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RESEARCH ARTICLE

Effects of COVID-19 Pandemic Period on Sheep Breeding

Hilal Tozlu Çelik[1]✉ iD • Hacer Tüfekci[2] iD

[1]Ordu University, Ulubey Vocational School, Department of Food Processing, Ordu/Türkiye
[2]Yozgat Bozok University, Faculty of Agriculture, Department of Animal Husbandry, Yozgat/Türkiye

ABSTRACT

The COVID-19 outbreak experienced worldwide has affected all production areas as well as livestock breeding. This study was aimed at determining the situation of sheep breeders in Ordu province during the COVID-19 outbreak and offering solution suggestions. In the study, data were collected through face-to-face interviews with 72 enterprises registered with the breeding sheep and goat breeders' association in Ordu province in 2023. In the study, it was determined that the breeders were negatively affected by the increase in feed prices (33.3%) during this period. Breeders' reasons for production (88.9%), land use (97.2%), and flock size (88.9%) were unaffected by the epidemic. It was noted that the weaning age of lambs did not change (97.2%) during the epidemic period. The pasture emergence period was not negatively affected during the epidemic period (94.4%). Breeders generally stated that they were not affected by the epidemic period in the use of roughage (84.7%), concentrate feed (83.3%), or vaccination practices (93.1%). Most of the breeders (94.4%) reported that their expectations from animal husbandry did not change after the pandemic period. After the pandemic, it was found that shepherds, fodder support, pasture improvement and health insurance support were more expected (94.4%). Breeders had problems finding a shepherd. In addition, health problems and quarantine practices affected the use of the labor force. Consequently, in order to ensure sustainable sheep production, the expectations and problems of local breeders should be taken into consideration and deficiencies should be eliminated.

1. Introduction

Economic, social, and biological factors experienced worldwide affect the lives of living things. Climate change and epidemics are seen as a significant threat to the sustainability of many species, ecosystems, and livestock production systems around the world (Tüfekci & Tozlu Çelik, 2021). The COVID-19 outbreak of the novel coronavirus (SARS-CoV-2), which has caused a global pandemic in the world, has negatively affected human health since December 2019. It showed its effects as a socio-economic crisis on 11 March 2020. The quarantine and health problems implemented during this period caused all sectors to be negatively affected. Restriction of animal movements, shortage of shepherds, disruption of

breeding activities, and veterinary practices led to a decrease in animal production. Animal welfare was also negatively affected during this period (Guan et al., 2020; Hashem et al., 2020; Vidaurreta et al., 2020). As in many other sectors worldwide, global livestock supply chains experienced a significant disruption due to the emergence of COVID-19 in early 2020 (Hashem et al., 2020; Vidaurreta et al., 2020; Almadani et al., 2022). The COVID-19 pandemic has affected many sectors such as the agricultural, feed, and food industries. Among the main impacts experienced by the agricultural sector, disruptions and delays in breeding activities in both animal and plant production due to the quarantine and restrictions imposed have caused producers to remain below their optimum production levels and thus reduced the income obtained. In

✉ Correspondence
E-mail address: hilalcelik@odu.edu.tr

addition, difficulties in the marketing of agricultural products, the decline in people's purchasing power, and the reduced ability of households to access affordable food have also affected the economic resilience of farmers and breeders (Faturokhman et al., 2022). Within livestock breeding, especially cattle, sheep, and goat breeding, marketing problems were undergone during the COVID-19 outbreak, which led to decrease in farmers' incomes. Although there was a decrease in purchasing power, data showed that the demand for processed livestock products during the pandemic period increased 3-4 times compared to the pre-pandemic period. This is due to a shift in consumer preferences for products with health and food safety quality standards, which points to a great opportunity for increased sales of products for beef cattle, sheep, and goats (Faturokhman et al., 2022). Food supply is a linear system in which one stage is fed by the other. It also includes many interconnected organizations, individuals, and businesses that make up this system and shape its functioning. The vulnerability of food supply and the need for transformative change are influenced by numerous shocks and stressors affecting food supply chains, such as climate change and the recent COVID-19 pandemic (Thornton et al., 2014; Garnett et al., 2020; Payne-Gifford et al., 2022; Bozma et al., 2023). Limited access to veterinary services and treatments has restricted the monitoring of animal health, resulting in a negative impact on animal welfare. This has posed risks for the spread of infectious animal diseases, human health, and food safety (Hashem et al., 2020; Mtimet et al., 2021). Problems with the supply chain have led to an increase in meat prices (Almadani et al., 2022). A study conducted in Australia reported that animal welfare is a shared responsibility during the COVID-19 pandemic and that a multi-sectoral approach to animal welfare during the crisis is therefore required (Baptista et al., 2021). It has become imperative to develop effective methods and technologies to protect animal health, human health, and food safety (Haque et al., 2021). To protect animal welfare during crises, nations should consider: national risk assessment, open communication channels, emergency plans for animal welfare, crisis response groups, and support systems for animal care providers. It is important to inform other countries to ensure that animal welfare is not jeopardized during unforeseen events (Baptista et al., 2021). The effects of global warming and the COVID-19 pandemic are dual challenges affecting high-quality livestock development (Hao et al., 2023). Challenges such as labor shortages have also emerged (Meuwissen et al., 2021).

Sheep and goat breeding is an effective animal husbandry activity that provides income in areas where active agricultural production cannot be carried out and prevents migration to cities. Pasture areas, natural vegetation, geographical characteristics, and the cultural and socio-economic structure of Türkiye are very suitable for sheep breeding. Both in the world and in Türkiye, small ruminant livestock is the most important

form of utilization of land where crop production cannot be carried out (Akçapınar, 2000). The Black Sea Region has a suitable structure for sheep breeding due to its mountainous, hilly land structure, climate, and socio-economic structure. The lack of sufficient land in the region, especially the presence of stony, sloping, and hilly lands, allows sheep and goat breeding as alternative animal husbandry. Sheep breeding is a part of the Black Sea ecology. It is an important animal husbandry activity for the region and Ordu province in terms of providing income during every period of the year (Tozlu Çelik, 2016). The rugged and inadequate land in the region severely limits the opportunities for the people living here to engage in different agricultural activities. For this reason, sheep breeding constitutes an important source of income for the people of this region in terms of having the potential to provide income continuously throughout the year and utilizing inefficient lands (Tozlu Çelik, 2016; Alkan & Türkmen, 2021). In this study, it was aimed at determining the effect of the COVID-19 outbreak on sheep breeders in Ordu province and making recommendations.

2. Materials and Methods

The Ordu province is the third-largest city in the Eastern Black Sea Region of the Black Sea Region. The provincial territory lies between 40°18' and 41°08' north latitudes and 36°52' and 38°12' east longitudes. The surface area of the province is 5.952 km^2. It has a topographically rugged and mountainous region, is dominated by the Black Sea climate. Hazelnut orchards dominate the province from the coastline up to an altitude of 1000 meters, and there are also various field lands and forests. There are pastures and plateaus in areas above 1000 meters. It is reported that the pasture potential of Aybastı, Gölköy, Kabadüz, Korgan, Kumru, and Mesudiye districts is suitable for the development of cattle and sheep breeding (T.C. Tarım ve Orman Bakanlığı, 2014).

Permission to conduct the study was obtained with the decision of the Ordu University Social Sciences and Humanities Research Ethics Committee dated March 2, 2023, and numbered 2023-36. For this study, data were collected by a face-to-face survey method in 2023 with 72 enterprises out of 1729 enterprises that are members of the Ordu Province Breeding Sheep and Goat Breeding Association (T.C. Tarım ve Orman Bakanlığı, 2023). In similar survey studies, it was reported that at least 3% (Yamane, 2010) or 10% (Sümbüloğlu & Sümbüloğlu, 2000) of the population would be sufficient to determine the sample size. In this study, data were collected from sheep breeders by face-to-face survey study survey using simple random sampling method. The evaluation of the obtained data was analyzed with the SPSS (26.0 Version) statistical program for analyze, descriptive statistics, and frequencies (SPSS, 2016). The results of the data collected within the research framework are presented as percentages and frequencies.

3. Results and Discussion

3.1. Socio-Economic Status of Breeders

The livestock sector in Türkiye is one of the sectors most affected by global climate change due to the predominance of rural economic structures and developing livestock-based industries. Health and welfare in animal production are integral parts of environmental sustainability (Tüfekci & Tozlu Çelik, 2021). Since sheep production in Türkiye is predominantly small-scale and pasture-based, the animal products obtained constitute the main food source for agricultural enterprises, and accordingly, the income from sheep production is generally at low levels (Şen et al., 2023).

The data on the socio-economic status of sheep breeders in the study are given in Table 1. According to the findings obtained, it was determined that 48.6% of the sheep breeders lived in the village, 44.4% in the district, and 7% in the province, and the majority of the sheep breeders were males (94.4%), and most of the respondents were in the middle age group of 41-60 years old (54.2%). The number of breeders with 1-4 households was 63.9%, and nuclear family structures constituted the majority. The level of education of the breeders with at least a primary school education was found to be 97.2%. Those who make a living only by farming are 80.5%, and the majority (90.3%) are property owners and nomadic sheep breeding is practiced in Ordu province.

In another study conducted in Ordu province, 59.21% of the 76 farmers surveyed were primary school graduates, 14.47% were secondary school graduates, 22.36% were high school graduates, and 3.95% were undergraduate graduates (Ateş & Çam, 2022). It was reported that the level of education did not make a difference in production, management, or additional business ventures (p = 0.139). In the same study, the level of education, experience, age, and moonlighting status of the breeders did not affect flock size, the number of lambs obtained per ewe, lamb survival, and enterprise management in sheep enterprises. It was reported that sheep breeders in Ordu province mostly carry out hazelnut production and animal production together, and some breeders participate in production with a small number of animals as a hobby because they cannot break away from their traditions (Ateş & Çam, 2022). It has been reported that there are very few female flock owners among the breeders; the average age of the flock owners is around 49, and the young generations are not very keen on sheep breeding (Ateş & Çam, 2022). In the study conducted to determine the effects of the COVID-19 pandemic on agricultural food production and farmers in Konya, the average age of agricultural workers was 53.95, and 91.3% of them were male. Of the respondents, 55.3% were primary school graduates; 68.7% had an income above their expenses; 72.0% had a nuclear family; 55.3% were engaged in dry farming; 90% were breeders; and 72.7% worked mainly in the summer months. As a result of the study, it was reported that 88% of those working in the agricultural sector were concerned about harvesting and sales and that these concerns were mostly (35.6%) due to price instability (Uğur & Buruklar, 2022). In our country, factors such as the low level of education and income of ovine breeders in general and breeding by very small family enterprises with production techniques that continue from father to son cause problems in the input supply of enterprises and the marketing of products (Sarıca et al., 2004; Ceyhan et al., 2015a, 2015b; Kandemir et al., 2015). The education, age, and gender findings obtained in the study were found to be similar to those reported in the literature (Ateş & Çam, 2022; Uğur & Buruklar, 2022).

Table 1. Data on socio-economic status of sheep breeders.

Residence	n	%	Age	n	%
Province	5	7.0	19-40 years	24	33.3
District	32	44.4	41-60 years	39	54.2
Village	35	48.6	Age 61 and over	9	12.5
Gender			**Occupation**		
Female	4	5.6	Farmer	58	80.5
Male	68	94.4	Public-private sector	14	19.5
Education			**Household**		
Illiterate	2	2.8	1-4 persons	46	63.9
Primary education and above	70	97.2	5-7 people and above	26	36.1
Cultivation Type			**Ownership Status**		
Nomad	72	100.0	Owner	65	90.3
			Tenant-partner	7	9.7

3.2. Breed of Sheep and Flock Size

The most common sheep breed in Ordu province is the Karayaka breed. It is important for animal breeders to carry out studies on the pure breeding of the Karayaka breed and increasing its productivity in the province. The Karayaka breed has varieties with different live weights depending on care and feeding. In Ordu province, it has been observed that there are Karayaka sheep reaching 40-45 kg live weight towards the coast and 30 kg live weight in high areas. It is estimated that this situation might be due to care and feeding. Karayaka sheep breed ranks second after Kıvırcık breed in terms of meat quality in Türkiye (Tozlu Çelik, 2016).

Data on sheep breeding practices are given in Table 2. The majority (84.7%) of the breeders stated that they do this work as the only source of livelihood, and 83.3% of them stated that they want to continue animal husbandry in the future. While more than half (51.4%) of the breeders stated that the purpose of production was to produce breeding, 31.9% for butchering, and 16.7% for sacrificial animals, 70.8% stated that they provided additional feeding to animals in certain periods. While 51.4% of the breeders reported that the breeding period of ewes was 6 years or more, 84.7% stated that the breeding period of rams was 1-4 years. In addition, 66.7% of the breeders stated that they used shepherds and had difficulty finding a shepherd.

Table 2. Data on some breeding practices in enterprises-1.

Reason for Keeping Livestock	n	%	How Many Years Have You Been Keeping Livestock?	n	%
Sole source of income	61	84.7	1-20 years	37	51.4
Additional source of income	11	15.3	More than 20 years	35	48.6
Purpose of Production			**Future Livestock Production Status**		
Butchery	23	31.9	Yes	60	83.3
Sacrifice	12	16.7	No	12	16.7
Breeding	37	51.4			
			Duration of Use of Females in Breeding		
Shepherd Status			1-5 years	35	48.6
Yes	48	66.7	More than 6 years	37	51.4
No	24	33.3			
			Duration of Use of Rams in Breeding		
Supplementary Feeding			1-4 years	61	84.7
Yes	51	70.8	More than 5 years	11	15.3
No	21	29.2			

3.3. Breeding Practices

When Table 3 is analyzed, it is seen that 87.5% of the breeders have a herd size of 50–200 heads, and 12.5% have a herd size of 200 heads or more. The weaning age of lambs is mostly 3-6 months (79.2%). It was determined that health controls were generally carried out by public veterinarians (70.8%). It was observed that vaccinations were performed by veterinarians. In 48.6% of the enterprises, it was determined that the breeder could also vaccinate himself and animal losses were higher (63.9%) in the winter months when cold temperatures were experienced. Improvement of pen conditions and care and feeding in winter conditions can prevent, especially the loss of offspring.

Ateş and Çam (2022) reported in their study in Ordu province that 59.27% of the enterprises had less than 100 heads of sheep, and the average number of animals per enterprise was 102 heads. As a result of the study, it was recommended that those with less than 100 sheep per enterprise should be encouraged to increase the number of animals, and the problem of finding a shepherd should be solved in order to reach a more

profitable, innovative, and sustainable situation for sheep farming enterprises in Ordu province.

Sheep breeding in Ordu province is generally carried out in rural areas with small flocks in the family enterprise style. In the breeding studies to be carried out in order to increase sheep yields, it is necessary to determine the current potential of the animals and to decide which breeding programs will be applied. For this reason, the records kept in the enterprises of the breeders are of great importance (Tozlu Çelik, 2016). In addition to the existing subsidies, it is important to promote sheep products and to provide subsidies to producers for the processing and marketing of the products obtained at a good price. In addition to the old animal shelters, the lack of knowledge of shepherds on issues such as herd management and animal diseases causes animal losses. The most important problem in sheep breeding enterprises is labor and feed expenses. In addition to these problems, the fact that the owners do not have sufficient knowledge about lamb growth and fattening and that they generally provide breeding from within the enterprise is among the general breeding problems (Ceyhan et al., 2015a, 2015b).

Table 3. Data on some breeding practices in enterprises-2

Herd Size	n	%	Vaccination Status	n	%
50-200	63	87.5	Yes	63	87.5
250 and above	9	12.5	No	9	12.5
Lamb Weaning Age			**Vaccination Person Vaccinated**		
2 months	11	15.3	Veterinarian	37	51.4
3-6 months	57	79.2	Breeder	35	48.6
More than 6 months	4	5.5	**Mother's Milk Intake in The First Week**		
			Yes	72	100.0
Health Control			**Season with High Animal Losses**		
Public veterinarian	51	70.8	Spring	26	36.1
Private veterinarian	21	29.2	Winter	46	63.9

3.4. Effects of COVID-19 Pandemic Period on Sheep Farming Enterprises

In the study, the data obtained from the questionnaire conducted to determine the impact of sheep breeders during the COVID-19 pandemic period are given in Table 4. In the study, it was stated that the breeders were negatively affected by the increase in feed prices (33.3%) during the epidemic period. The finding of increased feed costs in this study is similar to that reported by Mtimet et al. (2021). Breeders' reasons for production (88.9%), land use (97.2%), and flock size (88.9%) were not affected by the pandemic. The weaning age of lambs did not change (97.2%) during the pandemic and the period of pasture emergence was not negatively affected (94.4%). Breeders generally highlighted that they were not influenced by the epidemic period in the use of roughage (84.7%), concentrate feed (83.3%), or vaccination practices (93.1%). Most of the breeders (94.4%) reported that their expectations from animal husbandry did not change after the pandemic period. After the pandemic, it was determined that the expectations of shepherds, feed support, improvement of pastures, and health insurance support were higher (94.4%). Breeders had problems finding shepherds. Health problems and quarantine practices affected the use of the labor force. In general, 52.8% of sheep breeders in Ordu province stated that they were not affected by the COVID-19 outbreak. This finding was similar to that reported by Uğur and Buruklar (2022).

During the epidemic period, national and international restrictions on health control practices and budgetary constraints caused problems in the detection and control of animal diseases. During the COVID-19 pandemic, many logistics systems, such as livestock production systems and their supply chains, faced supply disruptions. In particular, there was a problem of access to agricultural inputs (such as animal equipment, vaccines, etc.) (Hashem et al., 2020). Studies have reported that the sales of farmers' crops and livestock products were adversely affected primarily due to quarantine practices during the COVID-19 outbreak (Jaacks et al., 2021). COVID-19 caused a decrease in the labor force during the pandemic period. Especially the problem of finding shepherds became more prominent during this period (Hashem et al., 2020; Uğur & Buruklar, 2022). The problem of difficulty in finding a shepherd in this study is similar to the ones reported in the literature (Biswal et al., 2020; Hashem et al., 2020; Uğur & Buruklar, 2022).

Due to the land and crop production structure of Ordu province, the fact that ovine breeding enterprises are small, widespread, and dispersed in the form of enterprises prevents the breeders from reaching the desired income in the marketing of the products. In addition to this, the fact that ovine breeders do not act together in the marketing stage of the products they obtain and the operating expenses of the ovine breeders cause many intermediaries between sellers and buyers, and this situation leads to economic problems. The fact that almost all of the available land is hazelnut, untimely, excessive, and irregular grazing of meadow and pasture areas, problems related to the supply of roughage, the problem of finding a shepherd, the lack of milk collection, and cold chain facilities in enterprises can be counted as general breeding problems (Tozlu Çelik, 2016).

During the pandemic period, restrictive measures did not have any impact on the parameters of sheep farming systems. In some studies, it was reported that the outbreak period negatively affected only the daily working routine of the breeders in enterprises with high sheep numbers (Yiakoulaki et al., 2022), while there are studies reporting problems in the raw material supply chain, finding laborers, and marketing of products (Biswal et al., 2020). In this study, the finding of not being affected by the COVID-19 outbreak as a general condition is similar to that reported by Yiakoulaki et al. (2022).

Table 4. Data reflecting the affected status of sheep breeders from COVID-19 pandemic period

Affected by COVID-19	n	%	Herd Size Affected?	n	%
Not affected	38	52.8	Not affected	64	88.9
Encouraged to keep livestock	3	4.2	Affected	8	11.1
Existing conditions became more difficult	7	9.7	**Was Pasture Access Affected?**		
Feed cost increased	24	33.3	Not affected	68	94.4
			Affected	4	5.6
Reason for Production Has It Been Affected?			**Has The Use of Roughage Been Affected?**		
Not affected	64	88.9	Not affected	61	84.7
Affected	8	11.1	Affected	11	15.3
Was Land Use Affected?			**Has The Use of Concentrated Feed Been Affected?**		
Not affected	70	97.2	Not affected	60	83.3
Affected	2	2.8	Affected	12	16.7
Did It Affect Weaning Age?			**Vaccination Affected?**		
Not affected	70	97.2	Not affected	67	93.1
Affected	2	2.8	Affected	5	6.9
Expectation After The Pandemic			**Has The Expectation Changed After The Pandemic?**		
Shepherd support	2	2.8	Yes	4	5.6
Feed support	2	2.8	No	68	94.4
Shepherd, feed, pasture improvement and health insurance support	68	94.4			

In a study conducted in Niğde province, it was aimed to examine the structural characteristics and biosecurity practices of sheep farms. As a result of the study, it was recommended to train breeders and implement strategic programs against diseases and other factors affecting production on sheep farms. In the same study, it was reported that the preparation of strategic programs for hygiene practices in sheep breeding against possible diseases may be an important support for future epidemic disease periods (Şen et al., 2023).

In a study conducted in Türkiye, it was reported that some of the negative effects on meat prices were compensated as a result of live animal imports to reduce the effects of uncertainty due to the COVID-19 outbreak. In order to secure access to lamb meat during the pandemic period, it has been suggested to improve price stability, support it through tax exemptions, carry out live animal sales through the live animal exchange, and establish digital sales channels (Mikail & Kaplan, 2021; Bozma et al., 2023).

4. Conclusion

The results of the study showed that flock size, pasture access, roughage, and concentrate feed use were not affected during the COVID-19 period in sheep breeding enterprises in Ordu province. During this epidemic period, it was reported by the breeders that the cost of feed increased and it was difficult to find a shepherd. It was determined that sheep breeders demanded especially feed, shepherd, pasture improvement, and health insurance support after the epidemic.

As a result, it is important to complete the deficiencies by taking into account the problems experienced during the COVID-19 epidemic period in order not to disrupt production and to ensure food safety. Taking precautions not only for the pandemic but also for changing climatic conditions is necessary for sustainable sheep breeding. Strategic plans should be made and more studies should be carried out so that sheep breeding is not adversely affected by epidemics and climatic changes. There is a need for self-sufficient animal food production planning at the national level.

