can impact offspring health. If teats are mounted directly on the side of the pens, it is recommended that they be mounted back from the wall of the pen to reduce damage to the teats from lambs chewing on the teats and to ensure there are no sharp edges as this could cause facial injury to lambs



Figure 12. Some examples of artificial rearing devices in lambs

4.5. Milk Feeding

The nutrition of newborn lambs relies entirely on milk to provide the nutrients and energy required to support growth and development. Because the rumen is dysfunctional at birth, the small intestine plays an important role in directing nutrient metabolism, immune function, and therefore survival and growth. Lambs must receive sufficient milk. In naturally reared lambs, competition between offspring (e.g. triplets and quadruplets) and low lactation performance of ewes due to malnutrition and/or animal health problems can reduce milk intake (Ünal et al. 2018). In such cases, lambs may need assistance with feeding or artificial rearing in the first few days of life. There are many different options for milk feeding of artificially raised lambs, from restricted

milk feeding to optional milk feeding. Feeding systems with both manual and automatic feeding equipment. Discover which system suits your budget, facilities, staff availability and skill level of existing staff.

Practical approaches or applications on the subject are given below, in order. These are respectively;

- Make sure that milk replacer is always freshly prepared and fed to the animals.
- Give warm milk replacer (skin temperature, i.e. not exceeding 37oC), especially to very young lambs, as this reduces the need to partition energy into heat production. It should be stirred to a consistent temperature for each feeding.
- Always follow the manufacturer's instructions for expressing milk replacer and do not mix at levels below recommendations. It is important to note that if an instruction states "200 g/L" this means 200 g of powder totalling 1 L of reconstituted milk replacer. Milk replacers can be concentrated (up to 250 g/L) to reduce milk volume and increase nutrient delivery to the lamb, especially in limited milkfeeding systems.
- Lambs should not be overfed. Maximum 350 ml per feed, depending on the size of the lamb. Watch the lamb's stomach and feeding should be stopped if the stomach begins to protrude beyond the ribs. Overfeeding can cause pet health problems such as diarrhoea and bloat.
- Equipment should be thoroughly cleaned and rinsed with warm soapy water after each use.
- Lambs should always be fed standing or with their heads in a natural position.

- A lamb should not be fed like a human baby while lying on its back. This can cause milk to leak into the lungs and cause the lamb to develop pneumonia or suffocate.
- Always ensure ad libitum fresh water is available, even when colostrum feeding.

4.6. A Plan Should Be Made for More Lambs Born After Multiple Pregnancy

If there is a high gestation with multiple triplets, it will be necessary to think and make a plan about how to feed these lambs. If the triplets are separated and fed effectively before lambing, it may be possible for them to raise their lambs, but often this may not be possible (Övet, 2023). Adopting excess lambs into single ewes can be a good way of dealing with excess lambs without encountering too many pets, but consideration must be given to how to do this and sometimes this is not always successful. In cases where it is not possible for the ewe to adopt (kill) another lamb, a plan will be needed to take care of motherless/orphan lambs. It is important to have a warm, dry area for these lambs. Having automatic feeding machines or feeders can greatly reduce the workload when there is more than one lamb (Figure 13). This also needs to be taken into account as lambs will need access to concentrated feed earlier than other lambs.



Figure 13. Examples of lamb rearing and individual lambing pens https://www.tirlanfarmlife.com/farm-advice/detail/article/top-10-tips-for-a-successful-lambing https://www.teagasc.ie/media/website/news/daily/sheep-photos/ewe-with-young-lambsfeatured.jpg

5. WHY ARE NEWBORN LAMBS SO VULNERABLE?

Unlike humans, there is no transfer of antibodies from mother to offspring, so they are born with no protection against disease and are therefore dependent on colostrum for passive immunity. Newborn lambs also have a very permeable intestinal tissue, or barrier, that can allow ingested bacteria and toxins to pass directly into the bloodstream. They are born with a limited amount of brown fatty tissue. This structure is effectively energy reserves from birth until it can be fed. Newborn lambs have a very large surface area to body weight ratio, making them more susceptible to heat loss, especially when wet. All lambs need to be identified as soon as possible and bonded well with their dams. No matter what breed you have, there is always the possibility of rejection. If this occurs in a situation where the bond cannot be rekindled, intervention needs to be done quickly. Lambs are often born in both cold and wet conditions. Because of this environment and its large ratio of surface area to body weight, it can often consume most of its limited energy levels simply by maintaining body temperature. It can save energy by keeping them dry. To minimize the possibility of disease problems, the lamb's belly should be treated with iodine as soon as possible after birth, ideally within 15 minutes. Make sure the lamb is breastfeeding and receiving adequate colostrum as soon as possible after birth (within 6 hours). This highly nutritious energy source will protect the lamb against diseases and help maintain its body temperature, thus ensuring its survival. Be sure to check that the ewe has sufficient milk supply for its lamb(s). While it is preferred to feed the lamb with sheep colostrum, if this colostrum is insufficient for any reason, it should be fed quickly from another source. This can be fresh or frozen from another sheep or, if necessary, a high-quality natural alternative such as Volostrum. Try to avoid unnecessary interventions. Ideally, assistance with lambing should only occur if the life of the ewe or lamb is at risk. Finally, make sure you always maintain good hygiene. One in five lambs may die due to an unhygienic environment and this can be prevented.