Compliance with Ethical Standards

Ethics committee permission for this study was received by the decision of Ordu University Social and Human Sciences Board dated March 2, 2023 and numbered 2023-36.

Conflict of Interest

The authors declare that they have no conflict of interest.

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RESEARCH ARTICLE

Determination and Pathogenicity of *Verticillium dahliae* Isolates Obtained from Tomato Plants (*Solanum lycopersicum* L.) in the Iğdır Province

Tuba Genç Kesimci✉ ⓘD

Iğdır University, Faculty of Agriculture, Department of Plant Protection, Iğdır/Türkiye

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ABSTRACT

This study was carried out to identification of Vegetative Compatibility Groups (VCGs) and pathogenicity of *Verticillium dahliae* isolates obtained from tomato plants in Iğdır province. As a result of survey studies conducted in 18 different regions, 14 isolates were obtained from 629 diseased tomato plants. In the complementation test, the seven isolates were found as VCG2A and VCG2B by using international reference isolates. VCGs of other isolates were not identified. Assessment of the aggressiveness of the KRS-2, YC-13, YY-14, and MLK3-4 isolates was evaluated on tomato (cv. Super). The disease severity was between 15-45% and MLK3-4 isolate had the highest disease severity (45%). Additionally, the effect of temperature on the growth of *V. dahliae* isolates (TSD-1, MLK3-4 and YY-14) was determined. The isolates showed optimal growth temperatures ranging from 20 to 25 °C (except for TSD-1). In control of fungal diseases, it is very important to describe the disease and pathogen. Therefore, the results of the study are important for tomato growers and researchers.

1. Introduction

Tomato (*Solanum lycopersicum* L.) is one of the most popular vegetables all over the world (Schreinemachers et al., 2018; Famuyini et al., 2020). The tomato plants are attacked by many biotic and abiotic disease during the production stage which it is known to be threatened by more than 200 diseases (Panno et al., 2021). Fungi constitute the most significant group of plant pathogens that causes economic damage in tomato. Specially, soil-borne fungal pathogens cause important yield loss in tomato (Yucel et al., 2008). Verticillium wilt caused by *Verticillium dahliae* Kleb. is a soil-borne pathogen serious threat to tomato cultivation areas all around the world. The

pathogen causes disease in 14 families from 4 different continents (Inderbitzin et al., 2011), and it causes billions of dollars of damage to various agricultural products (Pegg & Braddy, 2002; Klosterman et al., 2008; Inderbitzin et al., 2011). Although *Verticillium isaacii* Inderb. et al., *Verticillium nonalfalfae* Inderb. et al., *Verticillium tricorpus* Isaac and *Verticillium zaregamsianum* Inderb. et al. are known to cause vascular wilt diseases (Inderbitzin & Subbarao, 2014), *V. dahliae* is the most important and common species of the *Verticillium* genus on tomato (Li et al., 2022).

Initial symptoms of Verticillium wilt diseases on tomato include yellowing of the lower leaves, wilting and stunted

✉ Correspondence
E-mail address: tuba.genc@igdir.edu.tr

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growth (Kumar et al., 2018). The yellowing between the veins of the leaves with increasing wilt gradually becomes dry and brown (Senthamilselvi & Victoria, 2019). Generally, one side of the leaves remains green while other parts show typical wilt symptoms (Fradin & Thomma, 2006; Gazozcuzade, 2010), and causes V-shaped lesions on lower leaves. In the later stages of the disease, yellowing and stunting start from the lower leaves and gradually progresses to the upper leaves. Brown discoloration observed in the vascular tissues in the part close to the soil surface (Fradin & Thomma, 2006; Kumar et al., 2018).

Vegetative compatibility is used to determine the similarities and differences in the population of a species (Leslie, 1993; Burgess et al., 2009). The determination of Vegetative Compatibility Groups (VCGs) is approaching effectively for examine the genetic structure of soil-borne pathogens (Bao et al., 1998) and this method is commonly used in the genetic characterization of *V. dahliae* (García-Carneros et al., 2014). *V. dahliae* obtained from different hosts were determined five VCGs as VCG1, VCG2, VCG3, VCG4 (Joaquim & Rowe, 1990) and VCG6 (Bhat et al., 2003). VCG1 (VCG1A, VCG1B), VCG2 (VCG2A, VCG2B) and VCG4 (VCG4A, VCG4B) groups were categorized as two subgroups (Klosterman et al., 2009). VCGs of *V. dahliae* isolates obtained from various hosts have been studied by many researchers in Türkiye. This is known as VCGs; VCG1, VCG2A, VCG2B and VCG4B from cotton and cotton seeds (Göre, 2007; Göre et al., 2014), VCG1A, VCG2A, VCG2B and VCG4B from eggplant (Dervis et al., 2009), VCG1, VCG2A, VCG2B and VCG4B from chrysanthemum (Göre, 2009), VCG2B and VCG4B from weeds (Demirci & Genc, 2009), VCG1A, VCG2A and VCG4B from olive (Dervis et al., 2010), VCG2B and VCG4B from potato (Dane & Demirci, 2012), VCG2A, VCG2B and VCG4B from strawberry (Genc Kesimci & Demirci, 2020).

The tomato production is one of the most significant agricultural activities for growers in Iğdır province. It has been determined that there are important fungal diseases both in the survey studies carried out and in the diseased samples brought by the farmers. Therefore, studies were planned to identify fungal diseases that cause disease in tomato plants. We conducted this study: (i) to determination of *V. dahliae* isolates obtained from the tomato plants in Iğdır province (ii) to assess VCGs diversity of isolates *V. dahliae* (iii) to determine the virulence of isolates with pathogenicity test and (iv) to determine the effect of temperature on the growth of *V. dahliae* isolates.

2. Materials and Methods

2.1. Survey Studies

Survey studies were conducted in the tomato field of the Iğdır province and districts between August and November 2014-2016. Diseased tomato plants were randomly collected

from in the field (5-6 steps). The plants were taken in plastic bags and transported to the laboratory.

2.2. Isolations from Tomato Plants

Tomato plants were brought to the laboratory were washed with tap water and cleaned from the soil. For isolations, the root and stem tissues were cut in 1.5 cm size, surface sterilized in 2% NaOCl solution for 2-3 minutes, and then rinsed in sterile water 3 times. The plant parts were dried on the sterile paper for 15-20 minutes and were transferred 4 pieces (2 stems and 2 root collars) on each Petri plates (9 cm diameter) onto Water Agar (WA; 15 g agar per liter) amended with 50 mg L^{-1} streptomycin sulfate. The plates were incubated at 25 °C for 3-7 days. The colonies were examined under the microscope and conidia were transferred for pure growth with a sterile needle on WA or Potato Dextrose Agar (PDA). The isolates were stored +5 °C for use in the study.

Serial dilution method used for single spores. Firstly, isolates were grown at 25 °C for 7-10 days. Sterile water (1000 µL) was transferred in the first eppendorf tube (2 mL), and 900 µL sterile water was transferred in the 2nd, 3rd and 4th eppendorf tubes. A mycelial disc (5 mm diameter) of fungus grown on PDA was transferred in the first eppendorf tube. This eppendorf tube was vortexed for 1 min and then 100 µL conidial suspension was transferred from tube 1 to tube 2. After the eppendorf tube 2 was vortexed for 1 minute, 100 µL of taken was transferred from tube 2 to tube 3. The eppendorf tube 3 was vortexed for 1 min and the tube 3 containing spore concentration prepared as 1×10^3. The spore suspension (1000 µL) was poured into an empty the Petri plate (9 cm) and on top of this was added PDA slightly colder than the normal pouring temperature. The Petri were developed at 25 °C for 2-3 days and this pure colony was transferred to PDA. The isolates were kept in the slant test tubes +5 °C (Genc, 2012).

2.3. Morphological and Molecular Characterization

Verticillium dahliae (TSD-1, MLK3-4 and YY-14) isolates were grown on WA and PDA for morphological observations. The isolates were observed under the microscope and determined according to conidia and microsclerotia (Goud et al., 2003; Jabnoun-Khiareddine et al., 2010; Inderbitzin et al., 2011).

In the molecular test, two isolates (YY-14 and TSD-1) were selected. The isolates were grown to be used in DNA isolation in the Petri plates containing PDA 7-10 days at 25 °C and were identified using ribosomal DNA (rDNA)-ITS (Internal Transcribed Spacer) regions (ITS1, 5.8, ITS2) using ITS1 and ITS4 primers were performed by REFGEN (Ankara University Teknokent, Ankara, Türkiye). Genomic DNA isolation, PCR amplification using ITS1 and ITS4 primers (White et al., 1990), sequence analysis performed as described by Genc Kesimci et al. (2022).

2.4. **Generation and Characterization of *nit* Mutants**

Previous method of Korolev and Katan (1997) was modified for the determination VCGs of *V. dahliae* isolates. In this method, 30-50 g L^{-1} potassium chlorate ($KCIO_3$) and 0.2 g L^{-1} glucose were added to 17 g L^{-1} Corn Meal Agar (CMA) or 15 g L^{-1} WA. *Verticillium dahliae* isolates were grown on PDA and 5 mm mycelial discs taken from the isolates were transferred to each Petri plate (9 cm) as 6-8 pieces on Corn Meal Chlorate Agar (CMCA) or Water Agar Chlorate medium (WAC). The Petri plates were kept at 25 °C for 1-2 weeks. Each isolate was made in two replications. After the incubation period, fast and thin-growing sectors resistant to $KCIO_3$ were transferred to Czapex Dox Agar (CDA) and were determined as *nit* mutants. Then, sodium nitrate (0.5 g L^{-1}) ($NaNO_3$), sodium nitrite (0.5 g L^{-1}) ($NaNO_2$) and hypoxanthine (0.2 g L^{-1}) were added separately to CDA (45.4 g L^{-1}) and the phenotype of *nit* mutants were classified according to aerial mycelium growth in the presence of different nitrogen sources (Correll et al., 1987; Korolev & Katan, 1997).

2.5. **Complementation Tests with Tester Isolates**

Complementation tests were maintained on CDA using a method modified from Korolev and Katan (1997) and Korolev et al. (2000). All of the *nit* mutants obtained in this study with *nit* mutants of the international *V. dahliae* tester isolates were compared (*nit*1x*nit*M or *nit*Mx*nit*M) 1 to 1.5 cm apart on plates at least twice, and plates were incubated for 14-28 days at 25 °C in the dark. The complementations were characterized by growth at the contact zone between the two complementary *nit* mutants (Korolev et al., 2000).

2.6. **Effect of Temperature on the Mycelial Growth**

The optimal temperature of isolates *V. dahliae* was determined using a method modified from Bhat et al. (2003). TSD-1, MLK3-4 and YY-14 isolates were grown on PDA (9 cm) and were taken from 7-day-old colonies of each isolate mycelial plugs (5 mm in diameter) that transferred to PDA (9 cm). The petri were incubated at temperatures ranging from 5, 10, 15, 20, 25, 30 and 35 °C. Three replicates of each isolate and the sizes of the colony growth of each petri were measured as mm 7th, 14th and 21st days.

2.7. **Pathogenicity Test**

KRS-2, YC-13, YY-14 and MLK-3-4 isolates were selected for pathogenicity test. All isolates were cultured on PDA for 15 days. Surface disinfection of tomato seeds (cv. Super) were made in 70% alcohol for 3 min, rinsed with water three times and air-dried on filter paper. For each isolates were sown with 3 seeds in each pot (15-cm-diameter) filled with autoclaved torf/perlit soil mixture (2:1, v v^{-1}). The seeds were reduced to one seedling in each pot. The concentration of conidia in the suspension was adjusted to 10^6 conidia ml^{-1} using a hemocytometer. The tomato plants (2-3 leaf) were inoculated by flooding the soil around the roots with 10 ml spore suspensions for each isolate (Gong et al., 2017). The control plants were inoculated with sterile distilled water (10 ml). Hoagland solution added three times each 15 days (10, 20 and 30 mL). Plants were grown at 25±1 °C with a photoperiod of 24 h for 75 days. Five pots of tomato were used for each isolates and control. The scale was used according to the modified 0-4 scale of Hunter et al. (1968). The disease severity were calculated according to Townsend Heuberger's formula. As a result of pathogenicity test, were measured fresh weight and height of all tomato plants. Dry weight of the plants was calculated after drying at 80 °C for 48 h (Isaac et al., 2018). Re-isolations were made to complete Koch's postulate with the procedure of isolation. The data obtained as a result of study were subjected using the Statistical Package for the Social Sciences (SPSS) version 17.0 ($p<0.05$).

3. Results

3.1. **Survey, Isolation and Identification**

In this study, surveys were conducted from districts and villages in Iğdır province (Akyumak, Bayraktutan, Centre, Gölbaşı, Hakmemet, Küllük, Melekli, Obaköy, Yaycı, Yukarıçarıkçı, Kasımcan, Kuzugüden, Özdemir, Sarıçoban, Yüzbaşılar, Karakoyun, Taşburnu and Aralık). Isolations were performed from sypmtomatic 629 tomato plants. A total of 14 isolates of *V. dahliae* were obtained from the plants (Table 1). *Verticillium dahliae* could not be isolated from other regions except Aralık, Melekli, Taşburnu, Yukarıçarıkçı and Yaycı. The majority of the isolates were collected from stems (8 isolates), and remaining the isolates were isolated from root collar. *Verticillium dahliae* were also occured in the complex with other pathogens as *Rhizoctonia* spp. and *Fusarium* spp. which were significantly higher observed.

In morphological characterization, length and width of conidia and microsclerotia of *V. dahliae* isolates were measured (Table 2). Conidia arise singly at the apices of the phialides, irregularly sub-cylindirical, ellipsoidal. All of the isolates produced verticillate conidiophore. Microsclerotia is very variable in shape (irregular and elongated shaped or more spherical and scattered). Chlamydospores were not observed. Colonies has white then varied from white to black with the produced of microsclerotia during the 6-7 day incubations.

Table 1. Isolate code and location of *Verticillium dahliae* isolates obtained from root collar and stem of tomato plants in Iğdır province.

Species	Isolate code	Location	Plant colonization	
			Root collar	Stem
Verticillium dahliae	KRS-2	Aralık		+
	MLK1-D1	Melekli		+
	MLK1-D2	Melekli		+
	MLK1-D5	Melekli		+
	MLK3-D3	Melekli	+	
	MLK3-D4	Melekli	+	
	MLK3-D8	Melekli	+	
	TSD-1	Taşburnu	+	
	TS-ZE-8	Taşburnu	+	
	YC-1	Yukarıçarıkçı		+
	YC-13	Yukarıçarıkçı		+
	YC-14	Yukarıçarıkçı		+
	YC-16	Yukarıçarıkçı		+
	YY-14	Yaycı	+	
Total			6	8

Table 2. Isolate code, conidia-microsclerotia length and width of *Verticillium dahliae* isolates.

Species	Isolate code	Conidia length/ width*(µm)	Microsclerotia length/ width*(µm)
Verticillium dahliae	TSD-1	3-10 (5.7)/1.5-4.0 (3.0)	15-80 (37.4)/16-44 (28.8)
	MLK3-D4	3-7 (5.1)/1.5-3.5 (2.4)	17-70 (37.3)/16-40 (27.1)
	YY-14	3.5-10 (6.3)/1.5-4 (2.5)	15-59 (30.5)/13-45 (30.1)

*: Minimum-Maximum (Average).

In the molecular characterization, *V. dahliae* isolates (YY-14 and TSD-1) were performed by sequence analysis of the ITS regions of the ribosomal DNA (rDNA). BLAST analysis of the sequences obtained in the study was performed in NCBI, and the base sequences of the isolates were registered in the GenBank. The Accession number of TSD-1 and YY-14 was obtained as OR734907 and OR734908 (Sequence similarity of 100%), respectively.

3.2. Generation and Characterization of *nit* Mutants

Colonies that showed thin and expansive growth on CDA were determined as *nit* mutants. A total of 24 *nit* mutants from the nine isolates were obtained and phenotypes were identified (Table 3). Most mutants classified as *nit*1 (18). *nit*3 mutants could not be obtained. The other five isolates were not obtained *nit* mutant despite being repeated numerous times.

Table 3. Number of *nit* mutants and types of *Verticillium dahliae* isolates.

Isolate No	*nit*1	*nit*M	Total
MLK1-D1	1	-	1
MLK3-D3	-	2	2
MLK3-D4	2	-	2
MLK3-D8	2	1	3
TSD-1	3	1	4
TS-ZE-8	2	1	3
YC-1	5	-	5
YC-16	2	-	2
YY-14	1	1	2
Total	18	6	24

3.3. Vegetative Compatibility Groups

In the VCGs test, some of the isolates giving '+' reaction (Figure 1a) with reference test isolates and were included in a group. MLK1-D1, MLK3-D4, MLK3-D8, TS-ZE-8, YC-1 and YY-14 isolates were compatible with reference tester isolates (VCG2B Q115 and 39/5) and determined as VCG2B. Also, some of the isolates made '+/-' reaction (Figure 1b) with other groups. The weakly cross-reactions were observed between VCG2B (MLK3-D4, MLK3-D8, YC-1 and YY-14) and tester isolate of VCG2A PH. Similarity, VCG2A isolate (TSD-1) and all of the VCG2B isolates were made'+/-' with VCG4A BB, except VCG2B MLK3-D4 isolate. Two isolates (MLK3-D3 and YC-16) were incompatible and '-' reaction (Figure 1c) with tester isolates and VCGs could not be determined (Table 4).
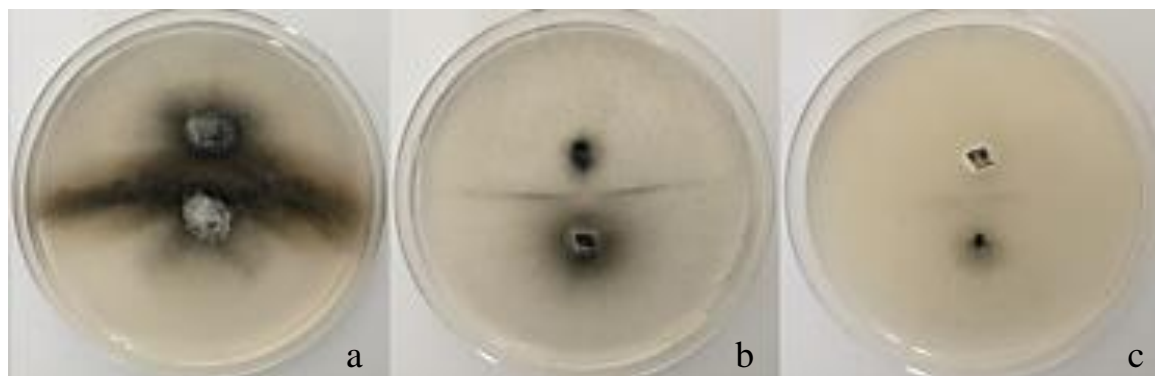
Figure 1. The degree of complementation; (a) '+' reaction; (b) '+/-' reaction; (c) '-' reaction.

Table 4. The reaction with test isolates of isolates *Verticillium dahliae*.

Isolate code	Mutant type	Tester isolates							VCGs
		VCG2A	VCG2B		VCG4A		VCG4B		
		PH	Q115	39/5	P103	BB	S39	MT	
MLK1-D1	1	-	+	+	-	+/-	-	-	2B
MLK3-D3	M, M	-	-	-	-	-	-	-	-
MLK3-D4	1, 1	+/-	+	+	-	-	-	-	2B
MLK3-D8	M, M	+/-	+	+	-	+/-	-	-	2B
TSD-1	M, 1	+	-	-	-	+/-	-	-	2A
TS-ZE-8	M, 1	-	+	+	-	+/-	-	-	2B
YC-1	1,1	+/-	+	+	-	+/-	-	-	2B
YC-16	1,1	-	-	-	-	-	-	-	-
YY-14	M, 1	+/-	+	+	-	+/-	-	-	2B

3.4. Effect of Temperature on the Colony Growth of Isolates *Verticillium dahliae*

TSD-1, MLK3-4 and YY-14 isolates were investigated for growth rate at seven temperatures (5 to 35 °C) *in vitro* conditions. The growth characteristics of the three isolates varied during the 1, 2 and 3-week incubation. The isolates showed optimum growth temperatures, ranging from 20 to 25 °C (except for TSD-1). None of the isolates *V. dahliae* grew at 5 °C and 35 °C for 7, 14 or 21 consecutive days (Figure 2).
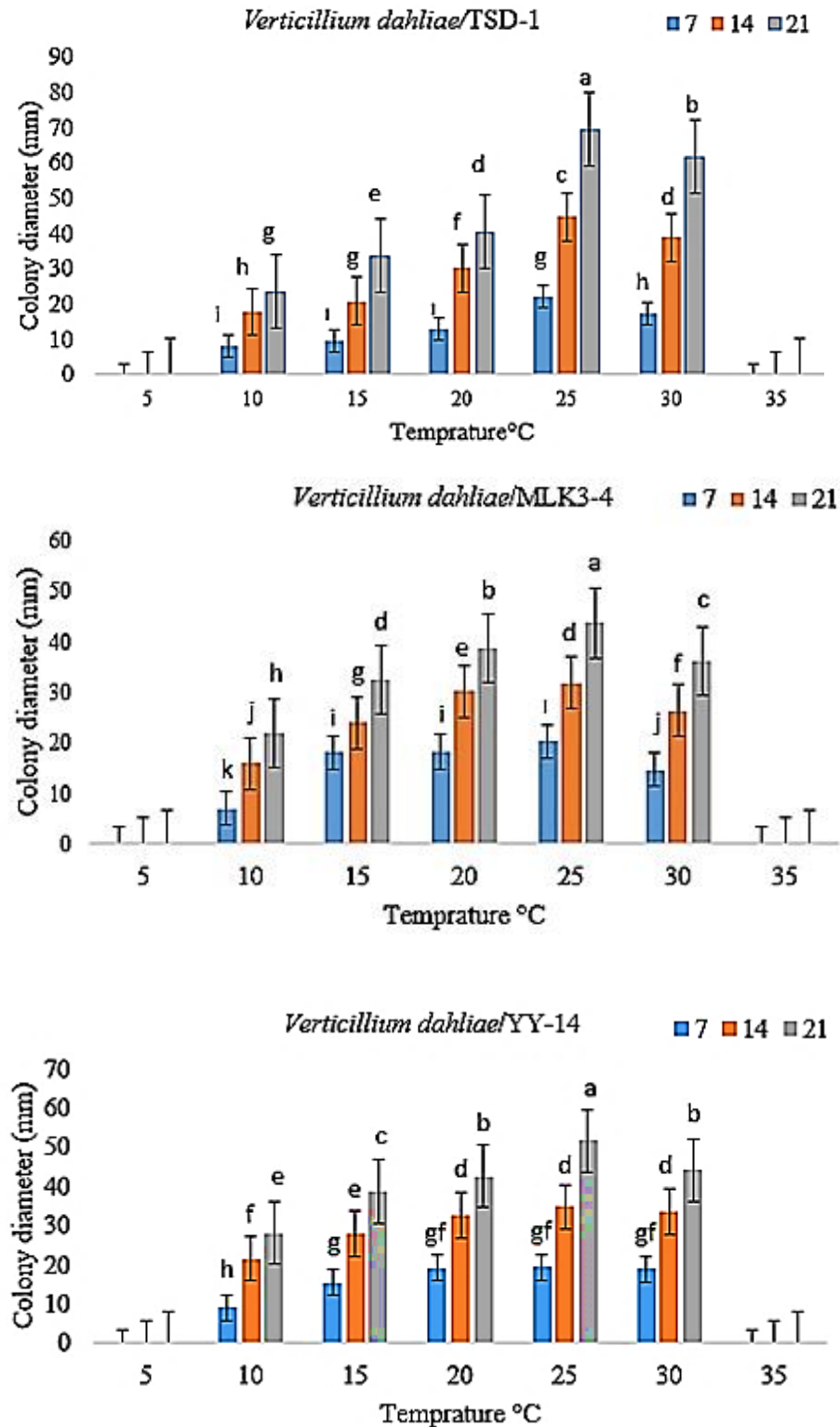
Figure 2. Colony growth of *Verticillium dahliae* isolates at different growth temperatures.