6. FEEDING of MILKING SHEEP

Nutrition during pregnancy affects colostrum yield and quality. Sheep needs are highest in the first 6-8 weeks of lactation. For optimum performance - feed according to needs - sheep must have body reserves (fat). Energy and protein requirements increased by 30% and 55% respectively. Insufficient energy intake increases protein needs. Milking sheep need plenty of clean, fresh water. A feed budget should be made and checked in advance to more accurately determine coarse and dense feed expenses in the farm (Abdelsaved et al. 2015). Grass yield and quality in pasture areas used for grazing vary throughout the year. There should be a clear understanding of what is owned, including the amount and inclusion. Over time, a profile should be created of your on-farm feed sources and if/when you may need additional supplements. Create a feed budget to understand what your animals' feed requirements are throughout the year and plan feeding management and stocking rates based on your available feed. Pregnancy check data can be used to estimate feed requirements for pregnancy and lactation. The greater the percentage of control/screening for pregnancy detection, the greater the requirements to meet the nutrient needs of multiple offspring. Feed requirements are ~50% higher for triplet ewes and ~20% higher for twin ewes compared to single-bearing ewes. Good nutrition (quality and quantity) is particularly important during gestation and lactation to ensure optimal survival and growth of lambs as well as the continued reproductive performance of ewes (next season). If you are producing crops, productivity and quality must be determined to inform your feed budget. If grain feed is required and economical, purchasing can be done.

7. CONCLUSION and SUGGESTIONS

There is evidence linking perinatal lamb behaviour and viability to survival and should therefore be investigated as a potential method to reduce mortality rates. Some of this variation can be explained by genetics, as lamb behaviour after birth is highly variable. Physiological differences between lambs may also help explain this variation, and it is reasonable to assume that 'metabolic maturity' at birth may influence behavioural progression. There is a clear need to classify maturity at birth in lambs, identify the factors that influence this, and investigate whether relationships exist with behaviour and vigour. In the current study, we aim to determine whether hormones and metabolites previously involved in 'metabolic maturity' are associated with perinatal lamb behaviour. It is hypothesized that lambs with increased behavioural progression and vigour after birth are born with increased metabolic maturity as defined by endocrine and metabolite changes, which may have consequences for postnatal lamb survival.

REFERENCES

- Abdelsayed M, Thomson PC, Raadsma HW. 2015. A review of the genetic and non-genetic factors affecting extended lactation in pasture-based dairy systems. Anim Prod Sci. 55:949–966.
- Abecia, JA., Canto, F. 2023a. Measuring lamb activity during artificial rearing by actigraphy. Conference: US Precision Livestock Conference, Knoxville, Tennessee
- Abecia, JA., Canto, F. 2023b. Circadian, feeding, and locomotor activities of artificially reared lambs measured by actigraphy. Journal of Applied Animal Research 2023, Vol.51, No.1, 234–241
- Banchero, GE., Milton, JTB., Lindsay, DR., Martin, GB., Quintans, G. 2016. Colostrum production in ewes: a review of regulation mechanisms and of energy supply. Animal, 9(5): 831-837
- Bancheva, T., Stoycheva, S., Dimitrova, T., Markov, N., Mondeshka, L.,
 Hristov, M. 2022. Impact of Various Factors on Live Birth Weight
 Lambs Review. Scientific Papers. Series D. Animal Science. Vol.
 LXV, No. 1, 2022 ISSN 2285-5750; ISSN CD-ROM 2285-5769; ISSN
 Online 2393-2260; ISSN-L 2285-5750.
- Bates, AL., McGrath, SR., Robertson, SM., Refshauge, G. 2022. Mating Conditions and Management Practices Influence Pregnancy Scanning Outcomes Differently between Ewe Breeds. Animals, 2022,12,2908
- Binns, S.H., Cox, I.J., Rizvi, S. Green, L.E., 2002. Risk factors for lamb mortality on UK sheep farms. *Preventive Veterinary Medicine*, 52 (3), 287–303.
- Brien, F., Hebart, ML., Smith, DH., Hocking Edwards, JE., Greef, JC., Hart, KW., Refshauge, G., Bird-Gardiner, TL., Gaunt, G., Behrendt, R., Robertson, MW., Hinch, G.N., Geenty, KG., van der Wef, HJ. 2010.
 Opportunities for genetic improvement of lamb survival. Animal Production Science, 50:1017–1025.

Canto, F., González, E., Abecia, JA. 2023. Effects of Implanting Exogenous Melatonin 40 Days Before Lambing on Milk and Colostrum Quality. Animals, 12,1257.

Caroprese, M. Sheep housing and welfare. Small Rumin. Res. 2008, 76, 21–25.

- Cloete SWP, Misztall, Olivier, JJ. 2009. Genetic parameters and trends for lamb survival and birth weight in a Merino flock divergently selected for multiple rearing ability. Journal of Animal Science 87, 2196–2208. doi:10.2527/jas.2008-1065
- Cristobal Carballo O, Khan MA, Knol FW, Lewis SJ, Stevens DR, Laven RA, McCoard S. 2019. Impact of weaning age on rumen development in artificially reared lambs. J. Anim. Sci. 97:3498-3510.
- DEFRA, 2003. Code of Recommendations for the Welfare of Livestock: Sheep, Defra Publications Admail 6000 London.
- Durak, MH., Ateş, A., Bilal, T. 2022. Effect of Energy Deficiency at The End of Pregnancy on Sheep and Lamb Birth Weights. Dicle Üniv Vet Fak Derg 2022;15(1):5-8
- Dwyer C.M. 2008. The welfare of the neonatal lamb. Small Ruminant Research, 76,31–41.doi:10.1016/j.smallrumres.2007.12.011
- Everett-Hincks JM, Dodds KG (2008) Management of maternal-offspring behaviour to improve lamb survival in easy care sheet systems. Journal of Animal Science 86, E259–E270. doi:10.2527/jas.2007-0503.
- Fridlund, C., Humblot, P., Bage, R. & Soderquist, L., 2013. Factors affecting the accuracy of pregnancy scanning in ewes. *Veterinary Record*, 173 (24), 606–606.
- Fthenakis, G.C., Arsenos, G., Brozos, C., Fragkou, I.A., Giadinis, N.D., Giannenas, I., Mavrogianni, V.S., Papadopoulos, E. & Valasi, I., 2012.
 Health management of ewes during pregnancy. *Animal Reproduction Science*, 130 (3–4), 198–212.

- Gebretensay, A., Alemayehu, G., Rekik, M., Alemu, B., Haile, A., Rischkowsky, B., Aklilu, F. & Wieland, B., 2019. Risk factors for reproductive disorders and major infectious causes of abortion in sheep in the highlands of Ethiopia. *Small Ruminant Research*, 177, 1–9.
- Genfor, E., Magnusson, U., Moliso, M. M., Wieland, B., König, U. Hallenberg, GS., Båge, R. 2023. Preventive herd management practices and their effect on lamb mortality in Ethiopia. Tropical Animal Health and Production (2023) 55:42.
- Goddard P, Waterhouse T, Dwyer C, Stott A 2006. The Perception of the welfare of sheep in extensive systems. Small Ruminant Research 62: 215-225.
- Holmøy IH, Waage S 2015. Time trends and epidemiological patterns of perinatal lamb mortality in Norway. Acta Veterinaria Scandinavica 57: 65-76.
- Jensen AC, Khan MA, Knol FW, Peterson SW, Morel PCH, McKenzie C, Stevens DR, McCoard SA. 2017. How does feeding meal affect the growth of artificially reared East Friesian-cross dairy lambs? Proceedings of the New Zealand Society of Animal Production 77:13-17.
- Kandemir Ç, Alkan İ, Yılmaz Hİ, Ünal HB, Taşkın T, Koşum N, Alçiçek A 2015. İzmir yöresinde küçükbaş hayvancılık işletmelerinin coğrafik konumlarına göre genel durumu ve geliştirilme olanakları. Hayvansal Üretim 56 (1): 1-17.
- Koyuncu, M., Duymaz, Y., Merve Karaca, M. 2018. Improvement of Survival in Newborn Offspring J. Biol. Environ. Sci., 2018, 12(34), 23-29.
- Linch G.N., F. Brien, 2016. Lamb survival in Australian flocks: a review. Animal Production Science 54, 656–666.

- Lynch, E.M.; Earley, B.; McGee, M.; Doyle, S. 2010. Characterisation of physiological and immunological responses in beef cows to abrupt weaning and subsequent housing. *BMC Vet. Res.* 2010, 6, 37.
- Matheson SM, Rooke JA, McIlvaney K, Jack M, Ison S, Bünger L and Dwyer CM 2011. Development and validation of on-farm behavioural scoring systems to assess birth assistance and lamb vigour. Animal 5, 776–783.
- McCoard SA, Stevens DR, Whitney TR. 2019. Sustainable sheep and goat production through strategic nutritional management and advanced technologies. Chapter 13 in "Animal Agriculture: Sustainability, Challenges and Innovations" Ed. Bazer FW, Lamb GC, Wu G. Academic Press.
- McCoard, SA, Knol, FW, Stevens, DR. 2020. Lamb Rearing Technical Manual. www.agresearch.co.nz. Pg:1-66.
- McHugh, N., Berry, D.P. Pabiou, T. 2016. Risk factors associated with lambing traits. Animal(2016),10:1,pp 89–95.
- McDowall, ML., Edwards, NM., Hynd, PI. 2011. The effects of short-term manipulation of thyroid hormone status coinciding with primary wool follicle development on fleece characteristics in Merino sheep. Animal (2011), 5:9, pp 1406–1413.
- Napolitano, F.; De Rosa, G.; Sevi, A. 2008.Welfare implications of artificial rearing and early weaning in sheep. Appl. Anim. Behav. Sci. 2008, *110*, 58–72
- Nieper BA, Khan MA, Ganesh S, Knol FW, Peterson SW, Stafford KJ, Stevens DR, McCoard SA. 2017. The effects of early access to meals on the behaviour of artificially reared dairy lambs. Proceedings of the New Zealand Society of Animal Production 77:18-22.
- Nowak R and Poindron P 2006. From birth to colostrum: early steps leading to lamb survival. Reproduction Nutrition Development 46, 431–446