3.5. Pathogenicity Test

Pathogenicity test was established with 4 isolates selected from *V. dahliae* isolates. As a result of the pathogenicity test of isolates *V. dahliae* was determined on differential virulence to the tomato plants. MLK3-4 isolate had the highest disease severity (45%); however, the KRS-2 isolate had the lowest disease severity (15%). The effects of all isolates on tomato plants (height, root length and fresh weight) were found to be statistically insignificant (Table 6). The control plants were observed any disease symptoms.

Table 6. Results of pathogenicity test of *Verticillium dahliae* isolates.

Species	Isolate codes	Height* (cm)	Root length (cm)	Fresh weight (g)	Dry weight (g)	Disease severity (%)**
Verticillium dahliae	KRS-2	37.4±6.36	18.8±3.36	8.1±1.14	1.1±0.19b	15±6.12b
	YC-13	37.8±3.74	17.4±0.92	8.4±1,04	1.3±0.15ab	35±6.12a
	YY-14	45.6±0.81	15.6±1.28	7.4±1.20	1.7±0.13a	40±10.0a
	MLK-3-4	39.8±1.39	19.4±2.52	8.8±1.49	1.7±0.10a	45±5.00a
	Control	35.6±3.64	16.0±1.51	7.1±0.66	1.1±0.10b	0±0.0b

*: Mean+/-standard error: Different letters indicate the difference between the averages (Duncan test, $p<0.05$).
**: Disease severity was calculated as a percentage on a 0-4 scale using the Tawsend-Hauberger formula.

4. Discussion

The determination of disease-causing pathogens is necessary for an effective control strategy and reduction of pathogen damage (Martinelli et al., 2015). In the current study, the presence of *V. dahliae* in tomato plants was investigated in Iğdır province and the pathogen was determined as associated with Verticillium wilt of tomato. The morphological identification is the first step in the classification of fungal pathogens (Sunpapao et al., 2022). *Verticillium* spp. have a resting structure characteristic as dark resting mycelium, chlamydospores and microsclerotia. The structures play an significant role both in the biology and taxonomy of the genus and was characteristic in distinguishing the species (Inderbitzin, 2011). All isolates obtained from this study showed the general characteristic features of *V. dahliae* by producing verticillate conidiophore and microsclerotia in morphological determination. The morphological identification was also confirmed by molecular identification of the selected isolates in the study.

Genetic variation is a significant concept in the life history of asexually reproducing plant pathogens. VCGs are the most effective method in the determination of the genetic relationships in *V. dahliae* population (McDonald & Linde, 2002). Also, knowledge of the variation within the pathogen groups is essential for improve the control strategy (Collado-Romero et al., 2008). VCGs test were applied to *V. dahliae* isolates obtained in this study and the VCGs of the isolates were determined as VCG2A and 2B which VCG2B isolates were isolated more frequently from tomato plants. The VCG2B is considered likely to be present in any *Verticillium* soil (Zeise & Von Tiedemann, 2002). Similarly, VCG2B is reported to be the most common group isolated from a variety of hosts (Bhat & Subbarao, 1999; Korolev et al., 2000). VCGs associated with tomato have been determined as VCG2A, VCG2B, and VCG4B at the International level. Different from our study, some studies were reported that VCG4B is the dominant group in tomato in South Africa, Greece and Israel. On the other hand, they showed that VCG2B had a limited distribution (Korolev et al., 2000; Papaioannou et al., 2013; Retief et al., 2023). Also, Akar et al. (2024) reported that VCG1A isolates are dominant group from grafted tomatoes in Antalya, Türkiye.

In this study, pathogenicity of one isolate was found to be different in levels of virulence of the other isolates used in the pathogenicity test. Other studies have shown that *V. dahliae* isolates have different virulence in various hosts (Tjamos, 1981; Joaquim & Rowe, 1991; Tsror et al., 2001; Sanei et al., 2008). A study conducted that by Genc Kesimci and Demirci (2020) were determined the disease severity between 27.5% and 75% on strawberry. Tjamos (1981) found that were differences found in the pathogenicity of tomato. Retief et al. (2023) reported that the aggressiveness of *V. dahliae* isolates varied between 2.08% and 51.94% on tomato. There are two races of *V. dahliae* infections on tomato, race 1 carries the avirulence gene VdAve1 and race 2 is not carrying (Panno et al., 2021). Fradin et al. (2009) observed that Ve gene provides resistance against race 1 of *V. dahliae* isolates. Therefore, it is considered that virulence differences may occur, especially in tomato. Also, the ability toxin production of isolates are very important (lead to necrosis and wilting) which the toxic metabolites and/or phytotoxins may induce plant defense mechanisms (Zhen & Li, 2004). Genetic exchange between isolates belonging to the same VCGs may lead to the emergence of more virulent isolates as it will provide diversity (Bhat et al., 2003; Jiménez-Díaz et al., 2006). At the same time, large number of host species may be effect the degree of virulence (Pegg & Brady, 2002).

The growth and distribution of fungi are affected by different environmental factors (Alsohaili & Bani-Hasan, 2018). Temperature is one of the most important environmental factors affecting the development of fungal diseases. In this study, temperature effect on radial growth of isolates *V. dahliae* was investigated *in vitro*. The colonies of *V. dahliae* well grown on PDA at 20-25 °C, and the optimal temperature was 25 °C. *Verticillium dahliae* not grown at 5 °C and 35 °C. These results were similar to with previous studies. Pegg and Brady (2002) has been reported that the optimal growth temperature of isolates as 22 and 25 °C. Jing et al. (2018) also found that the colony growth of isolates *V. dahliae* increased at 25 °C. Rampersad (2010) demonstrated that *V. dahliae* was grown at 25 and 30 °C, but not at 15 and 35 °C. In addition, Jabnoun-Khiareddine et al. (2006) found that *V. dahliae* was more greater disease higher temperatures (21 to 30 °C) than the lower

temperatures (17 to 21 °C) and adapted to higher temperatures in tomato plants.

5. Conclusion

Verticillium dahliae is one of the most common pathogens that causes yield and quality losses in tomato worldwide. In this study, we investigated to VCGs and pathogenicity of *V. dahliae* isolates obtained from tomato plants in Iğdır province. As a result of the study, VCGs were determined as VCG2A and VCG 2B. Also, variation in the pathogenicity of the isolates was observed. With the data obtained, a source was created for further studies to be carried out in tomato plants. The management of Verticillium wilt recommends control methods such as soil solarization, soil fumigation, and crop rotation, but the methods are inadequate to manage of the disease. Developing pathogen-specific control strategies such as selection of resistant varieties can contribute to more successful results.

Acknowledgment

Conflict of Interest

The author has no conflict of interest to declare.

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PRENSİP

RESEARCH ARTICLE

Effect of Parity and Pasture Period on Milk Lipid Composition in Awassi Sheep

Ülkü Dağdelen[✉] [ID] • Doğan Türkyılmaz [ID] • Nurinisa Esenbuğa [ID]

Atatürk University, Faculty of Agriculture, Department of Animal Science, Erzurum/Türkiye

ARTICLE INFO	ABSTRACT
	This study aimed to explore how pasture periods and parity impact the lipid profiles of 38 Awassi sheep raised in a traditional grazing system on natural pastures. Milk samples were gathered for lipid profile analysis at the commencement, midpoint, and conclusion of the pasture period. Data analysis employed repeated measurements through two-way ANOVA within the General Linear Model (GLM) framework. The statistical model encompassed the effects of pasture periods, parity, and their interactions. Although parity did not affect lipid profiles, the pasture period had a significant effect ($p<0.01$) on triacylglycerol (TAG), free fatty acid (FFA), and monoacylglycerol (MAN). A significant effect ($p<0.05$) on cholesterol (COL) was also observed. Furthermore, neither pasture period nor parity had any effect on phospholipids. As a result of the study, it was determined that parity had no effect on the lipid profile in milk, while pasture period had a very significant effect on triacylglycerol, free fatty acids, monoacylglycerol and a significant effect on cholesterol.

1. Introduction

Sheep farming holds a significant place in livestock organization. The selection of sheep breeds varies according to countries' climate conditions, consumption preferences, and cultural structures (Kaymakçı & Sönmez, 1996; Türkyılmaz, 2014; Dagdelen & Esenbuga, 2022). Türkiye ranks very high in the world in terms of the number of sheep. However, Türkiye numerical superiority is not valid in terms of productivity per animal (Epstein, 1985; Özbey & Akcan, 2000; Biçer et al., 2019). Sheep farming activity, which is an important part of animal production activities worldwide, is of great importance both economically and in terms of human health in Türkiye as well. Especially in the Central and Eastern Anatolia Region,

sheep farming is quite common, and the climate and pasture structure in this region make sheep farming an important source of income (Keskin et al., 2007; Özyürek, 2017; Türkyılmaz et al., 2021). Throughout history, sheep have played an important role in the economic and social life of Turkish communities both in nomadic migrations from Central Asia to Anatolia and in settled life (Özbey & Akcan, 2000; Biçer et al., 2019). It is of the utmost importance to ensure the nutritional needs of sheep are met at all times (Şahin et al., 2003). The performance of sheep in terms of fattening is influenced by both genetic and environmental factors. Among these environmental factors, the feeding system plays a significant role. Sheep are unable to fully express their genetic potential for fattening when raised under inadequate feeding systems (Keskin et al., 2010).

✉ Correspondence
E-mail address: ulku@atauni.edu.tr

Additional feeding or pasture periods are employed to ascertain the genetic potential of the subject. The pasture structures in Türkiye and the ongoing consumption habits of the people emphasize the importance of sheep products for healthy nutrition. Furthermore, the potential to create job opportunities with little capital further highlights the socio-economic importance of sheep farming.

The Awassi breed is a fat-tailed sheep breed raised in the Southeast Anatolia region. This breed adapts excellently to the hot climate conditions of the region and has the highest milk yield among native breeds. Awassi sheep, known for their flocking instinct and excellent walking ability, are recognized worldwide as a valuable sheep breed (Konar et al., 1991; Macit & Aksoy, 1996; Özyürek, 2017).

The components of sheep milk vary depending on various environmental factors such as genotype, parity, season, regional climate conditions, lambing interval and time, type of birth, care and feeding conditions, and udder structure lead to changes in the composition of sheep milk (Kesava Rao et al., 2003). However, generally, sheep milk contains 18.8% dry matter, 7.5% fat, 5.6% protein, and 4.6% lactose. These differences are typically most pronounced proportionally in milk fat (Park et al., 2007; Yılmaz et al., 2011).

Triglycerides are known as fat molecules that meet the body's energy needs and help maintain body temperature balance. They also support intracellular nutrient intake and body nourishment by participating in the structure of the cell membrane. Triglyceride levels can be measured through blood tests (Bitman & Wood, 1990). Normally, triglyceride levels are considered 150 mg/dl, with values between 150-199 mg/dl being borderline, values above 200 mg/dl considered high, and 500 mg/dl or above considered very high. Triglycerides are formed by the esterification of nutrients absorbed from the intestines in the liver.

Cholesterol, with its soft, fat-like structure, is one of the milk lipids found in all cells of the body. Cholesterol plays an important role in the production of cell membranes and some hormones necessary for a healthy body (Costa et al., 2009; Contarini & Povolo, 2013). However, high cholesterol levels are considered a key risk factor for coronary heart diseases such as heart attacks and strokes. The body obtains cholesterol from two sources: synthesis within the body and intake through food. While the liver and other cells produce most of the blood cholesterol, the remainder comes from food. Cholesterol, which is one of the building blocks of nerve cells, is also required for the synthesis of vitamin D and sex hormones (Kaynar et al., 2013). Cholesterol obtained from foods is hydrolyzed and absorbed by pancreatic cholesterol enzymes in the small intestine. It constitutes approximately 0.25-0.45% of total lipids in milk and is mostly present in esterified form with lipids. Cholesterol is transported in the blood by fat-protein complexes; low-density lipoproteins (LDL) carry cholesterol to tissues, while high-density lipoproteins (HDL) facilitate its return transport (Manlongat et al., 1998). LDL cholesterol, when present in excessive amounts in the bloodstream, can increase the risk of heart attacks and strokes and is often associated with the consumption of foods containing saturated fat, trans fat, and cholesterol. It can lead to the formation of thick and hard plaque known as atherosclerosis (Güldür et al., 2007).

The amounts of cholesterol obtained from animal-derived foods vary. It is 110 mg/dl in beef, 160 mg/dl in sheep meat, 1602 mg/dl in egg yolks, 3100 mg/dl in the brain, 375 mg/dl in kidneys, 88 mg/dl in cheese, and 19 mg/dl in sheep milk. Although phospholipids found in milk fat are present in small amounts, they perform important functions in the body. Phospholipids, especially those found in high concentrations in the brain, are known as molecules containing amino acids and fatty acids. Phospholipids are naturally present in all cells of the body, especially in the brain (Wat et al., 2009). Additionally, phospholipids have many functions related to tissues and organs, but the most important is their role in forming the building blocks of the brain, playing a key role for billions of cells (Turkyilmaz & Esenbuga, 2022).

2. Materials and Methods

The study was conducted at the Food and Livestock Application and Research Center of Atatürk University. In the study, 38 sheep of Awassi breeds were used. The sheep were divided into two groups, comprising those in the 3th (n=18) and 4th (n=20) parity. The sheep were grazed on the pasture in from May to September. Milk samples were collected on May (beginning of pasture), July (middle of pasture) and September (end of the pasture) for understanding the variation in milk lipid profile within each period is crucial for herd management and for guiding future research endeavors. During the pasture period, dry meadow grass obtained from the Plant Production Application and Research Center was also given to the animals ad libitum along with drinking water. In addition to pasture, the sheep were fed concentrate feed (13% crude protein, 8.70% crude cellulose, 2% crude fat, 9% crude ash, 0.35% sodium, 0.20% magnesium, Vitamin A 15 000 IU, Vitamin D3 3 000 IU and Vitamin E 30 mg/IU) at 1.5% of their live weight.

Control milking was conducted on the sheep used to determine to lipid profiles. These milking were initiated after the lambs were weaned and continued until the end of the pasture season. Control milking was started between 19:00 and 19:30 after the sheep returned from pasture and continued for approximately one hour. Before milking, the sheep's udders were cleaned with a moist cloth. Samples were taken at the beginning, middle, and end of the pasture season during the control milking to determine milk components and lipid profiles. These samples were taken to the Animal Science Department's Milk Analysis Laboratory at Atatürk University.

2.1. **Determination of Lipid Profile**

Lipid profiles, including triacylglycerol (TAG), phospholipid (PL), free fatty acid (FFA), monoacylglycerol (MAG), and cholesterol (COL), were analyzed by high-performance thin-layer chromatography (HPTLC) HPTLC plates were scanned at high resolution, and the Rf values were automatically calculated using TotalLab 1D (TL120) software, and the classification of lipids in milk samples was made (Figure 1). The area covered by each lipid spot was determined using a densitogram in TotalLab 1D (TL120) program (Figure 2), and the lipid class in total milk fat was expressed as a percentage (Kaynar et al., 2013).
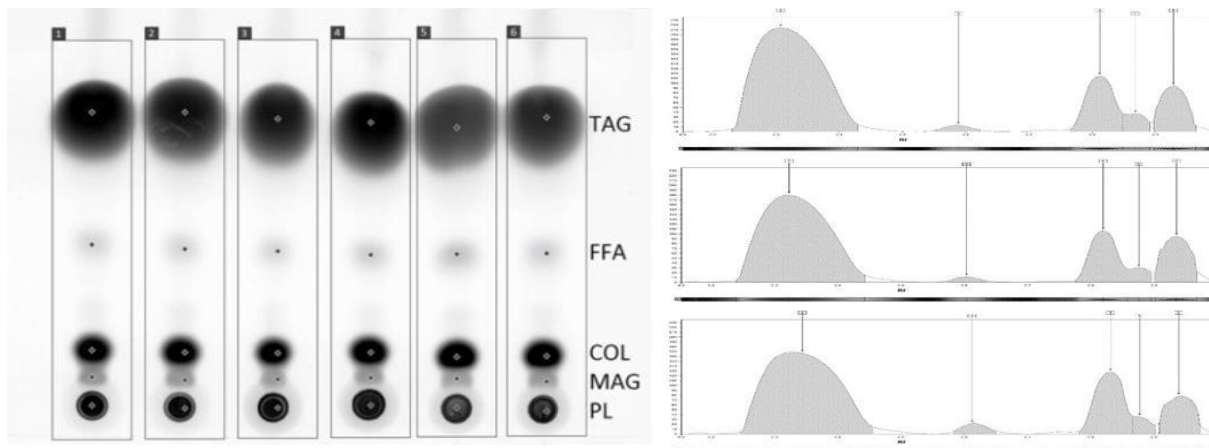
Figure 1. HPTLC image and densitogram.
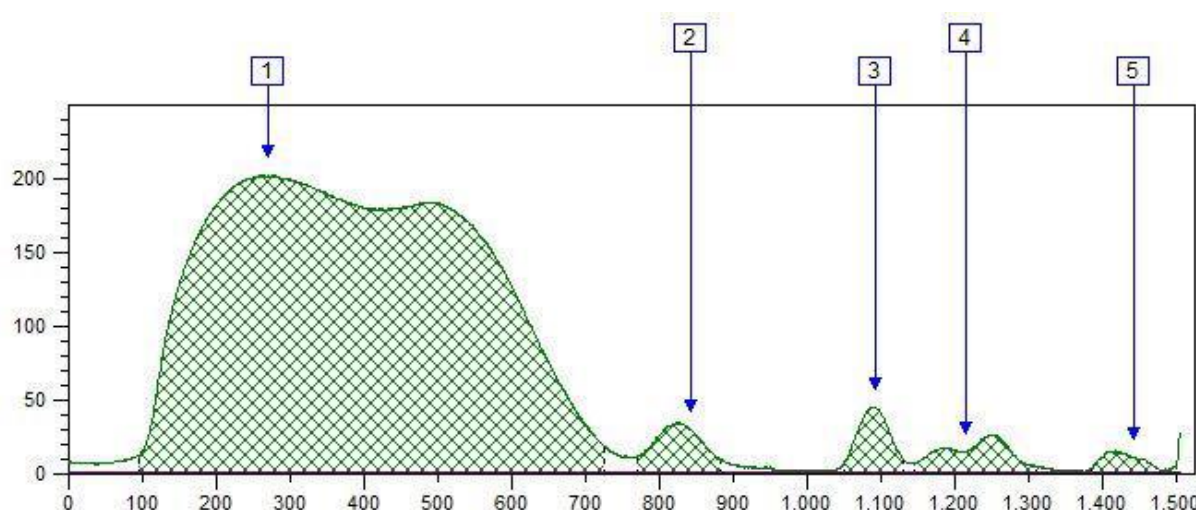
Figure 2. Densitogram image of lipid profiles in milk (1: Triacylglycerol, 2: Free fatty acids; 3: Cholesterol; 4: Monoacylglycerol; 5: Phospholipid).

2.2. **Statistical Analysis**

The data underwent analysis as repeated measurements via two-way ANOVA within the General Linear Model (GLM) procedure (SPSS Inc., Chicago, IL, USA). The mathematical model incorporated the effects of pasture periods and age, as well as all interactions.

3. Results and Discussion

The lipid profiles of milk from Awassi sheep are outlined in Table 1. Research on the lipid composition of sheep milk remains limited, but it's noted that lipids in sheep and goat milk are predominantly found in globules smaller than 3.5 μm, which is beneficial for digestibility and efficient lipid metabolism compared to cow's milk fat (Miller et al., 2006; Park et al., 2007; Kaynar et al., 2013; Merlin Junior et al., 2015).