- Övet, C. 2023. Colostrum-induced passive immune transfer in lambs. Journal of Istanbul Veterinary Sciences, 7(2), 80-88.
- Plush, KL. 2013. Metabolic maturity and vigour in neonatal lambs, and subsequent impacts on thermoregulation and survival. Bachelor of Animal Science (Honours). A thesis submitted for fulfilment of the requirements for the degree of Doctorate of Philosophy. The School of Animal and Veterinary Sciences. The University of Adelaide, Roseworthy, South Australia, Australia
- Richmond SE, Wemelsfelder F, Beltran de Heredia I, Ruiz R, Canali E and Dwyer CM. 2017. Evaluation of animal-based indicators to be used in a welfare assessment protocol for sheep. Frontiers in Veterinary Science Volume 4, Article 210 doi: 10.3389/fvets.2017.00210 – supplementary information.
- Stevens D, Knol FW, Neiper BA & McCoard SA. Post-weaning performance of East Friesian cross ewe lambs grazing ryegrass or plantain-based pastures after rearing on two contrasting diets. J. NZ Grasslands 79, 49-54 (2017).
- Stevens D, Samuelsson L, McCoard S, Day L, Young W, Bartlett N, Konui W, Gatley P, Hammond N, Macdonald T, King M, Hewittson J. 2017b. Using a co-innovation approach to accelerate the development of dairy sheep enterprises in New Zealand. 13th European IFSA Symposium, 1-5 July 2018, Chania (Greece).
- Swalha RM, Conington J, Brotherstone S, Villaneva B 2007. Analyses of lamb survival of Scottish Blackface Sheep. Animal 1, 151–157.
- Taşkın T 2014. Yetiştirme pratikleri. Koyun Keçi Genetik Islah Çalıştayı, 1113 Haziran, Uşak, s.79.
- Taşkın T, Ataç FE, Önenç SS 2005. Kuzulama sırasında karşılaşılan sorunlar en aza nasıl indirilebilir? Hasad Hayvancılık 3:1621.

- Taşkın T, Ünal HB, Canpolat Ö 2015. Koyunculuğun temel esasları. Hasad Yayıncılık Ltd. Şti, İstanbul.
- Tibbo, M., Mukasa-Mugerwa, E., Woldemeskel, M., Rege, J.E.O., 2003. Risk Factors for Mortality Associated with Respiratory Disease among Menz and Horro Sheep in Ethiopia. The Veterinary Journal, 165 (3) 276-287
- Tibbo, M., Woldemeskel, M., Gopilo, A., 2001. An outbreak of respiratory disease complex in sheep in Central Ethiopia. Tropical Animal Health and Production, 33 (5), 355-365.
- Tozlu Çelik, H., Aslan, FA., Kaşko Arıcı, Y., Kahveci, ME. 2023. Karayaka koyunlarında yaşın farklı dönemlerdeki vücut kondisyon durumu üzerindeki etkileri. Akademik Ziraat Dergisi 12(1): 145-152 (2023)
- Ünal, HB., Taşkın, T., Kandemir, Ç. 2018. Küçükbaş Hayvancılıkta Yavru Ölümlerinin Azaltılmasına Yönelik Barındırma ve Yetiştirme Uygulamaları. J. Anim. Prod., 2018, 59 (2):55-63, DOI: 10.29185/ hayuretim.430488.

CHAPTER 10

PROFITABLE MILK PRODUCTION IN SHEEP BREEDING

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INTRODUCTION

Ewe milk has a special importance in terms of the biochemical values it contains. Nowadays, ewe milk production is generally carried out under very difficult conditions. Although these difficult conditions have changed until today, they are generally similar. As a result, despite high labor costs, milk production remains low. For this reason, profitability in production of ewe's milk in Türkiye remains low.

In fact, there may be more than one reason for this economic inadequacy of ewe milk producers.

Among the agricultural sector activities, livestock breeding always has a very important place in terms of economy. Such sectors may have different characteristics. One of these is small ruminant breeding. It has a special importance especially for the Eastern Anatolia Region. In our country, methods of sheep and goat breeding vary or differ depending on various factors. These factors can be listed as follows: Factors such as the social, natural and economic situation of the regions, the existence of coarse or concentrated feed opportunities, the relationship of such factors with the plant production sector and the consumption habits of the population. Among the main agricultural sector components in Türkiye, livestock breeding, transhumance activities and semi-nomadic or nomadic sheep farming are prominent (Aygün and Demir, 2014; Aygün, 2017).

As in many farming practices, it is necessary to use modern methods for profitable milk production in sheep farming. It is important for sheep breeders to engage in modern production techniques in order to obtain better milk yield from their dairy herds and to earn more profit by producing more profitable milk (Sevinç, 1981).

As it is known, many sheep breeds, especially in certain regions of our country, are raised mainly for milk production, which is used to make highquality cheese. Ewe milk is also processed into other products such as yoghurt and butter. Mutton and wool production comes relatively later. It can be said that there is an increasing demand for sheep cheese and other ewe milk products. However, the increase in ewe milk production is not sufficient and is limited by many factors. This review has been prepared to help sheep breeders what they can do for profitable milk production in sheep husbandry.

STRUCTURAL FEATURES OF SHEEP HUSBANDRY IN TÜRKİYE

The effort to collect the desired features on a sheep according to today's economic needs constantly leads to the emergence of new breeds (Kaymakçı, 2006). Türkiye's native sheep breeds also have different characteristics according to regional distinctions. There are sheep breeds that stand out with their meat yield in some regions, their milk yield in some regions, and their combined yield in most regions. Traditional breeding practices are also common in sheep breeds that stand out with their milk yield. Modern production practices are not sufficient. Today, sheep breeding is mostly done for milk and meat yield. Wool productivity is relatively less. Nowadays, when ewe milk has become more important, breeders prefer sheep breeds with high milk yield. In this regard, it is necessary to give importance to breeding practices in order to obtain the highest level of efficiency from economically profitable breeds.

Türkiye has regions with very different characteristics in terms of climate and geographical conditions. For instance, some regions differ from others in terms of climatic conditions. Eastern Anatolia Region also has a different character from other regions. The emergence of extreme cold and severe storms is the biggest indicator that the winter months have arrived. However, in these seasons, although rarely, warm air waves coming from the north may occur in the high grasslands. However, during these stages, as the grasslands in the higher elevations become warmer, they are likely to receive more rainfall (Sözer, 1972). Despite all this, snow cover in Eastern Anatolia can remain on the ground for months. Under these conditions, the winter period becomes a dead period for Eastern Anatolia. Due to the strong continentality in Eastern Anatolia, the transition from winter to summer is almost sudden. As the cold wave coming from Siberia begins to lose its effect, spring rains begin, then the snow melts and the air warms up rapidly (Tunçdilek, 1978). Small ruminant husbandry activities, which have always had an important place in livestock sector in terms of economy, have been characterized by the prominence of different practices in a way that has adapted to the regional differences of Türkiye.

Raising small ruminants with conventional production in traditional communities is not only a form of economy but also a social and conventional sector, sometimes organized by following stereotyped processes.

In general, as always, small ruminant husbandry has a special place and importance. Small ruminant breeding is one of the most valuable animal husbandry practices in Türkiye, especially in mountainous regions where rural areas are concentrated. Sheep breeding will become increasingly important with the introduction of modern production techniques. According to some research conducted in our country, the predominance is in livestock breeding, especially sheep and goat breeding, which is done on plateaus and pastures (Aygün, 2021).

Meat and milk, as a food, constitute the basic protein sources of animal origin, which cannot be replaced by those of plant origin in human nutrition, in terms of the protein structures, mineral substances and vitamins they contain. Today, the amount of milk consumed per capita is considered an important indicator in determining the nutrition and development levels of countries. Characterization of sheep breeds both at the level of sheep phenotypes and their interaction with milk production systems and at the genetic level is most essential.



Figure 1. A flock of sheep-goat and shepherd on the highland (Photographer, Turgut Aygün).

Local sheep and goat breeds and some genotypes have important advantages such as their adaptation to natural breeding areas, their ability to utilize traditional and special products and areas unsuitable for agriculture. In addition, genetic traits associated with disease resistance, environmental adaptation or productivity are important issues that are emphasized today. In Türkiye, while the characterization of domestic sheep and goat breeds with significant genetic diversity and identification studies in terms of productivity characteristics continue, emphasis is also placed on breeding and protection studies. In terms of studies to be carried out on domestic sheep and goat breeds and genotypes in the country, it is important to carry out inventory determination and recording studies as a priority.

NUTRITION OF MILKING EWES DURING LACTATION PERIOD

Nutrition of ewes is neglected, especially at the beginning of lactation. Essential proteins are rarely provided. Even on good pasture, ewes require supplementary feeding before lambing and during lactation. Care should be taken in feeding dairy ewes during the lactation period. The nutritional level of ewes during the lactation period has important effects on the high and continuity of milk production and the condition of dairy ewes.