The major lipid components in sheep milk include triglycerides (TAG), free fatty acids (FFA), cholesterol (COL), monoacylglycerols (MON), and phospholipids (PL). TAG, with a complex composition, primarily resides in the core of milk globules, constituting nearly 90% of the lipids observed in this study. It was unexpected that parity did not appear to affect the content of sheep milk in terms of TAG, FFA and MON. However, the pasture period was found to exert a highly

significant effect (p<0.01) on these ratios. It's noteworthy that the TAG content observed in this study was lower than what was reported by Miller et al. (2006) for sheep milk.

It was observed that the cholesterol (COL) content in Awassi sheep milk was significantly higher, with a notable difference (p<0.05) noted. Furthermore, the pasture period was found to have a significant effect (p<0.05) on the COL levels. Specifically, the COL ratio was initially high at the beginning of the pasture period, gradually decreasing significantly as the period progressed towards the midpoint and end stages.

Phospholipids (PL) constitute a portion of the milk fat globule membrane, comprising approximately 0.2-1% of all milk lipids. They play a pivotal role in this membrane due to their highly surface-active properties. Some studies propose that PL may possess anti-cancer and anticholesterolemic functions. PL has been associated with tumor suppression by influencing cell proliferation and is acknowledged as a highly bioactive compound with bacteriostatic and cholesterol-lowering properties. Notably, the phospholipid (PL) levels in milk from both age groups were similar. The observed differences were not attributed to age but rather to the influence of varying pasture periods. It is noteworthy that the phospholipid (PL) levels in milk from different age groups exhibited similarity. The observed discrepancies were not attributable to age but rather to the impact of different pasture periods. Phospholipids (PL) are integral to the milk fat globule membrane, comprising 0.2-1% of all milk lipids and playing a crucial role due to their highly surface-active properties. Some

studies suggest PL may possess anti-cancer and anticholesterolemic functions, with reported tumour-suppressing properties by influencing cell proliferation and exhibiting bacteriostatic and cholesterol-lowering effects.

The PL level in milk from both age groups was similar, with differences not attributed to age but rather to various pasture periods. However, the effect of the pasture period on PL was not observed in this study. Additionally, the PL levels obtained were lower than those reported by some researchers (Boersma et al., 1991).

While no effect of age on free fatty acids (FFA) and monoacylglycerols (MON) was observed, a highly significant effect (p<0.01) of the pasture period on these variables was noted. The FFA and COL values were higher than those reported by Miller et al. (2006) but TAG and PL values were lower than those reported by Bitman and Wood (1990).

Lipids, along with other milk components, exhibit variability due to genetics, physiology, and environmental factors such as season. In this study, the pasture period emerged as crucial due to seasonal and climatic changes affecting the nutritional composition of animal feed and, consequently, animal physiology, thereby influencing milk quality. Seasonal variations in the lipid profile of milk during pasture periods have been documented in numerous studies. It was observed that the age and pasture period were not significant on lipid profiles (Lu et al., 2018).

Table 1. Lipid profiles of Awassi sheep milk (± standard error).

	N	TAG	FFA	COL	MON	PL
General Average		89.67±0.571	2.79±0.278	2.85±0.155	3.61±0.187	1.06±0.055
Parity		ns	ns	ns	ns	ns
3	18	89.35±0.676	2.99±0.357	2.98±0.219	3.64±0.186	1.04±0.084
4	20	90.67±0.624	2.33±0.329	2.64±0.202	3.28±0.172	1.07±0.078
Period of Pasture		**	**	*	**	ns
Beginning	16	87.22[b]±0.699	3.81[a]±0.369	3.25[a]±0.227	4.58[a]±0.193	1.12±0.087
Middle	10	91.52[a]±0.886	2.06[b]±0.467	2.66[b]±0.287	2.75[b]±0.244	1.01±0.111
End	12	91.29[a]±0.808	2.10[b]±0.426	2.50[b]±0.262	3.06[b]±0.223	1.04±0.101

ns: Non-significant; **: P<0.01, *: P<0.05; TAG: Triacylglycerol; FFA: Free Fatty Acids; COL: Cholesterol; MON: Monoacylglycerol; PL: Phospholipid.

4. Conclusion

Most parameters exhibited higher values at the end of the pasture compared to the beginning, indicating a notable change over time. The reason why the pasture period has a significant effect on triacylglycerol, free fatty acids, cholesterol, monoacylglycerol and phospholipid is thought to be due to the fatty acid conformation in the vegetative process of the plants in the pasture. Conversely, there were no discernible differences observed in milk composition and lipid profiles across different parities of sheep. This study addresses this gap

by highlighting the significant impact of pasture on lipid profiles rather than parity. Thus, to potentially manipulate milk composition and lipid profile, which are crucial for human nutrition, further research is recommended.

Compliance with Ethical Standards

The study protocol was approved in advance by Atatürk University Local Ethics Committee of Animal Trials with the decision number 49 dated 27.02.2015.

Conflict of Interest

The authors declare that they have no conflict of interest.

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RESEARCH ARTICLE

Larval Development of Penguin Tetra (*Thayeria boehlkei*): Morphological Observations

İhsan Çelik [ID] • Pınar Çelik[✉] [ID]

Çanakkale Onsekiz Mart University, Faculty of Marine Science and Technology, Department of Aquaculture, Çanakkale/Türkiye

ARTICLE INFO

ABSTRACT

In this study, the larval development stage of the penguin tetra fish (*Thayeria boehlkei*), a popular species in the freshwater aquarium fish industry, was investigated. For this purpose, penguin tetra larvae were morphologically observed from time to hatching until they reached the juvenile stage. Throughout the larval development phase, samples were randomly taken daily and captured on a camera using a stereomicroscope. Larval development stages were categorized into four periods: (I) Yolk-sac larva, (II) Pre-flexion larva, (III) Flexion larva, and (IV) Post-flexion larva. Larvae were fed with Artemia nauplii until they reached the juvenile stage for the entire research. Morphological changes, including the state of the yolk sac from hatching, mouth opening, changes in body shape and color, and fin formation processes, were recorded daily. According to the findings, the mouth opened on the 5th day, and external feeding also started on the 5th and 6th days. At the same time, free swimming movements were observed in the larvae, and it was determined that the yolk sacs were consumed. The flexion larva stage occurred on the 15-16th days. The larval development process of the penguin tetra fish was completed within 25 days after hatching. After this stage, the juveniles became morphologically identical to their parents.

1. Introduction

The global aquarium fish trade has turned into a multi-billion-dollar industry and continues to grow every year. According to a report published three years ago, the global aquarium fish market size was valued at USD 4.5 billion in 2020 and is expected to reach USD 6.3 billion by 2028 (Grand View Research, 2021).

Tetra fish, with more than 150 species, are among the important fish groups that are commercially valuable and popular among hobbyists (Helfman et al., 2009). Penguin tetra is in high demand by aquarium enthusiasts due to its

characteristics such as schooling behavior, unique colorful body patterns, small size, and adaptability to planted aquariums (Axelrod, 1996). It is a popular species among freshwater aquarium species (Weitzman & Palmer, 1997).

The penguin tetra (*Thayeria boehlkei*) is a freshwater fish species native to the Amazon basin, belonging to the Characidae family (Weitzman & Palmer, 1997). In nature, this species is distributed in rivers with dense vegetation (Géry, 1977). Penguin tetras are among the popular species in the aquarium sector due to their attractive colors and swimming behaviors (Axelrod et al., 1967). The popularity of this species in the aquarium trade can put pressure on natural fish stocks.

✉ Correspondence
E-mail address: pinarcelik@comu.edu.tr

To alleviate this pressure on natural populations and make this business sustainable, it is necessary to develop aquaculture practices (Monticini, 2010).

Morphological examination of the larval development of this tetra species is important for understanding the reproductive biology of the species and increasing the success in aquarium fish farming. The larval development stage is a critical period from hatching until the transition to the juvenile stage. During this period, newly hatched larvae undergo morphological and physiological changes until they become juveniles (Fuiman & Werner, 2009). The changes that occur at this stage are directly related to the survival and growth rates of the larvae (Houde, 1987).

In our literature review, we noticed that information on the larval development of penguin tetras is limited. No detailed scientific study on this subject has been found. Understanding the morphological changes during the larval development stage in every aspect will help us better understand the reproductive biology of the penguin tetra and increase the efficiency of the techniques applied in the cultivation of such species (Nakatani et al., 2001). In addition, the findings obtained in this way will allow a better understanding of the larval development of other species in the Characidae family and enable interspecies comparison (Oliveira et al., 2008).

The aim of this study is to examine the larval development stage of penguin tetra fish morphologically from hatching until the transition to the juvenile stage. Within the scope of the study, penguin tetra larvae were photographed daily from the first day they hatched until they reached the juvenile stage. The findings obtained enabled the definition of the larval development stage of this species and the determination of the morphological changes that occur during this period.

2. Materials and Methods

2.1. Broodstock Management and Larval Rearing

Mature individuals of the species, aged over one year, were utilized for the controlled reproduction in laboratory conditions. Female and male broodstock were maintained separately in 40 L glass aquaria (dimensions: 40 cm length × 30 cm width × 35 cm height; water level: 32-33 cm). All broodstock were fed a commercial aquarium diet (Tetramin Granulat, Tetra, Germany; composition: 46% protein, 12% oil, 3% fiber, 11% ash, 8% moisture) three times in a day. A photoperiod of 12 hours light and 12 hours dark was maintained, with lights on from 07:00 to 19:00. Water parameters in the broodstock tanks were kept constant at a temperature of 24±0.5 °C, pH of 6.0-6.5, and conductivity

between 100-200 μs, using 100-watt aquarium heaters for temperature regulation. For spawning, three females and three males were randomly selected from the broodstock aquaria and transferred to 15 L glass production tanks. Spawning occurred overnight, and the eggs were deposited on the bottom of the tank. After spawning, the broodstock were removed, and the eggs were incubated in the production tank for larval development monitoring. The eggs were maintained at a constant water temperature of 24±0.5 °C.

2.2. Larval Sampling

From the first day of hatching until the juvenile stage, random samples of larvae were collected daily. Larvae were observed using an Olympus SZX7 zoom stereomicroscope (Tokyo, Japan) and photographed with a connected video camera (Q Imaging, Micropublisher 3.3 RTV, Canada). Larval development stages were identified according to Kendall et al. (1984) and categorized into four periods: (I) yolk-sac larva, from hatching until yolk sac absorption; (II) preflexion larva, from yolk sac absorption until the start of notochord flexion; (III) flexion larva, from the start of notochord flexion until its completion; and (IV) post-flexion larva, from the completion of notochord flexion until the end of larval development and transition to the juvenile stage. Observations of larval development were terminated once the juvenile stage was reached.

2.3. Larval Feeding

Exogenous feeding commenced once the larval mouths opened. Larvae were fed with live *Artemia salina* nauplii (INVE Aquaculture Inc., Dendermonde, Belgium) once daily at a density of 5-10 individuals/mL until they reached the juvenile stage. Artemia cysts were hatched in 1.5 L cylindro-conical plastic containers at a water temperature of 25-26 °C and salinity of 25-30 ppt. Newly hatched Artemia nauplii were used for larval feeding.

3. Results

1 DAH: On the first day after hatching (1 DAH), the penguin tetra larvae have a large yolk sac (Figure 1). The eyes are in the formation stage, but eye development is not complete (Figure 1). The body is in primordial form, without pigmentation, and has a transparent appearance. From the outside, the digestive system appears to be a straight tube. On the first day, the mouth and anus are closed (Figure 1). There are a few small, scattered pigment spots on the head region. However, the head region also has a generally transparent appearance. Due to this transparency, the otolith structures inside the otic capsule appear as two small dots (Figure 1).
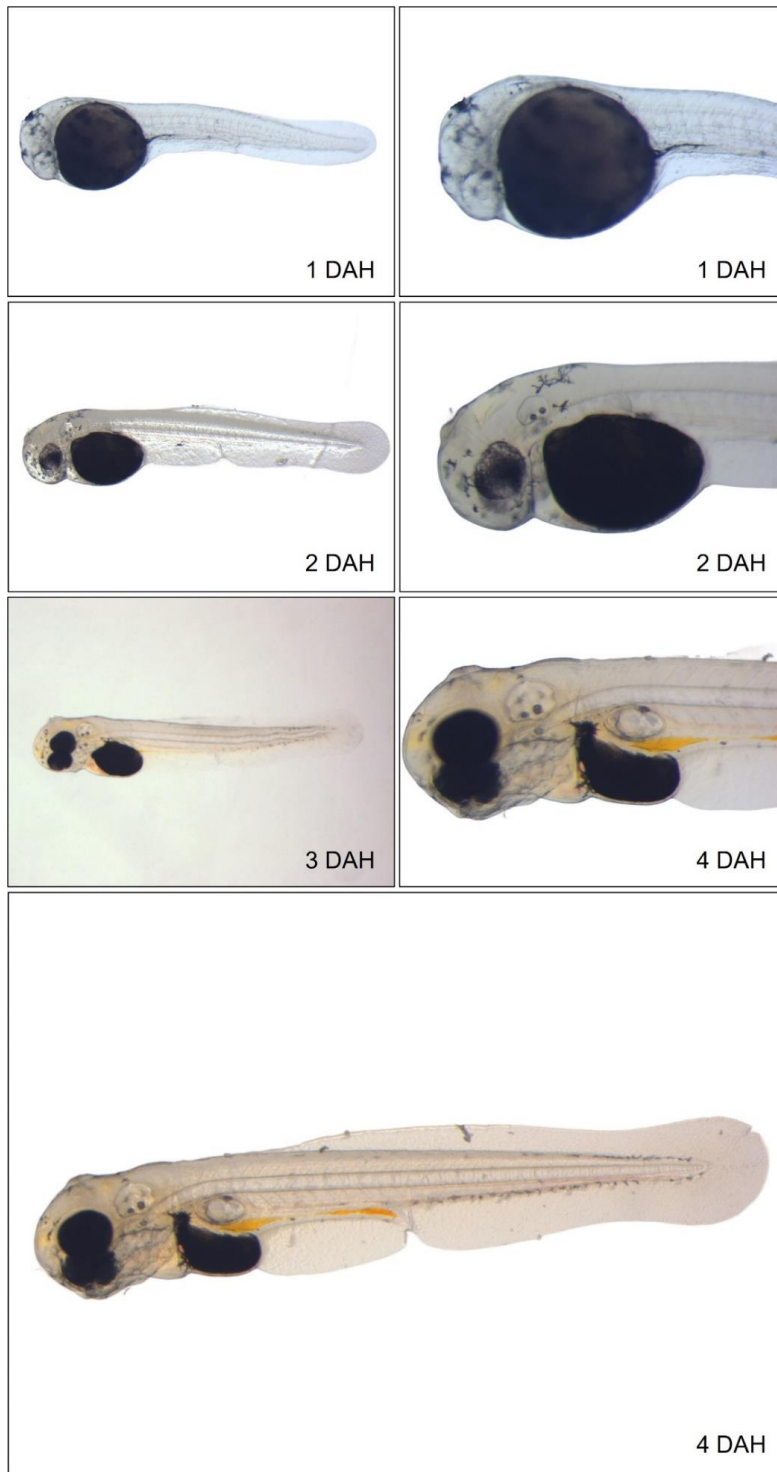
Figure 1. Larval development of penguin tetra (*Thayeria boehlkei*). Post-hatching stage (1 DAH), yolk-sac stage, the gas bladder was formed but not filled (4 DAH).

2 DAH: The proportion of black pigmentation in the eyes has increased (Figure 1). The eyes are slightly more developed compared to the first day. The head region and body are transparent. A pair of adhesive glands are noticeable on the anterior-upper part of the head region. There is a slight increase in pigmentation in the head region compared to the first day. The formation of the pelvic fins is noticeable. The mouth and anus are closed (Figure 1). The larva makes short-term tail movements. It cannot swim freely. The yolk sac has become even smaller compared to the previous day. The vertebral column and primordial fin structures can be more easily distinguished from each other. The primordial fin has widened (Figure 1).

3 DAH: Eye development appears to be complete (Figure 1). The head and body are still transparent. The vertebral column and myomere structures are clearly distinguishable.

The yolk sac has shrunk by approximately 2/3 compared to the first day. The mouth and anus are still closed. The digestive system can be seen as a long, straight tube extending to the anus (Figure 1). It can be noticed that the two small dots inside the otic capsule have grown slightly compared to the first days (Figure 1).

4 DAH: The yolk sac has become very small (Figure 1). The swim bladder has started to form as a small balloon. The adhesive gland on the anterior-upper part of the head is still present (Figure 1). The larva cannot swim freely. However, the duration of short tail movements has increased. The mouth and anus are closed (Figure 1). The digestive system is still in a straight structure (Figure 1). It is yellow in color. The notochord tip maintains its straight form. The body is transparent (Figure 1). Pigmentation continues to increase in the form of small black dots throughout the body.

5 DAH: The mouth has opened (Figure 2). The yolk sac is depleted (Figure 2). The larva has started free swimming movements. It can take external food during these days. The swim bladder has grown slightly. The notochord tip is not curved (Figure 2). The dorsal, anal, and caudal fins have not yet formed.
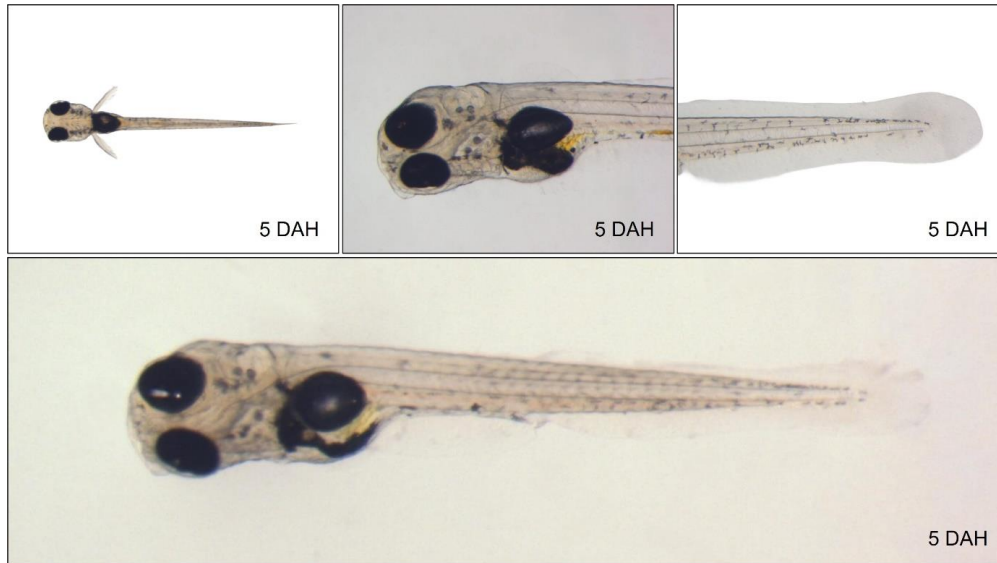
Figure 2. Larval development of penguen tetra (*Thayeria boehlkei*). Consumption of yolk-sac (5 DAH); opened-mouth stage (5 DAH); exogenous feeding stage.

6 DAH: The larva continues to take external food (Figure 3). The body is generally transparent, but the distribution areas of pigmentation continue to expand. Ingested Artemia can be seen in the stomach. Tail formations are not complete (Figure 3). The vertebral column is in a straight form. The notochord is not curved (Figure 3). The swim bladder is slightly elongated oval-shaped towards the back of the body.
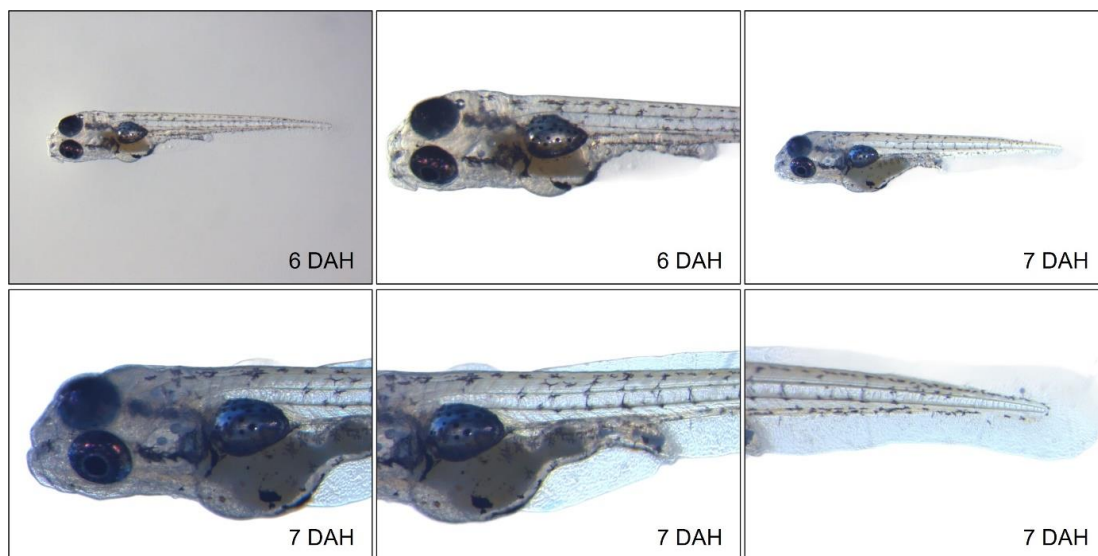
Figure 3. Larval development of penguin tetra (*Thayeria boehlkei*). Preflexion larva stage (6-7 DAH).

7-14 DAH: During these days, the larva can swim freely in series. They feed on live food. The density of pigmentation in their bodies increases slightly every day (Figure 3). The dorsal and anal fins have not yet completed their development (Figures 3 and 4). The caudal fin rays are also in the development stage. The notochord tip is not curved (Figures 4 and 5). Therefore, the preflexion larval period continues.
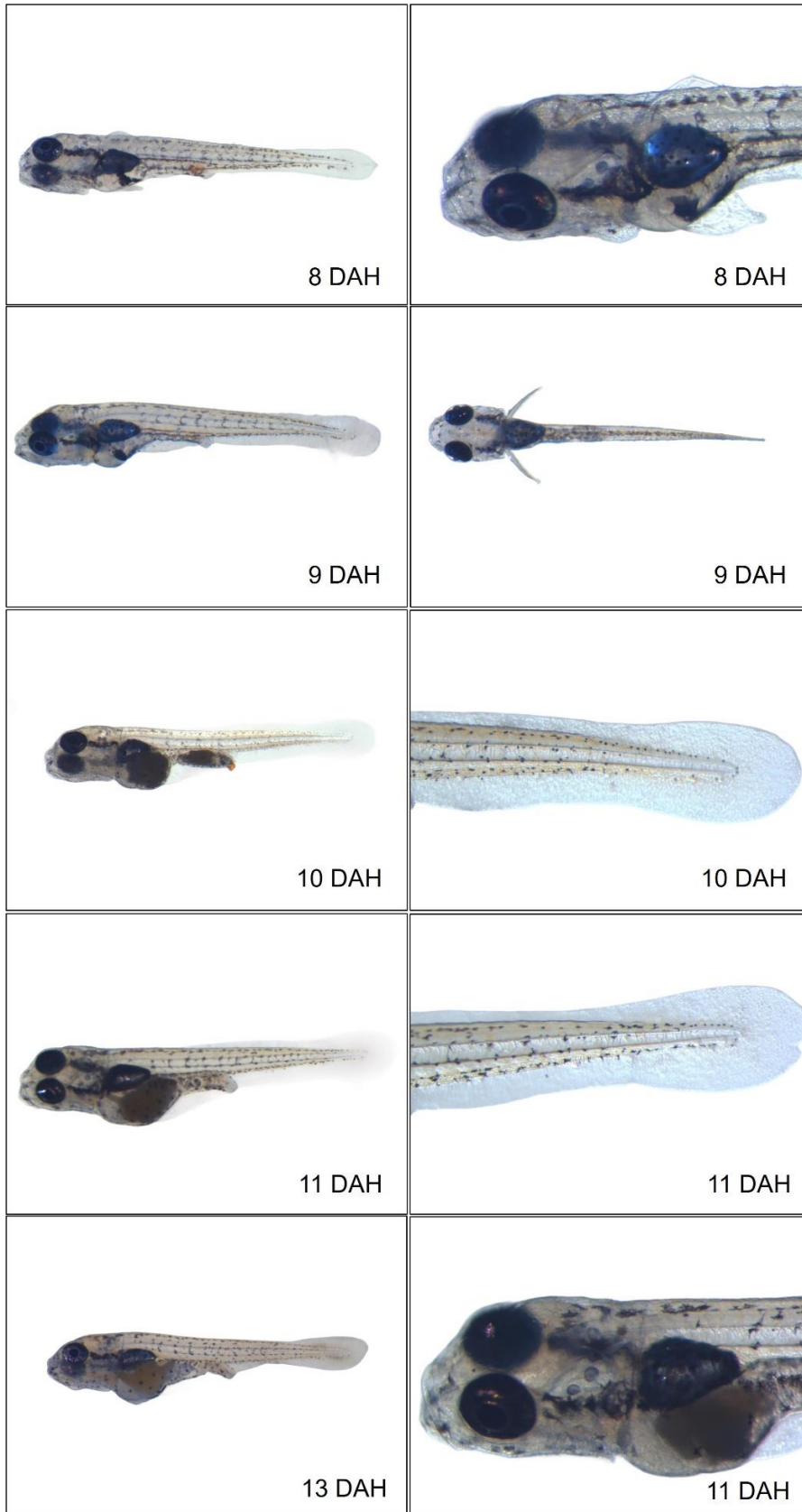
Figure 4. Larval development of penguin tetra (*Thayeria boehlkei*). Preflexion larva (8-11 DAH).

15-16 DAH: The most important morphological event in these days is the curving of the notochord tip (Figure 5). Therefore, these days can be defined as the flexion larval period for penguin tetra larvae. The formations of the anal and dorsal fins have started to become a little clearer (Figure 5). The caudal fin rays have also emerged. Body pigmentation has increased compared to previous days. On the 15th day, the second chamber of the swim bladder has formed. Thus, the swim bladder has become two-chambered (Figure 5).
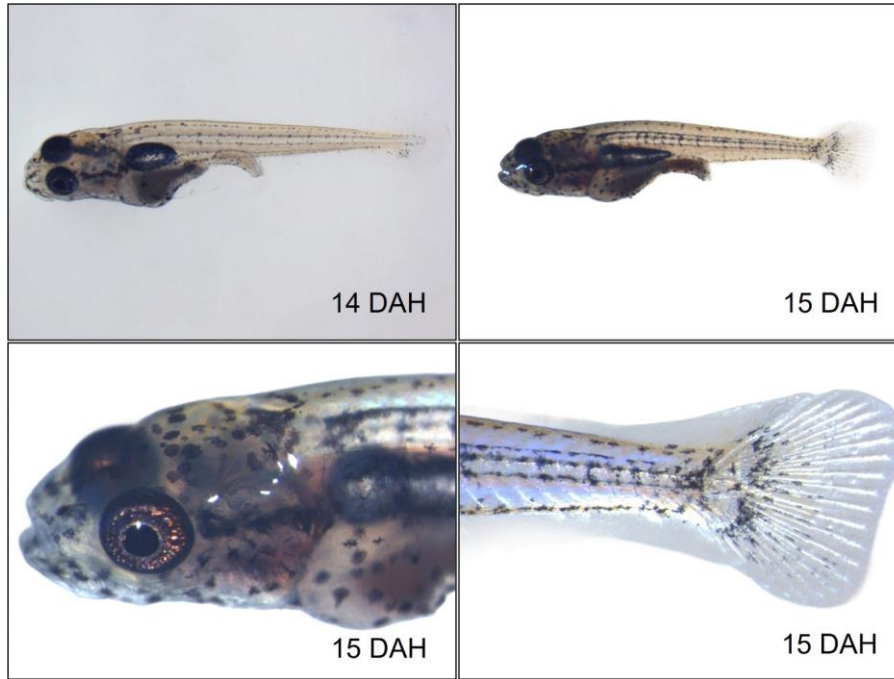
Figure 5. Larval development of penguin tetra (*Thayeria boehlkei*). Flexion stage, notochord flexion started (15 DAH); flexion stage, the notochord was completely flexed (16 DAH), swim bladder with two chambers was visible (15 DAH).

19-21 DAH: The dorsal and anal fins are quite developed, and the caudal fin has taken a forked shape (Figure 6). Body pigmentation has started to enter the color form of the parents (Figure 6). The transparent appearance of the larva is about to disappear.
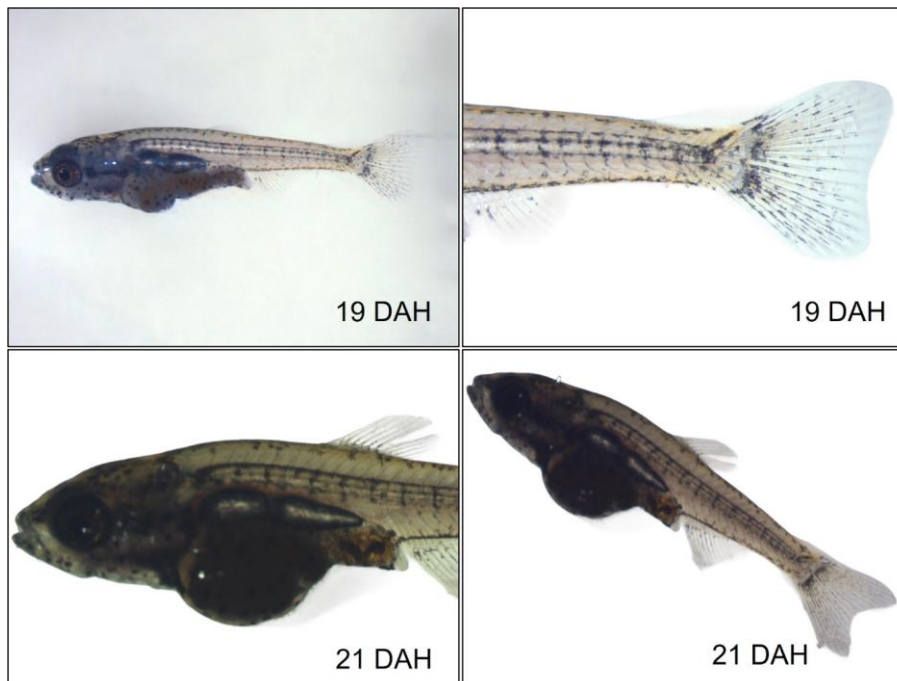
Figure 6. Larval development of penguin tetra (*Thayeria boehlkei*). Postflexion larva.

23 DAH: All fins have become more shaped and have reached the final stage of their development (Figure 7). The transparent body color of the larva has completely disappeared, and their colors resemble the color of adult individuals. During these days, a black line unique to the penguin tetra species has formed on the side of the body, extending from the head region to the lower fork tip of the caudal fin (Figure 7). Larval development is morphologically complete (Figure 7). The larvae have now taken on the color and body form specific to the penguin tetra species (Figure 7). Therefore, during these days, the larval stage is completed, and the juvenile stage has been entered.

Figure 7. Juvenile of penguin tetra (*Thayeria boehlkei*). End of metamorphosis. The body shape of larvae and pigmentation pattern were similar to those of the adult (25 DAH).
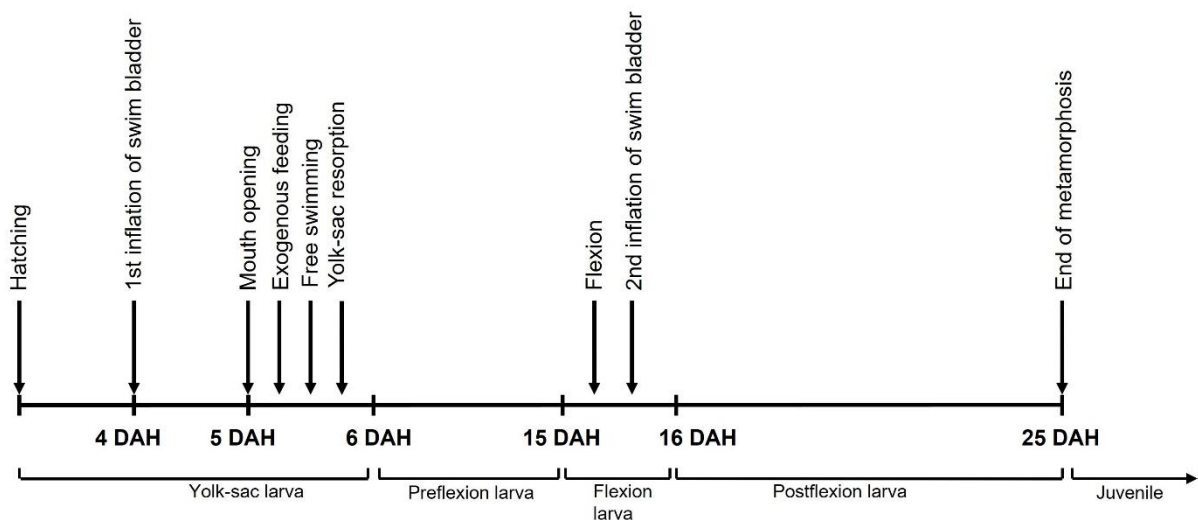
Figure 8. The main events of larval development in penguen tetra (*Thayeria boehlkei*).

4. Discussion

Tetra fish are small, colorful, and popular freshwater aquarium fish belonging to the Characidae family (Helfman et al., 2009). Understanding the larval development process of these fish from hatching to the juvenile stage can help comprehend the reproductive biology, growth potential, and survival rates of that species (Nakatani et al., 2001).

Tetra eggs are usually transparent, round, and small. The larvae that hatch from these eggs are quite small, usually transparent in color, and underdeveloped. Newly hatched larvae are dependent on their yolk sacs for nutrition until they start external feeding. For tetra larvae to develop and survive, it is critical to provide them with appropriate living conditions and nutritious live foods such as rotifers and Artemia nauplii (Çelik & Cirik, 2020; Lipscomb et al., 2020). Therefore, Artemia is preferred as the first feed in tetra larval feeding. In this study, Artemia nauplii were used for the initial feeding of penguin tetra larvae. The aquaculture practices, water quality parameters, and live food feeding protocol used during the larval rearing process directly affect the timing of morphological events in the developing larvae. For example, when comparing the timing of metamorphosis events during the larval development of black tetra and serpae tetra, it is observed that although these two species have similar morphological development findings, their larval growth rates slightly differ from each other. Looking at these two studies, it was reported that black tetra larvae developed slightly faster than serpae tetra larvae (Çelik & Cirik, 2020; Lipscomb et al., 2020). This situation is the same for penguin tetra larvae as it is for all other tetra species. Environmental conditions and feeding protocols are directly related to the development processes and timing of morphological events in larvae. It is known that larvae of the same species have different growth rates under different conditions. Therefore, new studies on the growth and survival of the species are needed to accurately determine the larval development rates of a species.

In this study, it was observed that the body shape, organs, fins, and body color structure of penguin tetra larvae gradually became more distinct within the first month after hatching. Primarily, the development of the head and eye region was observed, followed by the development of the caudal fin, dorsal, ventral, and anal fins. This situation has also occurred in the same way in some other tetra species (Çelik & Cirik, 2020; Lipscomb et al., 2020). Free swimming started after the initial inflation of the swim bladder.

Pigmentation is one of the most important morphological findings of larval development. These findings are even used in species identification of larvae. Therefore, pigmentation formation and intensification are also mentioned when describing larval development. In this study, the distribution of pigments on the body was also mentioned. Fish larvae initially appear transparent. Later, over time, pigment cells form and develop in their bodies, and when they reach the juvenile stage, the offspring acquire the color structure of their parents. Pigmentation colors and patterns also differ in different fish species. The same situation applies to different tetra species (Helfman et al., 2009). Pigmentation occurs in species-specific color patterns (Nakatani et al., 2001). As larvae grow and approach the juvenile stage, they become colored similarly to their parents' colors. Therefore, describing the pigmentation process during the larval development of fish is critically important.

Just as pigmentation varies according to species, the duration of larval development also varies depending on the species and environmental conditions. It is known that the development stages of tetra larvae generally take 3-4 weeks (Çelik & Cirik, 2020; Lipscomb et al., 2020). At the end of this period, the larval stage is completed, and the juvenile stage is reached. In this study, the larval development process of the penguin tetra examined was also completed in 3-4 weeks. These offspring have become morphologically identical to their parents. However, it may take much longer for larvae to reach sexual maturity. This also varies from species to species.

Examining the larval development of a fish species of economic importance and documenting the findings are important for scientific research. On the other hand, it is also important for professionals who culture this species to improve their aquaculture practices. Examining the larval development of tetra species is similarly important for scientific research. In addition, it is also beneficial for amateur and professional producers who are engaged in the aquarium hobby and breed tetras. Because accurately describing the larval development process of a fish species can reveal valuable information about the reproductive biology, growth dynamics, and survival capacity of that species (Nakatani et al., 2001). Furthermore, detecting diseases and abnormalities that may occur during larval rearing can contribute to raising healthy products for the aquarium fish industry.

In conclusion, the larval development of the penguin tetra species is critically important for understanding the life cycle of this species and using this information to raise higher quality fish generations. Describing the life cycle of an economically important fish species widely used in the aquarium industry, such as the penguin tetra, from hatching to the juvenile stage is important for the continuity of that species and the sustainability of the aquarium industry. Understanding the larval development process of this species can contribute to raising healthier and higher quality fish in the aquarium fish industry.

Compliance with Ethical Standards

The study was conducted when ethics committee approval was not required. Both authors possess certificate of handling experimental animals.

Conflict of Interest

The authors declare that they have no conflict of interest.

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PRENSİP

RESEARCH ARTICLE

Researches on Seafood Consumption Behaviors in Kütahya Province

Nurcan Şimşek ID • Gökhan Arslan✉ ID

Atatürk University, Faculty of Fisheries, Erzurum/Türkiye

ABSTRACT

Seafood is among the most important recommended foods due to its rich protein, mineral and fat content. In recent years, food has become an extremely important product in terms of maintenance due to increasing food demand and decreasing contractual production. In the current research, face-to-face questions were examined with 128 people living in Kütahya province and its central districts, and the answers given were evaluated with the Chi-square interval test. 76 women (59.4%) and 82 men (40.6%) that participated in this survey. According to the age analysis, 21.8% are under 21; 9.4% are between 21-30; 21.8% were between 31-40; 21.8% were between 41-50; 6.4% were between 51-60 and 18.8% were between 61-70. It was observed that there was a relationship between increasing income and fish meat consumption, and fish meat was preferred at a higher level of income. In addition, it has been an important data that young people in later periods consume more fish.

1. Introduction

Decreasing food resources and increasing world population are considered to be the source of an important nutritional problem for the future. Experts on the subject emphasize that food production should double in parallel with the increase in world population in the 2050s. The sector that can meet this need apart from agricultural production is aquaculture, considering its current potential (Arslan, 2019).

It has been observed that the sector has been growing faster than other food sectors worldwide in recent years. In this sense, it is predicted that the aquaculture sector will continue to increase its importance in terms of food supply in the near future (Arslan & Oğuzhan Yıldız, 2021).

The development of the aquaculture sector and similarly all

sectors is undoubtedly closely related to the balance of supply and demand. In this sense, fish consumption is extremely important for the development of the sector. Many studies have been conducted at different times and regions on fish consumption habits in Türkiye (Atay et al., 2002; Aydın & Karadurmuş, 2012; Beyazbayrak, 2014; Çiçek et al., 2014; Abdikoğlu et al., 2015; Ercan & Şahin, 2016; Gürel et al., 2017; Şen & Şahin, 2017; Arslan, 2019).

These studies show that fish consumption in our country is below the world average. As of 2019, our annual fish consumption per capita in our country was 6.26 kg (TÜİK, 2019). This situation is also important for the development of the sector. A significant part of the consumption took place in coastal areas. For this reason, studies in provinces that do not have a coastline can be data sources regarding low

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✉ Correspondence
E-mail address: gokhan.arslan@atauni.edu.tr

consumption.

In this study, fish consumption habits of Kütahya province were examined. Data was collected by asking face-to-face questions to 128 participants, taking into account gender, age, education level, income level and occupational characteristics.

2. Materials and Methods

Our study was conducted by asking face-to-face questions to 128 people residing in Kütahya in September and October 2023, and evaluating the data obtained with the Chi-square independence test. Kütahya Province is a province in the Aegean region of Türkiye. Its area is 11,634 km^2 and its population is 580,701 in 2022 (Wikipedia, 2024).

3. Results and Discussion

In this part of the study, descriptive statistics of the individuals participating in the survey and findings and comments obtained as a result of data analysis are included.

3.1. Demographic Characteristics and Frequency Distributions of the Individuals Participating in the Study

The demographic characteristics of the individuals participating in the study and the frequency distributions of their answers to the questions asked about the study are given in the Table 1.

Table 1. Demographic characteristics and frequency distributions of the individuals participating in the study.

Gender	Male	Female			
N	52	76			
%	40.6	59.4			
Age	<21	21-30	31-40	41-50	51-60	61-70
N	28	12	28	28	8	24
%	21.8	9.4	21.8	21.8	6.4	18.8
Educational status	Primary school	High school	University	Postgraduate	
N	40	52	28	8	
%	31.25	40.6	21.8	6.2	
Income level	<4000	4000-6000	6000-8000	8000-10000	10000+
N	24	4	12	16	72
%	18.8	3.2	9.4	12.8	56.2
Occupational	Public	Private sector	Student	Retire	Self-employment	Housewife
N	8	12	32	24	12	40
%	6.4	9.4	25.2	18.8	9.4	31.2

According to the demographic characteristics given in Table 1, 40.6% of the individuals participating in the survey are male and 59.4% are female. In the analysis made according to ages, It was determined 21.8% are under 21; 9.4% are between 21-30; 21.8% were between 31-40; 21.8% were between 41-50; 6.4% were between 51-60 and 18.8% were between 61-70. In the study conducted by Oğuzhan Yıldız and Arslan (2021) in Erzurum, the lowest age group of the participants was determined as the 61-70 age group with 4.92%, while the highest age group was determined as the 21-30 age group with 41.80%.

Considering the answers given by the individuals participating in the survey according to their education level, it was revealed that 31.25% were primary school graduates, 40.6% were secondary school graduates, 21.8% were university graduates and 6.2% were postgraduates. Considering the income levels, 18.8% are below 4000, 3.2% are between 4000-6000, 9.4% are between 6000-8000, 12.8% are between 8000-10000 and 56.2% are It was determined that 2 of them had an income of 10000 or more. When the occupations of the individuals participating in the study are examined, 6.4% work in the public sector, 9.4% work in the private sector, 25.2% are students, 18.8% are retired, 9.4% are self-employed and 31.2% are self-employed. It was determined that 2 of them were housewives. In the study conducted by Karakaya and Kırıcı (2016) in Bingöl, 6.8% of the consumers are tradesmen, 7.3% are retired, 7.8% are self-employed, 8.3% are workers, 18% are 5% were students, 23.4% were housewives and 27.9% were civil servants.

3.2. Analysis of the Answers Given to Questions Related to Demographic Characteristics using the Chi-Square Independence Test

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which type of animal product do you consume more" differs depending on gender (P<0.05). While 68.4% of women preferred chicken meat, this rate was found to

be 30.8% for men. Additionally, while 23.1% of men consume fish meat, 5.3% of women consume fish meat. In the study conducted by Olgunoğlu et al. (2014) in Adıyaman, it was found that the consumption rate of red meat and chicken was higher than the consumption rate of fish.