Feeding is also effective in the onset of heat and pregnancy after lambing. Low levels of feeding prolong cycles. On the other hand, imbalances in feeding levels, especially during the breeding season, cause fertilized egg deaths. In sheep species, breastfeeding and milking generally prevent the effectiveness of estrus, and the pregnancy rate is low during this period. It is known that ewes that do not breastfeed their lambs have a shorter postnatal anoestrus period than those that do. However, researchers report that milking does not affect estrus, pregnancy and lamb production, provided that balanced and adequate nutrition is available. As a result, it can be said that lactation suppresses estrus, but under certain conditions, lactating or milking ewes can mate, and there is a variation within breeds and breeds in terms of estrus activity during this period (Kaymakçı, 2006).

In cases where the requirements of milk-producing sheep breeds are relatively low, it may be sufficient to feed them with feedstuffs with high cellulose content that are insufficient in terms of nutrients. Condensed feeds, called factory feeds, can also be given in addition to roughage, depending on their quality, content and quantity. Here, quantity and nutritional content, that is, quality, are decisive. However, it is necessary to balance the deficiency and fill the nutritional deficiencies in the context of the needs of the sheep in some specific stages and the calorie, i.e. energy level, provided by the quality forage. A ration should be prepared by providing balanced and high-quality feed content for the animals during the approximately 30-day period before lambing, during the lactation period, before and for 30 days after fertility. Accordingly, the energy ratios of feeds with an ideally balanced nutritional content are as follows (Table 1). Milk yield in sheep generally reaches its peak in the first 6 weeks of lactation and then slows down. In this first period, the nutritional level of dairy sheep is extremely important. The nutritional needs of dairy sheep are guided by the functions that the animals need to perform in their bodies. These vary depending on the stages of the reproductive cycle.

Feed ingredient	Metabolized energy values per kg dry matter
Barley	13.0
Corn	13.5
Soybean meal	12.3
Dry weed	7.5-10.0
Green silage	9-11.5

Table 1. Energy values of some feedstuffs (Kaymakçı, 1986).

THE IMPORTANCE OF HYGIENE AND CONTROL OF DISEASES IN MILKING EWES

The mammary glands of ewes consist of two lobes. Each breast lobe has a nipple and duct. Ewes may have more than two teats. These are mostly in front of the normal nipples. The amount of blood passing through the mammary glands of an ewe in 24 hours can reach approximately 1200 liters. Approximately 350-400 liters of blood must pass through the mammary glands to produce one liter of milk. The average lactation period in sheep is 140-150 days. However, it is known that this period can extend to 8-9 months in some dairy sheep breeds (Kaymakçı, 2006). When fresh milk from healthy animals leaves the udder, only a few microorganisms are present. Studies have shown that there are no microorganisms in the alveoli, up to the milk treasury. In contrast, the teat canal is a bed of microorganisms. This is where the first contamination of milk with microorganisms occurs. Between 100-1000 microorganisms pass into 1 ml of milk from the teat canal. There are almost no bacteria passing from the air into milk. In poorly ventilated shelters, the number of spore-forming bacteria transferred to milk was found to be between 100-200 per 1 ml. Other sources that are effective in bacterial contamination of milk are poorly cleaned udder, milking containers, milk jugs, and washing water used in milking and enterprise (Kaymakçı, 2006).

In these sources of contamination, various microorganisms pass into the milk. However, the type of microorganisms that enter the milk after milking is important as well as the number. The bacterial species seen in the milk of healthy animals are saprophytic bacteria that do not cause any clinical symptoms. The milk of sick animals contains bacteria that cause diseases to humans and animals. An important issue in terms of ewe milking hygiene is that ewe milk is more exposed to fecal contamination than cow milk. Thus, milk contains coli bacteria. Disease-causing bacteria such as Salmonella contaminate milk. For this reason, it is useful to take precautions to prevent fecal contamination during milking period of sheep and goat.

If considered from a health and technological perspective, qualified milk should be understood as follows: Milk with a normal cell count, no pathogenic bacteria, no toxins and harmful chemicals, and a normal taste and smell.

A milk containing these features can only be obtained by knowing and fulfilling hygienic conditions. If we briefly summarize these conditions;

(1) Adequate litter and ventilation must be provided to prevent the absence of carbon dioxide, ammonia and other harmful gases in the milking environment.

(2) Stinky, moldy and spoiled feeds should not be used as they will spoil the bacteriological quality, aroma and smell of the milk.

(3) Milkers must not carry pathogenic bacteria that cause diseases in animals. In addition, the milker must pay attention to all cleaning principles.

(4) During milking, attention should be paid to the cleanliness of the animals' udder, at least the teats should be washed with a disinfectant, and if possible, the first milk should be placed in a separate container. Milking containers should be suitable for the purpose, they should be washed thoroughly after milking and their mouths should be closed. Milkers should use clean aprons to wear only during milking.

(5) After milking, milk should not be kept in the pen, but should be filtered immediately in the filtering chamber.

(6) Cooling is required to slow down the activity of microorganisms in milk and to prevent spoilage to some extent. The simplest is cooling with low-grade water. Milk to be cooled should be placed in jugs in pools filled with cold water, and the water in the pool should be at the level of milk in the jugs. Water should be given from the bottom and drained from the top (Kaymakçı, 2006).