Table 2. Distribution of the question "Which type of animal product do you consume more?" depending on gender.

		Which type of animal product do you consume more?		
		Chicken	**Fish**	**Meat**
Gender	Male	16	12	24
		30.8%	23.1%	46.2%
	Female	52	4	20
		68.4%	5.3%	26.3%

Pearson Chi-Square: 19.612, P= 0.000.

Table 3. Distribution of the question "What do you pay attention to when buying fish?" depending on gender.

		What do you pay attention to when buying fish?				
		Price	**Taste**	**Freshness**	**Small Fishbone**	**All**
Gender	Male	0	4	24	8	12
		0.0%	8.3%	50.0%	16.7%	25.0%
	Female	4	12	16	0	36
		5.9%	17.6%	23.5%	0.0%	52.9%

Pearson Chi-Square: 26.953, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "What do you pay attention to when buying fish differs depending on gender?" (P<0.05). While 50% of men pay attention to freshness, this rate was found to be 23.5% for women. Additionally, while 5.9% of women pay attention to the price, 0% of men do not pay attention to the price.

Table 4. Distribution of the question "Which type of animal product do you consume more?" depending on age.

		Which type of animal product do you consume more?		
		Chicken	**Fish**	**Meat**
Age	<21	20	0	8
		71.4%	0.0%	28.6%
	21-30	4	0	8
		33.3%	0.0%	66.7%
	31-40	20	4	4
		71.4%	14.3%	14.3%
	41-50	12	0	16
		42.9%	0.0%	57.1%
	51-60	8	0	0
		100.0%	0.0%	0.0%
	61-70	4	12	8
		16.7%	50.0%	33.3%

Pearson Chi-Square: 65.039, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which type of animal product do you consume more" varies depending on age (P<0.05). While 100% of the 51-60 age group consumed chicken meat, this rate was found to be 16.7% in the 61-70 age group. In addition, while 50% of individuals between the ages of 61-70 consumed fish, it was found to be 0% in the age ranges under 21, 21-30, 41-50 and 51-60.

Table 5. Distribution of the question "If your answer is no, what is your reason?" depending on age.

		If your answer is no, what is your reason?		
		Taste	**Price**	**Difficulty purchasing**
Age	21-30	4 100.0%	0 0.0%	0 0.0%
	31-40	0 0.0%	4 100.0%	0 0.0%
	41-50	4 100.0%	0 0.0%	0 0.0%
	61-70	0 0.0%	0 0.0%	4 100.0%

Pearson Chi-Square: 32.000, P= 0.000.

If your answer is no, a significant difference emerged as a result of the Chi-square independence test performed to determine whether the question "What is your reason?" varies depending on age (P<0.05). While 100% of individuals between the ages of 21-30 and 41-50 stated that they do not consume fish due to taste incompatibility, individuals between the ages of 31-40 stated that they do not consume fish due to its price. 100% of individuals between the ages of 61-70 stated that they do not consume fish due to the difficulty of purchasing it.

Table 6. Distribution of the question "If your answer is yes, how often do you consume fish" depending on age.

		If your answer is yes, how often do you consume fish		
		Once a week or more	**Once a month or more**	**Once a year or more**
Age	<21	8 28.6%	4 14.3%	16 57.1%
	21-30	0 0.0%	4 50.0%	4 50.0%
	31-40	4 16.7%	12 50.0%	8 33.3%
	41-50	4 16.7%	12 50.0%	8 33.3%
	51-60	0 0.0%	0 0.0%	8 100.0%
	61-70	12 60.0%	4 20.0%	4 20.0%

Pearson Chi-Square: 38.076, P= 0.000.

If your answer is yes, a significant difference emerged as a result of the Chi-square independence test performed to determine whether the question "How often do you consume fish differs depending on age" (P<0.05). While 60% of individuals aged 61-70 consume fish once a week or more, 20% stated that they consume fish once a year or more. While 100% of individuals aged 51-60 stated that they consumed fish once a year or more, 14.3% of individuals under 21 years of age stated that they consumed fish once a month or more.

Table 7. Distribution of the question "What kind of seafood do you consume most?" depending on age.

		What kind of seafood do you consume most?	
		Marine fish	**Freshwater fish**
Age	<21	24 85.7%	4 14.3%
	21-30	4 50.0%	4 50.0%
	31-40	24 100.0%	0 0.0%
	41-50	20 83.3%	4 16.7%
	51-60	4 50.0%	4 50.0%
	61-70	20 83.3%	4 16.7%

Pearson Chi-Square: 17.216, P= 0.004.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "What kind of seafood do you consume most" varies depending on age (P<0.05). When the individuals participating in the survey were asked about the type of seafood they consume most, 100% of individuals in the 31-40 age group stated that they consumed marine fish, while 50% of individuals in the 21-30 and 51-60 age groups stated that they consumed marine fish. While the rate of participants consuming freshwater fish is 50% between the ages of 21-30 and 51-60, this rate is 14.3% for participants under 21. In the study conducted by Bolat and Telli (2019) in Denizli, 72% of the individuals participating in the survey preferred marine fish, while the rate of those who preferred both marine fish and freshwater fish was 23%. The rate of those who prefer freshwater fish remained at 5%.

Table 8. Distribution of the question "Which of the following fish do you consume more?" depending on age.

		Which of the following fish do you consume more?		
		Trout	**Anchvoy**	**Bonito**
Age	<21	8 28.6%	16 57.1%	4 14.3%
	21-30	4 50.0%	4 50.0%	0 0.0%
	31-40	0 0.0%	24 100.0%	0 0.0%
	41-50	4 16.7%	16 66.7%	4 16.7%
	51-60	4 50.0%	4 50.0%	0 0.0%
	61-70	4 16.7%	20 83.3%	0 0.0%

Pearson Chi-Square: 27.816, P= 0.002.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which of the following fish do you consume more?" varies depending on age (P<0.05). The proportion of individuals participating in the survey who consume trout is 50% in the 21-30 and 51-60 age range, while it is 0% in the 31-40 age range. While 100% of individuals between the ages of 31-40 consumed anchovies, 14.3% of individuals under 21 years old stated that they consumed bonito. Gürel et al. (2017) in the study conducted in the central district of Ağrı province, when the fish species most preferred by consumers were examined, it was found that 60.70% consumed anchovy, 13.55% consumed trout and at least 2.71% consumed fish. It was determined that they consumed horse mackerel.

Table 9. Distribution of the question "What do you pay attention to when buying fish" depending on age.

		What do you pay attention to when buying fish?				
		Price	Taste	Freshness	Small Fishbone	All
Age	<21	0	4	12	0	12
		0.0%	14.3%	42.9%	0.0%	42.9%
	21-30	0	4	0	4	0
		0,0%	50.0%	0.0%	50.0%	0.0%
	31-40	4	4	4	4	8
		16.7%	16.7%	16.7%	16.7%	33.3%
	41-50	0	0	16	0	8
		0.0%	0.0%	66.7%	0.0%	33.3%
	51-60	0	0	4	0	4
		0.0%	0.0%	50.0%	0.0%	50.0%
	61-70	0	4	4	0	16
		0.0%	16.7%	16.7%	0.0%	66.7%

Pearson Chi-Square: 81.752, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was performed to determine whether the question "What do you pay attention to when buying fish varies depending on age" (P<0.05). 66.7% of individuals between the ages of 31-40 stated that they pay attention to freshness when buying fish. 14.3% of the participants under 21 stated that they pay attention to the taste.

Table 10. Distribution of the question "What is the most important reason for fish consumption?" depending on age.

		What is the most important reason for fish consumption?		
		Delicious	Healthy	Both of them
Age	<21	8	0	20
		28.6%	0.0%	71.4%
	21-30	8	0	0
		100.0%	0.0%	0.0%
	31-40	4	12	8
		16.7%	50.0%	33.3%
	41-50	0	8	16
		0.0%	33.3%	66.7%
	51-60	0	4	4
		0.0%	50.0%	50.0%
	61-70	0	4	20
		0.0%	16.7%	83.3%

Pearson Chi-Square: 73.434, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was performed to determine whether the question "What is the most important reason for consuming fish?" varies depending on age (P<0.05). While 100% of the surveyed individuals between the ages of 21-30 stated that they consume fish because it is delicious, 16.7% of individuals between the ages of 61-70 stated that they consume fish because it is healthy. Yüksel et al. (2011) in their study in Tunceli province, when asked about their reasons for choosing fish, 31% stated that they consumed fish only for a healthy and balanced diet, 7% stated that they consumed fish only for taste, and 62% stated that they consumed fish for both reasons.

Table 11. Distribution of the question "Which type of animal product do you consume more?" depending on education level.

		Which type of animal product do you consume more?		
		Chicken	Fish	Meat
Educational status	Primary school	32 80.0%	4 10.0%	4 10.0%
	High school	32 61.5%	8 15.4%	12 23.1%
	University	4 14.3%	4 14.3%	20 71.4%
	Postgraduate	0 0.0%	0 0.0%	8 100.0%

Pearson Chi-Square: 50.000, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which type of animal product do you consume more" differs depending on education level (P<0.05). While 80% of primary school graduates consume more chicken meat, 14.3% of university graduates stated that they consume chicken meat. While 100% of postgraduate graduates consumed more red meat, 10% of primary school graduates stated that they consumed fish.

Table 12. Distribution of the question "Do you consume fish" depending on education level.

		Do you consume fish?	
		Yes	No
Educational status	Primary school	32 80.0%	8 20.0%
	High school	52 100.0%	0 0.0%
	University	20 71.4%	8 28.6%
	Postgraduate	8 100.0%	0 0.0%

Pearson Chi-Square: 17.241, P= 0.001.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Do you consume fish" differs depending on education level (P<0.05). While 100% of the individuals who participated in the survey with secondary and postgraduate education stated that they consumed fish, 20% of primary school graduates and 28.6% of university graduates stated that they did not consume fish.

Table 13. Distribution of the question "If your answer is no, what is your reason?" depending on education level.

		If your answer is no, what is your reason?		
		Taste incompatibility	Price	Difficulty purchasing
Educational status	Primary school	0 0.0%	4 50.0%	4 50.0%
	University	8 100.0%	0 0.0%	0 0.0%

Pearson Chi-Square: 16.000, P= 0.000.

If your answer is no, a significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "What is your reason?" differs depending on education level (P<0.05). Although 100% of the participants who were university graduates stated that they did not consume fish due to incompatibility of taste, 50% of the primary school graduates stated that they did not consume fish due to the difficult price

and 50% due to the difficulty of purchasing.

Table 14. Distribution of the question "If your answer is yes, how often do you consume fish" depending on education level.

		If your answer is yes, how often do you consume fish?		
		Once a week or more	Once a month or more	Once a year or more
Educational status	Primary school	8 25.0%	4 12.5%	20 62.5%
	High school	8 15.4%	24 46.2%	20 38.5%
	University	12 60.0%	4 20.0%	4 20.0%
	Postgraduate	0 0.0%	4 50.0%	4 50.0%

Pearson Chi-Square: 28.100, P= 0.000.

If your answer is yes, a significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "How often do you consume fish differs depending on education level?" (P<0.05). While 60% of university graduates stated that they consumed fish once a week or more, 12.5% of primary school graduates stated that they consumed fish once a month or more. 62.5% of primary school graduates and 20% of university graduates stated that they consume fish once or more a year.

Table 15. Distribution of the question "Which of the following fish do you consume more?" depending on education level.

		Trout	Anchovy	Bonito
Educational status	Primary school	12 33.3%	24 66.7%	0 0.0%
	High school	8 15.4%	40 76.9%	4 7.7%
	University	4 20.0%	12 60.0%	4 20.0%
	Postgraduate	0 0.0%	8 100.0%	0 0.0%

Pearson Chi-Square: 14.787, P = 0.022.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which of the following fish do you consume more?" varies depending on educational level (P<0.05). While 33.3% of primary school graduates stated that they consumed more trout, 100% of postgraduate graduates stated that they consumed more anchovies. In addition, 20% of university graduates stated that they consumed bonito more, while 7.7% of secondary school graduates stated that they consumed bonito more.

Table 16. Distribution of the question "What do you pay attention to when buying fish, depending on education level?"

		What do you pay attention to when buying fish, depending on education level?				
		Price	Taste	Freshness	Small Fishbone	All
Educational status	Primary school	4 11.1%	4 11.1%	12 33.3%	0 0.0%	16 44.4%
	High school	0 0.0%	8 15.4%	16 30.8%	8 15.4%	20 38.5%
	University	0 0.0%	4 20.0%	4 20.0%	0 0.0%	12 60.0%
	Postgraduate	0 0.0%	0 0.0%	8 100.0%	0 0.0%	0 0.0%

Pearson Chi-Square: 37.252, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "What do you pay attention to when buying fish varies depending on education level?" (P<0.05). While 11.1% of primary school graduates stated that they pay attention to the price when buying fish, 100% of postgraduate graduates pay attention to its freshness; While 20% of university graduates stated that they pay attention to whether it is delicious, 15.4% of secondary school graduates stated that they pay attention to whether it has fewer strings.

Table 17. Distribution of the question "What is the most important reason for consuming fish?" depending on education level.

		What is the most important reason for consuming fish?		
		Delicious	**Healthy**	**Both of them**
Educational status	Primary school	0	8	28
		0.0%	22.2%	77.8%
	High school	16	16	20
		30.8%	30.8%	38.5%
	University	4	4	12
		20.0%	20,0%	60,0%
	Postgraduate	0	0	8
		0.0%	0.0%	100.0%

Pearson Chi-Square: 24.471, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was performed to determine whether the question "What is the most important reason for consuming fish?" varies depending on education level (P<0.05). While 30.8% of secondary school graduates stated that they consume fish because it is delicious, 30.8% stated that they consume fish because it is healthy. 100% of postgraduate graduates stated that they consume fish for both reasons.

Table 18. Distribution of the question "Which type of animal product do you consume more?" depending on income level.

		Which type of animal product do you consume more?		
		Chicken	**Fish**	**Meat**
Income level	<4000	24	0	0
		100.0%	0.0%	0.0%
	4000-6000	0	0	4
		0.0%	0.0%	100.0%
	6000-8000	0	12	0
		0.0%	100.0%	0.0%
	8000-10000	12	0	4
		75.0%	0.0%	25.0%
	10000>	32	4	36
		44.4%	5.6%	50.0%

Pearson Chi-Square: 125.576, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which type of animal product do you consume more" varies depending on income level (P<0.05). Among the respondents, the rate of those who consume chicken meat more is 100% among those with an income of less than 4,000, while it is 44.4% among those with an income of over 10,000. While 100% of the participants between 6000-8000 stated that they consume fish meat more, the rate of those consuming fish meat more is 5.6 among participants over 10000.

Table 19. If your answer is yes, distribution of the question "How often do you consume fish" depending on income level.

		If your answer is yes, how often do you consume fish?		
		Once a week or more	Once a month or more	Once a year or more
Income level	<4000	0 0.0%	4 16.7%	20 83.3%
	4000-6000	0 0.0%	4 100.0%	0 0.0%
	6000-8000	4 33.3%	8 66.7%	0 0.0%
	8000-10000	4 33.3%	4 33.3%	4 33.3%
	10000>	20 33.3%	16 26.7%	24 40.0%

Pearson Chi-Square: 38.267, P= 0.000.

If your answer is yes, a significant difference emerged as a result of the Chi-square independence test performed to determine whether the question "How often do you consume fish differs depending on income level?" (P<0.05). Of the individuals who said they consumed fish, 83.3% of those with an income below 4000 stated that they consumed fish once a year or more, while 33.3% of individuals between 8000-10000 stated that they consumed fish once a month or more. Those who consume fish once a month or more include 100% of the individuals in the range of 4000-6000. Karakaya et al. (2020) in the study conducted in Erzincan province, when asked about the frequency of fish consumption, the rate of people consuming fish once every five days was found to be 50%, the rate of people consuming fish once a month was 40%, and the rate of people consuming fish once a week was 10%.

Table 20. Distribution of the question "What kind of seafood do you consume most?" depending on income level.

		What kind of seafood do you consume most?	
		Marine fish	Freshwater fish
Income level	<4000	20 83.3%	4 16.7%
	4000-6000	4 100.0%	0 0.0%
	6000-8000	12 100.0%	0 0.0%
	8000-10000	8 50.0%	8 50.0%
	10000>	52 86.7%	8 13.3%

Pearson Chi-Square: 16.014, P = 0.003.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "What kind of seafood do you consume most" varies depending on income level (P<0.05). When the participants were asked about the marine fish they consume most, 100% of individuals with incomes between 4000-6000 and 6000-8000 stated that they consumed marine fish, while 13.3% of individuals with income over 10000 and 16.7% of individuals with incomes between 4000-6000 stated that they consumed freshwater fish. They stated that they consumed more freshwater fish.

Table 21. Distribution of the question "Which of the following fish do you consume more?" depending on income level.

		Which of the following fish do you consume more?		
		Trout	Anchovy	Bonito
Income level	<4000	4 16.7%	16 66.7%	4 16.7%
	4000-6000	0 0.0%	4 100.0%	0 0.0%
	6000-8000	0 0.0%	12 100.0%	0 0.0%
	8000-10000	8 50.0%	8 50.0%	0 0.0%
	10000>	12 20.0%	44 73.3%	4 6.7%

Pearson Chi-Square: 18.597, P = 0.017.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which of the following fish do you consume more?" varies depending on income level (P<0.05). While 50% of individuals with income between 8000-100000 stated that they consumed more trout, 50% stated that they consumed more anchovy. While 6.7% of individuals with income over 10000 stated that they consumed bonito more, 100% of individuals with income between 4000-6000 and 6000-8000 stated that they consumed anchovy more.

Table 22. Distribution of the question "What do you pay attention to when buying fish?" depending on income level.

		What do you pay attention to when buying fish?				
		Price	Taste	Freshness	Small Fishbone	All
Income level	<4000	0 0.0%	8 33.3%	12 50.0%	0 0.0%	4 16.7%
	4000-6000	0 0.0%	0 0.0%	0 0.0%	4 100.0%	0 0.0%
	6000-8000	4 33.3%	4 33.3%	0 0.0%	0 0.0%	4 33.3%
	8000-10000	0 0.0%	4 25.0%	0 0.0%	4 25.0%	8 50.0%
	10000>	0 0.0%	0 0.0%	28 46.7%	0 0.0%	32 53.3%

Pearson Chi-Square: 142.454, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "What do you pay attention to when buying fish varies depending on income level" (P<0.05). When the participants were asked about the features they pay attention to when buying fish, 33.3% of individuals with an income between 6000-8000 said that they pay attention to the price, while 100% of individuals with an income between 4000-6000 said that they pay attention to whether it has small fishbones. While 50% of individuals with an income below 4000 pay attention to its freshness, 25% of individuals with an income between 8000-10000 stated that they pay attention to its deliciousness. 53.3% of individuals with income over 10,000 stated that they pay attention to all of these features.

Table 23. Distribution of the question "What is the most important reason for consuming fish?" depending on income level.

		What is the most important reason for consuming fish?		
		Delicious	Healthy	Both of them
Income level	<4000	12 50.0%	4 16.7%	8 33.3%
	4000-6000	4 100.0%	0 0.0%	0 0.0%
	6000-8000	0 0.0%	4 33.3%	8 66.7%
	8000-10000	4 25.0%	4 25.0%	8 50.0%
	10000>	0 0.0%	16 26.7%	44 73.3%

Pearson Chi-Square: 53.418, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "What is the most important reason for consuming fish?" varies depending on income level (P<0.05). While 25% of individuals with income between 8000-10000 stated that the most important reason for consuming fish is its taste, 100% of individuals with income between 4000-6000 stated that they consume fish for its taste. While 16.7% of individuals with incomes between 4000-6000 before while 16.7% of the individuals with an income over 10000 people stated that they consume fish because it is healthy. 73.3% of individuals with income over 10,000 stated that they consume fish for both reasons.

Table 24. Distribution of the question "Which type of animal product do you consume more?" depending on the profession.

		Which type of animal product do you consume more?		
		Chicken	Fish	Meat
Occupational	Public	0 0.0%	0 0.0%	8 100.0%
	Private sector	4 33.3%	0 0.0%	8 66.7%
	Student	24 75.0%	0 0.0%	8 25.0%
	Retire	4 16.7%	12 50.0%	8 33.3%
	Self-employment	8 66.7%	0 0.0%	4 33.3%
	Housewife	28 70.0%	4 10.0%	8 20.0%

Pearson Chi-Square: 68.677, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which type of animal product do you consume more" differs depending on the profession (P<0.05). Among the respondents, 75% of students who were students stated that they consumed chicken meat more, while 10% of housewives stated that they consumed fish meat more. While 16.7% of retirees stated that they consumed more chicken meat, 50% stated that they consumed more fish meat.

Table 25. Distribution of the question "Do you consume fish" depending on profession.

		Do you consume fish?	
		Yes	No
Occupational	Public	8 100.0%	0 0.0%
	Private sector	8 66.7%	4 33.3%
	Student	32 100.0%	0 0.0%
	Retire	24 100.0%	0 0.0%
	Self-employment	12 100.0%	0 0.0%
	Housewife	28 70.0%	12 30.0%

Pearson Chi-Square: 26.819, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was performed to determine whether the question "Do you consume fish" differs depending on the profession (P<0.05). While 100% of public employees, students, retirees and self-employed people state that they consume fish, this rate is 66.7% for private sector employees. 33.3% of private sector employees and 30% of housewives stated that they do not consume fish.

Table 26. If your answer is yes, distribution of the question "How often do you consume fish" depending on your profession.

		How often do you consume fish?		
		Once a week or more	Once a month or more	Once a year or more
Occupational	Public	0 0.0%	8 100.0%	0 0.0%
	Private sector	0 0.0%	4 50.0%	4 50.0%
	Student	8 25.0%	4 12.5%	20 62.5%
	Retire	12 50.0%	4 16.7%	8 33.3%
	Self-employment	4 33.3%	4 33.3%	4 33.3%
	Housewife	4 14.3%	12 42.9%	12 42.9%

Pearson Chi-Square: 37.675, P= 0.000.