In conclusion, regarding milking hygiene, various difficulties arise in the processing for milk of ewe and goat milked in unhealthy environments into products. This situation directly concerns animal health as well as human health. For this reason, the necessary hygienic precautions before and after milking should be carried out sensitively.

OVERVIEW OF GENERAL PROBLEMS OF SHEEP HUSBANDRY

It is obvious that Türkiye is under the influence of climatological features related to geography and soil structure, and that semi-nomadic or fully nomadic goat-sheep activities continue in most parts of Türkiye, with the influence of its flora.

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Farmer families, who continue their lives by dealing with small ruminant, experience problems and deadlocks in choosing pastures, grazing and wintering places, and obtaining the necessary conditions for this type of livestock farming. Of course, there has been a decline and decline in the rural population in some regions due to the security problem that continues, albeit to a lesser extent, today. Ones living in these mountainous regions had to leave these areas. However, as a result of the recent improvements in Türkiye and the provision of public order, the plateaus and pastures in the Eastern provinces will continue to be of great importance for semi-nomadic or nomadic tribes engaged in sheep and goat breeding, and they will have the opportunity to benefit from them again.



Figure 2. Preparation for milking on the highland (Photographer, Turgut Aygün).

The most convenient way to milk sheep is to reserve a separate section within the pen suitable for milking. In this way, milk yield inspections become easier. Health inspections of sheep and collection of milked milk under hygienic conditions can be ensured. In such pens, sheep should be lined up without being squeezed. It should not be frightened in a way that causes undesirable physiological reactions.



Figure 3. Preparation for milking on the highland (Photographer, Turgut Aygün).

The important thing here is how the sheep will take their place and be kept without fear until the milking is finished. The milking parlor should be large enough and generally above where the milker moves. During milking, ewes are tied in different ways mostly around their necks.

There are many different methods applied in different countries. In some countries, sheep are milked in front of a feed trough at a certain height from the ground. This method also allows the sheep to get used to milking easily. There is enough space for a moving pole to rotate. The milker can sit or stand. Sometimes milkers use a seat that slides on a rail, so they can move it with very little force.



Figure 4. Berivan (female milker) and milking ewes on the highland (Photographer, Turgut Aygün).

Ewes can also be milked manually in special sections called "Kotra", which are very useful. The number of stalls in the farms depends on the number of milkers used. The sheep are herded into a compartment at the back of the cutter and they are taken into the compartments from the back or side of the milkers.

Of course, breeders who raise small livestock have many problems. One of these is the issue of education. Especially the education of girls in the family should be approached from a different perspective. It is often problematic for families who go to high pastures to stay in places called tents. In this way of life, small and young individuals who help the farmer family in all their work contribute to the family economy. In a nomadic or semi-nomadic lifestyle, these individuals play an important role in meeting the basic needs and requirements of the family at every stage. Another thing that should be known here is that in this lifestyle, children generally have to start their education life a little behind. In addition, especially girls face serious problems in transhumance or nomadic lifestyle.

BREED SELECTION IN DAIRY SHEEP HUSBANDRY

Small ruminant breeding is a sector with different dynamics within the livestock sector. Approximately 9% of the milk production obtained at the end of production in our country consists of milk obtained from sheep. Among Türkiye's native breeds with high milk yield, the Awassi sheep breed has a special place and importance. In addition to its increasing importance, it is a local breed that stands out in terms of economic breeding (Yıldız and Yıldız, 2002). Therefore, our country is among the most important countries in terms of hosting various sheep breeds with different characteristics in many aspects. These breeds have adapted to different environmental and care-management conditions. This is also important in that the necessary expenses in terms of cultivation do not require additional costs in broadcasting (Gürsu, 2011; Gürsu and Aygün, 2014).



Figure 5. The end of milking ewes on the highland (Photographer, Turgut Aygün).

The average lactation duration and lactation milk production for Awassi sheep are given as 165.40 days and 110.00 l, respectively. It was revealed that the effect of age and gender on these two characteristics in Awassi sheep was not statistically significant. Similarly, the fat rate, dry matter rate, density, freezing point of milk, and protein rates in milk for Awassi sheep are given as follows, respectively: 6.40%, 11.61%, 1.0364 g/cm3, -0.59 °C and 6.09% (Gürsu and Aygün, 2014).

Milking in sheep husbandry is a seasonal job. It is usually done twice a day, at regular intervals. All our local sheep and dairy sheep are milked. In local breeds, there is a 3-5 month milking period after the lambs are weaned. In dairy breeds, this period may increase to 7-8 months. Milking operations require a significant workforce. In Türkiye, most of the sheep flocks are kept in small flocks. Milk yield levels are also not high. Therefore, milking is naturally done by hand (Gökdal, 1998). On the other hand, in countries where dairy sheep farming is developed, machine milking is becoming increasingly common in large sheep flocks in terms of both labor force increase (savings) and milk and milking hygiene.