If your answer is yes, a significant difference emerged as a result of the Chi-square independence test performed to determine whether the question "How often do you consume fish differs depending on the profession" (P<0.05). While 100% of the participants working in the public sector stated that they consumed fish once a month or more, 62.5% of the students stated that they consumed fish once a year or more. While 50% of retirees stated that they consumed fish once a week or more, 16.7% stated that they consumed fish once a month or more.

Table 27. Distribution of the question "What kind of seafood do you consume most?" depending on profession.

		What kind of seafood do you consume most?	
		Marine fish	**Freshwater fish**
Occupational	Public	8 100.0%	0 0.0%
	Private sector	8 100.0%	0 0.0%
	Student	24 75.0%	8 25.0%
	Retire	20 83.3%	4 16.7%
	Self-employment	8 66.7%	4 33.3%
	Housewife	28 87.5%	4 12.5%

Pearson Chi-Square: 7.371, P = 0.0194.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "What kind of seafood do you consume most" differs depending on the profession (P<0.05). Among the participants, 100% of those working in the public and private sectors stated that they consumed marine fish the most, while 75% of the students stated that they consumed marine fish the most. While 33.3% of self-employed workers stated that they consumed more freshwater fish, 12.5% of housewives stated that they consumed freshwater fish.

Table 28. Distribution of the question "Which of the following fish do you consume more?" depending on profession.

		Which of the following fish do you consume more?		
		Trout	**Anchovy**	**Bonito**
Occupational	Public	0 0.0%	4 50.0%	4 50.0%
	Private sector	0 0.0%	8 100.0%	0 0.0%
	Student	12 37.5%	16 50.0%	4 12.5%
	Retire	4 16.7%	20 83.3%	0 0.0%
	Self-employment	4 33.3%	8 66.7%	0 0.0%
	Housewife	4 12.5%	28 87.5%	0 0.0%

Pearson Chi-Square: 43.155, P= 0.000.

A significant difference emerged as a result of the Chi-square independence test, which was conducted to determine whether the question "Which of the following fish do you consume most" differs depending on the profession (P<0.05). 100% of private sector employees stated that they consume more anchovy. While 12.5% of the students stated that they consumed more bonito, 12.5% of the housewives stated that they consumed more trout. While 87.5% of housewives report that they consume more anchovies, this rate is 50% for public employees.

4. Conclusion and Recommendations

According to the results of our study, 76 women (59.4%) and 82 men (40.6%) that participated in this survey of the individuals who participated in our survey were women and 52 were men. When asked which type of animal product they prefer, 68.4% of women answered chicken, while this rate was 30.8% for men. Therefore, it is seen that a significant portion of women living in the region choose chicken meat in their animal product choices. The fact that 50% of men responded to

the question of what do you pay attention to when buying fish, while this rate was 23.5% for women, also supports the previous consumer behavior. In age-related consumption preferences, the increase in fish meat preference as age increases indicates that fish is important for health. When asked which type of fish they consume, a significant portion of the participants answered sea fish, which can be attributed to the region's proximity to the sea. Regarding the relationship between education levels and fish consumption, it has been observed that fish consumption increases as the education level increases. In this sense, it is understood that the health benefits of fish meat are more clearly understood with the education level and thus it is preferred. Another conclusion drawn from the study that income level also affects preferences significantly is that there is a direct relationship between income level and fish consumption preference. In this case, it should be evaluated especially by sector officials. Parameters such as taste, health, and price change fish consumption preferences.

This study we have conducted is about how individuals shape their fish meat preferences; price, freshness and health significantly affect their preferences. The fact that participants with higher education levels know that fish is an important food item for health when it is associated with the level of education has led us to the fact that the health benefits of fish meat should be explained in this region and similar regions. These and similar studies are extremely valuable data sources for the future of our country, which has fish consumption well below the world average.

Acknowledgment

Conflict of Interest

The authors declare that they have no conflict of interest.

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RESEARCH ARTICLE

Investigation of Quality Parameters of Trout Fishballs Cooked with Sous-Vide Method

Nesim Maskan [ID] • Pınar Oğuzhan Yıldız[✉] [ID]

Atatürk University, Faculty of Fisheries, Erzurum/Türkiye

ABSTRACT

This study was conducted to determine the changes in the quality of rainbow trout (*Oncorhynchus mykiss*) meatballs cooked with the sous-vide technique. Vacuum-packaged fishball were cooked using the sous-vide method under two different temperature (60 °C and 80ºC) and two different times (15 and 20 minute) and stored at 4±1ºC for 25 days. The results showed that sous vide cooking technique at 80 °C for 20 min. effectively limited bacterial growth. It was determined that sous vide cooking technology significantly reduced the levels of total volatile base nitrogen (TVB-N) and thiobarbituric acid reactive substances (TBARS) at the end of storage. As the cooking temperature increased, the pH values of the meatball samples also increased. According to the sensory evaluation results, the meatballs cooked with the sous vide technique received higher scores than the control group, and the most preferred cooking temperature and time was determined to be 60ºC for 20 minutes.

1. Introduction

Nowadays, the increase in the number of working women and the population and the difficult working conditions have increased the demand for nutritious and delicious foods that can be prepared in a short time. In this context, the ready-made food sector has gained importance (Oğuzhan & Yangılar, 2014).

Sous vide (vacuum cooking) is a cooking method that finds application in homes and in the ready-made food industry. Cooking methods are important in terms of both nutrition and health. Sous vide has recently become widely used as a popular cooking method around the world. Sous vide cooking is a healthier technique because less nutritional value of meat is lost compared to traditional cooking (Kaya, 2021).

It is also a method that provides quality food for the ready-made food industry. It is also advantageous both in terms of sensory aspects and in terms of significantly inhibiting microorganism activities (Creed & Reeve, 1998; Seyyar, 2015).

Sous vide is the process of cooking food at controlled temperatures and in vacuum packages. With this technique, foods are cooked alone or together with products such as sauce and spices in vacuum packaging (Haskaraca & Kolsarıcı, 2013).

Sous vide cooking is the process of cooking vacuum-packaged foods by immersing them in a temperature-controlled

water bath for the desired time. In French, sous vide means "under vacuum". The cooking process is generally carried out at low temperatures below 100°C and requires longer cooking time than traditional cooking methods (Bozova, 2020; Yıldız & Yılmaz, 2020; Ceylan, 2021; Öztürk et al., 2021).

Seafood is very important in human nutrition due to its unique composition. It is considered a quality food for human consumption due to its high protein content, essential amino acid profile and other nutritional benefits such as omega-3/omega-6 fatty acids. It is also known to be rich in unsaturated fatty acids, minerals and vitamins (A, B1, B2, D and K) (Erol, 2013; Abdel-Wahab et al., 2020). It contains many functional properties such as high quality and easily digestible proteins, polyunsaturated fatty acids beneficial to health, and vitamins and minerals necessary for human nutrition (Jayasinghe et al., 2013).

Seafood is a food with extremely high nutritional value and beneficial for health and has an important place in consumption. Therefore, products prepared with seafood are of great importance in ready meal consumption. In this context, fish meatball cooked with the sous vide technique are a delicious and nutritious alternative product for the ready-made food industry (Bilgin & Metin, 2022). The sous-vide technique improves the sensory properties of foods and preserves their nutritional values. In addition, this method preserves various vitamins and fatty acids found in foods (Yıldız & Yılmaz, 2020).

Especially for young people and school-age children, products prepared with fish (meatball, croquette, nugget, finger, sausage, cake) are healthy and delicious alternative products for children with fast food habits. Today, in addition to traditional cooking methods, there is a need to apply innovative techniques that have many advantages (fast and safe, longer shelf life, preserves nutritional value, and is heated for consumption). In the literature review, no study was found on cooking rainbow trout meatballs with the sou vide technique. With this study, the possibilities of cooking fish meatball using the sous vide technique at appropriate temperatures and times was be investigated and the quality parameters of rainbow trout balls cooked with the sous vide technique at different temperatures and times was be examined.

2. Materials and Methods

2.1. Material

Rainbow trout fillets (approximately 10 kg) an average of 250±25 g weight and 28±0.8 cm lengths were supplied from Atatürk University Faculty of Fisheries, and meatball ingredients were be supplied from a local market in Erzurum.

2.2. Method

The filleted fish meat was passed through a meat grinder

(Empero EM.08). The formulation of meatball dough is shown in Table 1.

Table 1. Meatball formulation.

Meatball composition	Amount (%)
Minced fish	75
Breadcrumbs	7.5
Salt	1.5
Ground red pepper	0.25
Ground black pepper	0.25
Cumin	0.25
Allspice	0.25
Garlic powder	15

This mixture was kneaded well to obtain meatball dough and the meatballs were shaped into 20 grams. Then the trout meatballs were packaged using a vacuum packaging machine (Multivac A 300/16 Sepp Haggenmuller D 87787 Wolfertschwenden, Germany), the packaged samples were cooked using sous vide cooking equipment (Lacia SVC107). They were divided into 5 groups: K: control, A: 60ºC for 15 min, B: 60ºC for 20 min, C: 80ºC for 15 min, D: 80ºC for 20 min. Fish meatball in the control group were grilled at 175°C for 3 minutes on each side. The cooked meatballs were cooled quickly (30 minutes) in ice water. All meatball samples were stored in the refrigerator at 4±1 °C for 25 days and sensory, microbiological and chemical sensory analyses of the samples were performed on days 0, 5, 10, 15, 20 and 25 of storage. The study was carried out in three replications and two parallels, and the results are given as average.

2.3. pH, TBARS and TVB-N

A 10 g sample was taken from the meatball sample, 100 ml of pure water was added, homogenized and pH value measured (Gökalp et al., 2001).

For TBARS analysis, 2 g of each meatball sample was taken and 12 ml of Trichloroacetic acid solution was added to it. It was homogenized in ultra-turrax and filtered through filter paper. 3 ml of the filtrate was taken and 3 ml of Thiobarbituric acid (0.02 M) was added to it, the mixed and cooled test tubes were centrifuged (2000 g). Readings were made at the absorbance value in the spectrophotometer (530 nm) (Lemon, 1975).

TBARS (μmol malonaldehyde/g): [(absorbance/ k(0.06) x 2/1000) x 6.8] x 1000/ sample weight

TVB-N value was determined as reported by Kızılkaya (2020). An amount of 10 g of the meatball sample was taken, 90 ml of 0.6 N perchloric acid solution was added to it, homogenized with ultra-turrax, filtered, 50 ml of the mixture was taken and phenol phthalein was dropped on it. Then, 6.5 ml of 20% NaOH solution was added, 100 ml of boric acid solution and 3-5 drops of methyl red + methylene blue indicator were

added to the distillate flask and the process was continued until 100 ml of distillate was collected. The resulting distillate was titrated with 0.1 N HCl solution until it turned gray, and the total amount of volatile base-nitrogen was calculated by taking into account the HCl solution spent in the titration.

TVB-N (mg/100g) = n x 16.8 mg nitrogen

2.4. Microbiological Analysis

For microbiological analyses, 10 grams of the meatball sample was taken and placed in a stomacher bag under aseptic conditions, and 90 ml of sterile saline (0.85% NaCl) was added to it and homogenized in the stomacher device. Then, appropriate dilutions were prepared from the homogenate and inoculated into the medium using the surface spreading method. The results are given as log cfu/g. Plate Count Agar was used for total aerobic mesophilic and psychrotrophic bacteria analyses, and the media were stored at 30 °C for 2 days and 10 °C for 7 days, respectively (Baumgart et al., 1986; Vanderzant & Splittstoesser, 1992). De Man Rogosa Sharpe Agar was used for lactic acid bacteria count, and Violet Red Glucose Agar was used for Enterobacteriaceae count, and the media were incubated for 2 days at 30 °C under anaerobic conditions (Baumgart et al., 1986; Gökalp et al., 2001). For yeast-mold counting, Rose Bengal Chloramphenicol medium was used and incubated at 25ºC for 5 days (Halkman, 2005). For Pseudomonas counting, Pseudomonas Agar Base+CFC Selective Agar+Supplement Agar was used and the media were incubated under aerobic conditions at 25ºC for 2 days (Gökalp et al., 2001).

2.5. Sensory Evaluation

The meatballs were subjected to sensory analysis by a group of 10 panelists in terms of appearance, texture, odor, taste, color and general acceptability. Meatball samples were given scores between 1-9 (1: unconsumable; 2: very bad; 3: bad; 4: somewhat bad; 5: neither good nor bad; 6: somewhat good, 7: good, 8: very good, 9: quite good) (Choi et al., 2014). The samples were presented to the panelists after being removed from the vacuum packages without any further processing at room temperature.

2.6. Statistical Analysis

The data obtained from the research were evaluated by applying the analysis of variance technique according to the random trial plan with the SPSS program. All measurements were performed twice. Duncan's multiple comparison tests (p<0.05) was used to compare the means of significant sources of variation.

3. Results and Discussion

3.1. Microbiological Analysis

The microbiological analysis results of samples are given in Table 2. According to the results of analysis of variance, the difference between the groups was found to be statistically significant (p<0.05)

Although the limit value for the total number of bacteria in order to see spoilage in fish is 7-8 log CFU/g, most regulations accept 6-7 log CFU/g as the limit to ensure more reliability for human health (Mol Tokay, 2009). The initial total mesophilic aerobic number of fresh rainbow trout was determined as 3.32 log CFU/g, and a significant decrease in bacterial load was detected after the sous vide process. In our study, the number of mesophilic bacteria in the groups that underwent sous vide treatment varied between 2.00 and 5.95 log CFU/g. While the number of bacteria reached the upper limit value in the control group on the 15th day of storage, it was determined that this value was not reached in the sous vide treated groups. As the cooking process and time increased, bacterial growth slowed down due to exposure to heat treatment. It was observed that the applied cooking treatment had a positive effect on slowing down the growth of total mesophilic aerobic bacteria. After sous vide cooking, significant decreases were determined in the TMAB counts of all samples compared to the control group. Erdem et al. (2022) observed that the decrease in bacterial count may be due to the breakdown of the, denaturation of proteins, plasma membrane and change in the permeability of the cell wall of microorganisms. Additionally, the advantage of the sous vide cooking technique is to prevent aerobic bacterial growth. (Haskaraca & Kolsarıcı, 2013; Erdem & Gökmen, 2022). Gürel İnanlı and Yaz (2020) reported that the numbers of aerobic mesophilic bacteria were higher in the control group (raw pike fish) compared to the sous vide applied groups, which is in agreement with our study. Pongestkul and Benjakul (2022) noted that the total bacterial counts of dried sour-salted fish treated with the sous vide technique at different levels increased in parallel with storage.

Psychrotrophic bacterial (TPAB) growth was slower in the sous vide technique applied groups compared to the control group. The lowest number of bacteria was observed in group D meatball samples. It has been determined that as the temperature and period increases, the growth of psychrotrophic bacteria slows down accordingly. Mol et al. (2012) emphasized that the numbers of psychrotrophic bacteria were lower in whiting fish cooked with the sous vide technique. Çağlak et al. (2017) while the total number of psychrophilic bacteria was 3.72 log CFU/g in fresh zander samples, it was detected below the value of 1.47 log CFU/g in the first 7 days in the sous vide treated groups.

The initial yeast-mold counts of all meatball samples were found to be 2.00 log CFU/g and increased as the storage period progressed. It is thought that this situation is caused by both heat treatment and vacuum packaging. Bozova and İzci (2021), no yeast or mold was detected in meagre fillets applied sous vide during the 42-day storage period. Russo et al. (2023) noted that mussel samples cooked sous vide at temperatures higher

than 80ºC inhibited the growth of yeast and mold.

Differences between groups and storage days were found to be statistically significant (p<0.05) on lactic acid bacteria (LAB) count. LAB counts were determined as 2.00 log CFU/g in all groups at the beginning of storage, and an increase was observed during the storage period. Bongiorno et al. (2018) reported that LAB was not detected in mussel samples after the sous vide process.

Enterobacteriaceae counts were initially detected as 2.00

log CFU/g in all meatball groups. Depending on the heat treatment applied, lower Enterobacteriaceae numbers were obtained in the sous vide method groups compared to the control group during the storage period. Similarly, while the number of Enterebacteriaceae was determined as 2.87 log CFU/g in raw pikeperch fillets, it was determined as 2.00 log CFU/g, <100 log CFU/g and <100 log CFU/g in fillets applied sous vide at different temperatures and times (65ºC 45 min, 75ºC 20 min. and 90ºC 10 min.), respectively (Modzelewska-Kapituła et al., 2022).

Table 2. Microbiological analysis results of count of raw and sous vide-cooked trout meatballs.

Samples	Storage period (days)	Microbiological Analysis (log CFU/g)				
		TMAB	TPAB	TYM	LAB	ENTERO
K	0	3.32±0.23[f]	3.21±0.29[f]	<2.00[e]	<2.00[e]	<2.00[f]
	5	4.99±0.04[e]	4.93±0.10[e]	2.10±0.06[e]	2.41±0.19[d]	2.34±0.12[e]
	10	6.37±0.16[d]	6.36±0.07[d]	3.10±0.24[d]	2.38±0.08[d]	2.70±0.20[d]
	15	7.14±0.13[c]	7.13±0.17[c]	4.73±0.16[c]	2.32±0.04[c]	3.08±0.09[c]
	20	9.41±0.24[b]	7.33±0.16[b]	5.56±0.25[b]	4.13±0.18[b]	3.95±0.08[b]
	25	11.51±0.09[a]	11.05±0.08[a]	6.35±0.13[a]	4.97±0.11[a]	4.65±0.13[a]
A	0	<2.00[e]	<2.00[e]	<2.00[c]	<2.00[d]	<2.00[d]
	5	2.03±0.04[e]	2.11±0.15[e]	<2.00[c]	<2.00[d]	<2.00[d]
	10	2.92±0.04[d]	2.90±0.18[d]	2.14±0.12[c]	2.15±0.01[d]	2.01±0.02[d]
	15	4.04±0.05[c]	3.90±0.04[c]	2.63±0.23[b]	2.68±0.13[c]	2.16±0.01[c]
	20	5.00±0.07[b]	4.95±0.07[b]	3.00±0.00[b]	3.00±0.01[b]	2.83±0.08[b]
	25	5.95±0.05[a]	5.63±0.21[a]	3.51±0.32[a]	3.42±0.19[a]	3.00±0.00[a]
B	0	<2.00[e]	<2.00[d]	<2.00[d]	<2.00[d]	<2.00[c]
	5	2.02±0.03[e]	2.05±0.07[d]	<2.00[d]	<2.00[d]	<2.00[c]
	10	2.57±0.19[d]	2.30±0.21[d]	2.10±0.14[d]	2.04±0.05[d]	<2.00[c]
	15	3.72±0.24[c]	3.40±0.24[c]	2.50±0.11[c]	2.35±0.16[c]	2.07±0.09[c]
	20	4.58±0.21[b]	4.72±0.22[b]	2.95±0.07[b]	2.93±0.04[b]	2.45±0.09[b]
	25	5.78±0.16[a]	5.50±0.41[a]	3.25±0.15[a]	3.15±0.09[a]	2.83±0.04[a]
C	0	<2.00[d]	<2.00[e]	<2.00[c]	<2.00[c]	<2.00[c]
	5	<2.00[d]	<2.00[e]	<2.00[c]	<2.00[c]	<2.00[c]
	10	2.36±0.25[d]	2.35±0.09[d]	<2.00[c]	<2.00[c]	<2.00[c]
	15	3.10±0.19[c]	2.95±0.07[c]	2.11±0.04[c]	2.11±0.15[c]	<2.00[c]
	20	4.07±0.25[b]	3.90±0.14[b]	2.90±0.01[b]	2.67±0.14[b]	2.26±0.12[b]
	25	5.35±0.12[a]	5.07±0.10[a]	3.14±0.19[a]	2.99±0.02[a]	2.54±0.06[a]
D	0	<2.00[d]	<2.00[d]	<2.00[c]	<2.00[b]	<2.00[b]
	5	<2.00[d]	<2.00[d]	<2.00[c]	<2.00[b]	<2.00[b]
	10	<2.00[d]	<2.00[d]	<2.00[c]	<2.00[b]	<2.00[b]
	15	2.59±0.13[c]	2.45±0.16[c]	<2.00[c]	<2.00[b]	<2.00[b]
	20	3.22±0.04[b]	3.13±0.18[b]	2.28±0.12[b]	2.14±0.12[b]	2.07±0.09[b]
	25	4.88±0.14[a]	4.61±0.12[a]	2.85±0.02[a]	2.82±0.10[a]	2.53±0.04[a]

Different letters in the same column indicate statistical difference (p<0.05). K: Control, A: 60ºC for 15 min, B: 60ºC for 20 min, C: 80ºC for 15 min, D: 80ºC for 20 min.

3.2. pH, TBARS and TVB-N

pH, TBARS and TVB-N analysis results of samples are given in Table 3. While it was determined that the group and storage days had a significant effect on the pH value (p<0.05), the interaction of group and storage days did not have a significant effect on the pH value (p>0.05). While the pH value of raw trout meatballs were 6.34 at the beginning of storage, it increased to 6.35, 6.39, 6.42 and 6.45 in groups A, B, C and D after sous vide cooking, respectively. As can be seen in Table 2, the pH value was affected by the cooking process. As the cooking temperature increased, the pH values of the meatball samples also increased because of heat treatment. del Pulgar et al. (2012) reported that this increase in pH was attributed to the formation of disulfide bonds during the cooking process. Becker et al. (2016) stated that increasing temperature causes an increase in pH, mainly due to protein denaturation and change in protein load. Our study is consistent with previous studies showing that the pH value of fish meat increases with increasing temperature (Mohan et al., 2017; Cui et al., 2019; Cropotovaa et al., 2019; Erdem et al., 2022; Pongsetkul et al., 2022).