Ewe milk, along with goat milk, is among the products preferred by breeders. Goat milk has different properties compared to other milk in terms of its content (Kaymakçı et al., 2005).

CONCLUSION AND SUGGESTIONS

Existing genetic productivity capacities in the livestock sector must be improved through breeding programs and projects. Animal diseases and pests, which are one of the important problems in export activities, should be combated more effectively.

The high input prices in milk production have brought production to a limiting stage. In this regard, support policies should be reviewed and input subsidies should be made. Livestock loan interest rates should also be reduced and maturities extended.

The establishment of modern enterprises that can produce economically profitable products should be supported.

The training of milk producers on modern sheep breeding should be increased to a more effective level.

Organizations of ewe milk producers should be supported.

Unregistered production, which has especially increased recently, should be regularly inspected and necessary sanctions should be applied.

The necessary socio-economic programs should be applied to ewe milk producers. Various measures need to be taken to accommodate the population in rural areas where they are located and to stop the economic deterioration. It is obvious that the problems of business owners producing ewe milk, both in rural areas and in places close to cities, need to be identified and solutions must be sought.

REFERENCES

- Aygün T., Demir, F. (2014). Highland sheep husbandry and zoma life in Hakkari. Uluslararası Mezopotamya Tarım Kongresi, 22-25 Eylül 2014, Diyarbakır, Abstract Book, s: 934-935.
- Aygün, T. (2017). The Occupational Health and Safety for Berivans at Zoma Life in Hakkâri Province (Hakkâri ilindeki zoma yaşamında berivanların sağlığı ve iş güvenliği). Uluslararası Katılımlı 1. Tarım ve Gıda Etiği Kongresi, 10-11 Mart 2017, 163-164, Ankara Üniv. Ziraat Fakültesi, Ankara.
- Aygün, T. (2021). Nomadic activities of small ruminant husbandry in Muş province of Eastern Anatolia in Turkey. XII. International Scientific Agriculture Symposium, "AGROSYM 2021", 7-10 October 2021, Saray Bosna, Bosna Hersek, pp: 1170-1175.
- Gökdal, Ö. (1998). The Milk Yield, Reproductive Performance, Some Body Measurements and Growth Characteristics of Karakaş Sheep (Karakaş Koyunlarının Süt ve Döl Verimleri ile Dışyapı ve Büyüme-Gelişme Özellikleri)," (Doktora tezi). Ph.D. thesis, Department. Animal Science, Yüzüncü Yıl University, Van, Türkiye.
- Gürsu, G. (2011). Characteristics of Milk Yield and Blood Serum Levels of Ca, K, P and Co of Awassi Sheep Maintained at Village Conditions in Gaziantep City (Gaziantep İlinde Köy Koşullarında Yetiştirilen İvesi Koyunlarının Süt Verim Özellikleri ile Kan Serum Ca, K, P ve Co Düzeyleri)," M.S. Thesis, Department Animal Science, Yüzüncü Yıl Univ., Van, Türkiye.
- Gürsu, G., Aygün, T. (2014). Some Characteristics of Milk Yield in Awassi Ewes Maintained at Village Conditions. Journal of Advanced Agricultural Technologies 1(1): 19-23.
- Kaymakçı, M. (1986). Profitable Milk Production (Translation). U.S. Feed Grains Council, England, pp: 15.

- Kaymakçı, M., Tuncel, E., Güney, O. (2005). Türkiye'de süt keçisi ıslah çalışmaları. Süt Keçiciliği Ulusal Kongresi.26-27 Mayıs 2005, Ege Üniv. Ziraat Fak. Zootekni Bölümü, Bornova, İzmir, s: 4-10.
- Kaymakçı, M. (2006). İleri Koyun Yetiştiriciliği. (in Turkish) (İzmir İli DKKYB Yayınları No.1, İzmir).
- Sevinç, A. (1981). Türkiye'de Hayvansal Üretimin Yapısı, Potansiyeli, Geliştirme Olanakları ve Önlemleri. Ankara Üniversitesi, Veteriner Fakültesi Yayınları, 367, Ankara. 85.
- Sezgin, Y. (2006). Bitlis İlinde Göçebe Küçükbaş Hayvancılık Faaliyetleri (yüksek lisans tezi, basılmamış). Yüzüncü Yıl Üniv., Fen Bilimleri Enst., Zootekni Anabilim Dalı, Van, 60 s.
- Sözer, A.N. (1972). Kuzey Doğu Anadolu'da Yaylacılık. Ankara. 128.
- Tunçdilek, N. (1978). Türkiye'nin Kır Potansiyeli ve Sorunları. İstanbul Üniversitesi, 2364, İstanbul. 304.
- Yıldız, A., Yıldız, N. (2002). Milk yield and lactation duration of Awassi ewes raised in ceylanpinar state farm (Ceylanpınar Tarım İşletmesi'nde yetiştirilen İvesi koyunlarının süt verimi ve laktasyon süresi). YYÜ Vet. Fak. Derg., 13: 117-121.