The difference between the groups was found to be statistically significant (p<0.05) on TBARS value. As can be seen in Table 2, increases were detected depending on the progression of storage time and the highest TBARS value was determined in the control group samples at the end of storage. Varlık et al. (1993), the consumable limit value for fresh fish is between 7-8 mg MDA/1000 g, and it was observed that only the control group samples (8.00±0.20 µmol MA/kg) exceeded this limit value on the 20th day of storage. There are studies showing that the TBARS value decreases as the cooking temperature increases in the sous vide cooking technique for the same period of time. It is thought that decreasing the cooking time due to increasing temperature reduces lipid oxidation. It is also stated that the compounds detected by TBARS analysis are very reactive and the TBARS value may be determined lower since the compounds they form by interacting with different compounds such as amino acids and proteins in meat are not determined as a result of the analysis (Özyürek, 2021). Yuan et al. (2023) found that the TBARS values of Russian sturgeon meat cooked with the sous vide technique increased depending on storage. Karki et al. (2023) stated that the TBARS values of tilapia fish cooked with the sous vide technique were within acceptable consumption limits until the end of storage. Yang et al. (2023) reported that the TBA value of tilapia fillets processed using the sous-vide method (0.47 ± 0.06 mg MDA/kg) was significantly lower compared to the control group (0.69 ± 0.03 mg MDA/kg) (p<0.05). They observed that the sous-vide cooking method has the capacity to effectively inhibit the lipid oxidation process in meat.

While the initial TVB-N value in raw trout meatballs was determined as 8.16 mg/100 g, it were found to be 6.99, 6.56, 5.95 and 5.20 mg/100 g in group A, B, C and D meatball samples after sous vide cooking, respectively. As the storage period increased, TVB-N values increased accordingly. At the end of storage, TVB-N values of all groups were determined below the acceptable limit (≤35 mg/100 g) (Varlık et al., 1993). Similarly, Zhan et al. (2022) found that the TVB-N contents of three groups of scallop samples cooked at different temperatures (70°C and 75°C) with control and sous vide techniques increased with increasing storage time. Kurt Kaya (2022) emphasized that when the quality of marinated crayfish was examined in brine, vacuum packaging and sous-vide cooking, increases were detected depending on time in all groups during storage (p < 0.05). Gokoglu et al. (2024) noted that they determined lower TVB-N values in fish and shrimp cooked with sous-vide compared to control samples. It has been reported that this condition is associated with bacterial growth and microbial proliferation.

Table 3. pH, TVB-N and TBARS values of raw and sous vide-cooked trout meatballs.

Samples	Storage period (days)	Analysis		
		TVB-N (mg/100 g)	TBARS (µmol MA/kg)	pH
K	0	8.16±0.14[f]	1.25±0.07[f]	6.34±0.07[d]
	5	13.61±0.52[e]	2.84±0.09[e]	6.38±0.03[cd]
	10	17.06±0.15[d]	4.30±0.07[d]	6.50±0.02[abc]
	15	20.38±0.24[c]	6.27±0.17[c]	6.48±0.08[bc]
	20	24.40±0.23[b]	8.00±0.20[b]	6.57±0.02[ab]
	25	28.46±0.45[a]	9.65±0.19[a]	6.62±0.02[a]
A	0	6.99±0.07[f]	1.05±0.07[f]	6.35±0.08[d]
	5	10.40±0.16[e]	1.72±0.11[e]	6.45±0.04[cd]
	10	14.73±0.23[d]	2.97±0.15[d]	6.56±0.06[bc]
	15	17.66±0.31[c]	4.37±0.26[c]	6.48±0.03[cd]
	20	21.30±0.17[b]	5.46±0.11[b]	6.64±0.03[ab]
	25	24.70±0.02[a]	6.24±0.13[a]	6.70±0.02[a]

Table 3. (continued)

Samples	Storage period (days)	Analysis		
		TVB-N (mg/100 g)	TBARS (µmol MA/kg)	pH
B	0	6.56±0.14[f]	1.12±0.09[f]	6.39±0.07[c]
	5	9.70±0.25[e]	1.79±0.09[e]	6.49±0.02[bc]
	10	13.90±0.04[d]	3.19±0.06[d]	6.61±0.02[ab]
	15	16.98±0.27[c]	4.70±0.05[c]	6.55±0.07[b]
	20	20.73±0.20[b]	6.00±0.04[b]	6.68±0.03[a]
	25	23.99±0.02[a]	6.72±0.08[a]	6.70±0.11[a]
C	0	5.95±0.08[f]	1.17±0.04[f]	6.42±0.08[c]
	5	9.01±0.17[e]	1.88±0.15[e]	6.55±0.00[bc]
	10	13.17±0.25[d]	2.57±0.28[d]	6.60±0.0[ab]
	15	16.27±0.38[c]	4.36±0.43[c]	6.65±0.07[ab]
	20	20.30±0.22[b]	6.33±0.07[b]	6.75±0.07[a]
	25	23.22±0.05[a]	6.98±0.03[a]	6.75±0.04[a]
D	0	5.20±0.17[f]	1.19±0.01[f]	6.45±0.08[c]
	5	8.72±0.10[e]	2.00±0.07[e]	6.57±0.04[bc]
	10	12.65±0.32[d]	2.99±0.19[d]	6.62±0.02[ab]
	15	15.65±0.28[c]	4.98±0.28[c]	6.73±0.03[a]
	20	19.54±0.28[b]	6.74±0.06[b]	6.74±0.02[a]
	25	22.72±0.09[a]	7.20±0.12[a]	6.75±0.07[a]

Different letters in the same column indicate statistical difference ($p<0.05$). K: Control, A: 60ºC for 15 min, B: 60ºC for 20 min, C: 80ºC for 15 min, D: 80ºC for 20 min.

3.3. Sensory Evaluation Results

Sensory features are one of the important factors that consumers consider (Cai et al., 2021). Sensory evaluation results of samples are given in Figure 1 (a-d). According to the sensory analysis results, it was determined that group and storage days had a significant effect ($p<0.05$), while the interaction of group and storage days did not have a significant effect ($p>0.05$). It was found that the meatballs cooked with the sous vide technique during storage had higher scores than the control. It is thought that this is due to the fact that the aroma components in the product remain in the package by processing the products in vacuum packaging in the sous-vide technique,

increasing their sensory preferability. A decrease was observed in all sample groups in parallel with storage. It was determined that group B meatballs were the most preferred group, receiving the highest score in terms of sensory parameters. Similarly, Ceylan and Gokoglu (2022), emphasized that the sensory scores of squid cooked sous vide at 60 °C were higher than those cooked at 80 °C. Karki et al. (2023) reported that the sensory evaluation scores of tilapia samples cooked with sous vide were lower in the herb supplemented group than in the control group at the beginning of storage. It appeared to significantly improve the texture and overall acceptability of sous vide-treated fish burgers, but had no effect on color or odor (Zhou et al., 2021).
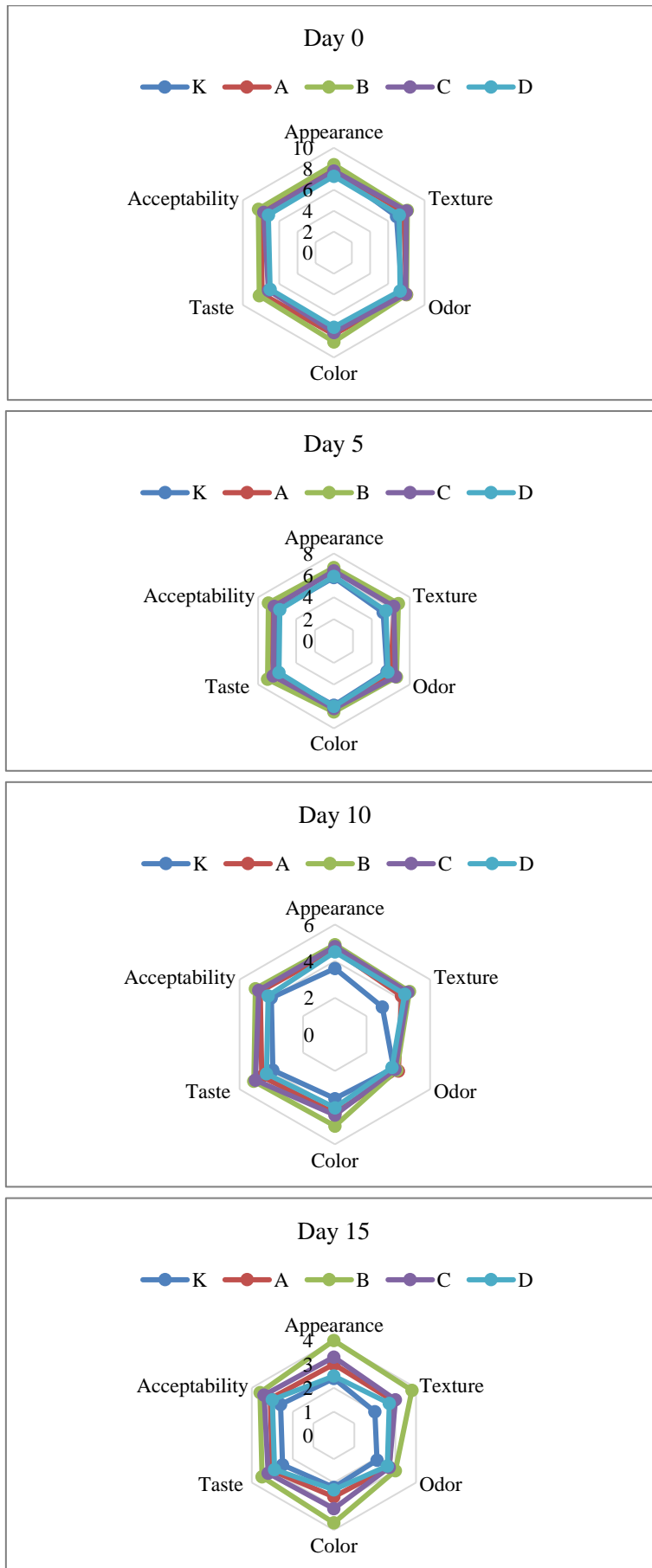
Figure 1. Sensory evaluation results of raw and sous vide-cooked trout meatballs.

4. Conclusion

Today, in addition to traditional cooking methods, there is a need to apply innovative techniques that have many advantages (fast and safe, longer shelf life, preserves nutritional value, and is heated for consumption). Sous vide is a modern culinary and processing method with these advantages. Sous vide cooking significantly affected the chemical, sensory and microbiological properties of rainbow trout meatballs. As a result, it was clearly observed that higher cooking temperature or longer processing time led to lower microorganisms. In addition, it was determined that the TBARS and TVB-N values of the samples applied with this technique remained well below the levels considered inconsumable throughout the storage period. Sensory analysis results showed that sous-vide cooking method increases the flavor of fish and is a good alternative method for cooking it. It was found that the sous vide technique is an effective cooking method on the quality parameters of trout meatballs. While there were a limited number of studies on the application of sous vide technique to fish and fish products in the literature review, no studies were found in which this technique was applied to rainbow trout meatballs. In this context, the results of the research will shed light on future studies.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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RESEARCH ARTICLE

Growth, Yield Components and Tuber Yield Responses of Potato (*Solanum tuberosum* L.) Varieties in High Altitude Regions of Türkiye

Ünlühan Tufan[1] iD • Erdoğan Öztürk[2✉] iD

[1]Food Control Laboratory Directorate, Ankara/Türkiye
[2]Atatürk University, Faculty of Agriculture, Department of Field Crops, Erzurum/Türkiye

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ABSTRACT

Potato is one of Türkiye's strategic crops regarding food and economic benefits. However, the need for well-adapted and productive varieties is a production problem for the country. Therefore, evaluating such varieties with high yield potential and suitable for local environmental conditions is essential. Thus, this study was conducted to select potato varieties with good agronomic characteristics suitable for high-altitude regions such as Erzurum province in 2022. In the study, 12 potato varieties, seven local (Bahar, Kaya, Kafkas, Petek, Taş, Ayaz, Deniz) and five fixed (Pomqueen, Alegria, Marabel, Agria, and Marfona) were used as plant material. It was established according to the "Randomized Blocks" experimental design with three replications. The study's results revealed that the cultivars significantly affected all the variables. The highest plant height was recorded in the Pomqueen (89.6 cm) and Kaya (85.5 cm) varieties, while Kaya had the highest number of stems per hill (6.1 pieces). Concerning yield and yield components, the highest number of tubers per hill was recorded in Ayaz (17.5 pieces), while the highest tuber yield per hill and total tuber yield were recorded in Petek (1287.1 g and 5251.5 kg da^{-1}). This variety was followed by standard Alegria (4497.5 kg da^{-1}) and local Kaya (4441.0 kg da^{-1}) varieties in total tuber yield. Therefore, considering the yield potential, especially local Petek and Kaya and standard varieties Alegria and Marabel were promising and recommended to growers in regions with high altitudes and similar agroecology.

1. Introduction

One of the world's most widespread and vital crops, the potato is a staple food for most of the world's population and has significant health benefits. In world production, potatoes (*Solanum tuberosum* L.) are the world's third largest crop for human consumption after wheat and rice. Rich in carbohydrates, vitamins, protein, and minerals, potatoes are consumed by more than one billion people worldwide (FAO, 2008; CIP, 2010). Therefore, today, the potato crop is an inevitable solution to the problem of population growth and hunger in the world, primarily since it can be grown at many

latitudes and in almost all soil and climatic conditions (except in the equatorial region) (Shitikova et al., 2022).

Since potatoes can be grown from mid-altitude regions to the highest mountain peaks, from humid to dry areas, improvements in their productivity require the development of varieties that are best adapted to a wide range of environments (Kolech et al., 2015). Therefore, plant breeding programs should improve a crop's adaptability and tolerance to biotic stress to increase yield (Tessema et al., 2020).

Producers need high-yielding and stable varieties to reduce food demand and increase yield and quality significantly.

Varieties with these traits can be easily adapted for long-term production in different environments. Therefore, varieties with these traits may overcome the challenges of genotype x variety interactions (Gedif & Yigzaw, 2014).

Variety performance is determined by the genotypic main effect, environmental influences, and the interaction between genotypes and environments (Yan et al., 2001). Hongyu et al. (2014) stated that it is essential to determine varieties' responses to different environments as part of the evaluation process, which will help breeders accurately determine the stability and adaptability of genotypes.

It has been emphasized that it is essential to identify the best-performing varieties on a regional basis to produce high-yielding potato tubers of high marketing and processing quality (Bilate & Mulualem, 2016; Habtamu et al., 2016; Bekele & Haile, 2019). There are no varieties that are difficult to find and have many potentials suitable for all environments and all uses (Bradshaw, 2007). Therefore, evaluating genotype × environment interactions can provide the most appropriate response to environmental adaptation. Tesfaye et al. (2012) reported that genotype, location, and genotype × environment interactions significantly affected dry matter content, starch content, and yield of potato varieties. Similar results were reported by many potato researchers (Abbas et al., 2011; Asefa et al., 2016a; Wassu, 2016; Matin et al., 2017; Nasiruddin et al., 2017).

Selecting the suitable variety for the correct region is a critical parameter, as is taking many measures to grow high-yielding and high-quality potatoes. Nowadays, many variety breeding studies are carried out for potatoes, and these varieties are transferred to different regions to determine the varieties with good adaptation, high quality, and high yield. This study aimed to select the potato varieties developed in our country with high adaptability and superior performance in terms of yield and quality for high-altitude regions such as Erzurum province.

2. Materials and Methods

The research was conducted in the farmer's field in the Yarımca Neighborhood of Aziziye District of Erzurum Province in 2022. The region is at 1853 m, at coordinates 39° 55' N and 41° 61' E. A total of 202.5 mm of rainfall was recorded during the plant growth period (May to October) of the experimental year, with the highest rainfall in May (89.3 mm) and June (80.4 mm). There was no rainfall (0 mm) in August. The average maximum and minimum temperatures during the study months were 9.1 and 22.0 °C, respectively. The highest temperatures were recorded in July (19.3 °C) and August (22.0 °C). As a result of the analysis of the physical and chemical properties of the soil of the test site, it was determined that it was slightly alkaline, low in lime, total nitrogen, and available phosphorus, medium in organic matter, and rich in plant-available potassium.

In the study, 12 potato varieties, seven local (Bahar, Kaya, Kafkas, Honeycomb, Taş, Ayaz, Deniz) and five fixed (Pomqueen, Alegria, Marabel, Agria, and Marfona) developed by Doga Seed in the borders of Gülşehir (The district of Nevşehir province in Türkiye) for different ecologies of the country were used as plant material (Table 1).

Table 1. Potato varieties and some characteristics.

Variety	Origin	Time of Maturity	Utilisation Characteristics
Agria	Germany	Mid Late	Table / Industrial
Alegria	Germany	Mid Early	Table
Ayaz	Türkiye (Nevşehir)	Late	Crisp
Bahar	Türkiye (Nevşehir)	Late	French Fries / Table
Deniz	Türkiye (Nevşehir)	Early	Table
Taş	Türkiye (Nevşehir)	Early	Table
Kafkas	Türkiye (Nevşehir)	Mid Early	Table
Kaya	Türkiye (Nevşehir)	Late	French Fries / Table
Marabel	Austria (Kaltenberg)	Mid Early	Table
Marfona	Netherlands	Mid Early	Table
Petek	Türkiye (Nevşehir)	Mid Early	French Fries / Table
Pomqueen	Germany	Late	French Fries

The research was established using the "Randomized Blocks" experimental design with three replications. In the study's area, farmyard manure was homogeneously mixed with the soil in autumn before planting potatoes and left for winter. In addition, ammonium sulfate (12 kg da^{-1}) of nitrogen fertilizers and 10 kg of phosphorus fertilizer triple super phosphate (P_2O_5) were applied before planting as a supplement to farm manure. Seed potato tubers of the varieties were planted by hand in May 2022 in hills with 70 cm between rows and 35 cm above rows. When the plants reached 5-10 cm in height, the

first and second hoeing were 20-25 days after this hoeing. The earthing up and irrigation procedures were carried out according to climate and soil conditions from the beginning of flowering. Potato harvesting was carried out when the green parts of the potato yellowed and dried from the bottom, the stolons were separated from the main plant and the umbilical cord, and the tuber peel reached an average thickness, unpeelable feature, and a specific size. The data on plant height (cm), number of main stem per hill, number of tuber per hill, tuber yield per hill and total tuber yield (g and kg da⁻¹) were obtained from the two outermost rows and the middle two rows by ignoring the plants at the ends.

2.1. Statistical Analysis

All the data was analyzed using the SPSS package (SPSS, Version 20.0, SPSS Inc, Chicago, IL, USA). When the F-test indicated statistical significance at the $p=0.05$ level, the protected least significant difference (Protected DUNCAN) was used to separate the means (Steel & Torrie, 1980).

3. Results and Discussion

3.1. Analysis of Variance

The results of the analysis of variance showed that potato cultivars had highly significant ($p<0.01$) differences in plant height, number of main stem per hill, number of tuber per hill, tuber yield per hill and total tuber yield (Table 2).

Table 2. F values on yield and yield component response variables of potato (*Solanum tuberosum* L.).

Source	DF	F Values				
		Plant Height	Stem Number Per Hill	Tuber Number Per Hill	Tuber Yield Per Hill	Total Tuber Yield
Block	2					
Variety	11	21.920**	6.220**	13.110**	3.160**	6.506**
Error	22					

**: Highly significant at 1% level of probability; Df: Degree of freedom.

3.2. Mean Performance of Varieties for Growth Traits and Yield Components

The mean plant height of potato varieties varied between 43.3-86.6 cm. The highest height was measured in Pomqueen, and the shortest in Ayaz. The average plant height was 65.4 cm. In terms of the highest plant height, Pomqueen (86.6 cm), Kaya (85.5 cm), and Alegria (79.6 cm) had a statistically insignificant plant height, while they differed significantly from the other varieties. Ayaz (43.3 cm), Marfona (46.9 cm), and Taş (52.6

cm) varieties had shorter plant heights than all other varieties (Table 3, Figure 1). The different ripening periods (early, medium early, medium late, and late) of the potatoes used in the study may have caused variability in terms of plant growth. In addition, plant height is also a cultivar trait. It can be directly related to many factors such as genetic structure, growing period (Karakuş et al., 2011), light period, temperature, irrigation, care treatments, and soil fertility. Therefore, significant differences were found among cultivars.

Table 3. Mean value of plant height, stem and tuber number per hill, tuber yield per hill and total tuber yield as affected by potato varieties.

Variety	Plant Height (cm)	Stem Number Per Hill	Tuber Number Per Hill	Tuber Yield Per Hill (g)	Total Tuber Yield (kg da⁻¹)
Agria	65.0cd	5.7ab	6.5f	834.3de	3344.5cd
Alegria	79.6ab	5.6abc	14.0b	1102.3abc	4497.5ab
Ayaz	43.3f	4.8cde	17.5a	974.3bcd	3974.5bcd
Bahar	67.7c	5.2bcd	9.5e	719.5e	3145.0de
Deniz	74.4bc	5.1bcd	10.5de	1163.3ab	3895.5bcd
Taş	52.6ef	4.2e	10.0de	917.5bcde	3753.7bcd
Kafkas	57.5de	5.4abcd	11.0cde	810.6de	3304.5cd
Kaya	85.5a	6.1a	9.5e	1089.2abc	4441.0ab
Marabel	57.8de	5.9ab	12.5bcd	1048.8abcd	4277.0bc
Marfona	46.9f	4.0e	6.5f	567.9f	2315.0e
Petek	67.8c	4.5de	13.5bc	1287.1a	5251.5a
Pomqueen	86.6a	4.7cde	9.5e	902.0cde	3678.5bcd
Mean	65.4	5.2	10.9	1026.4	3823.2

Means followed by different letters in the same column are significantly different at 1% level of probability.
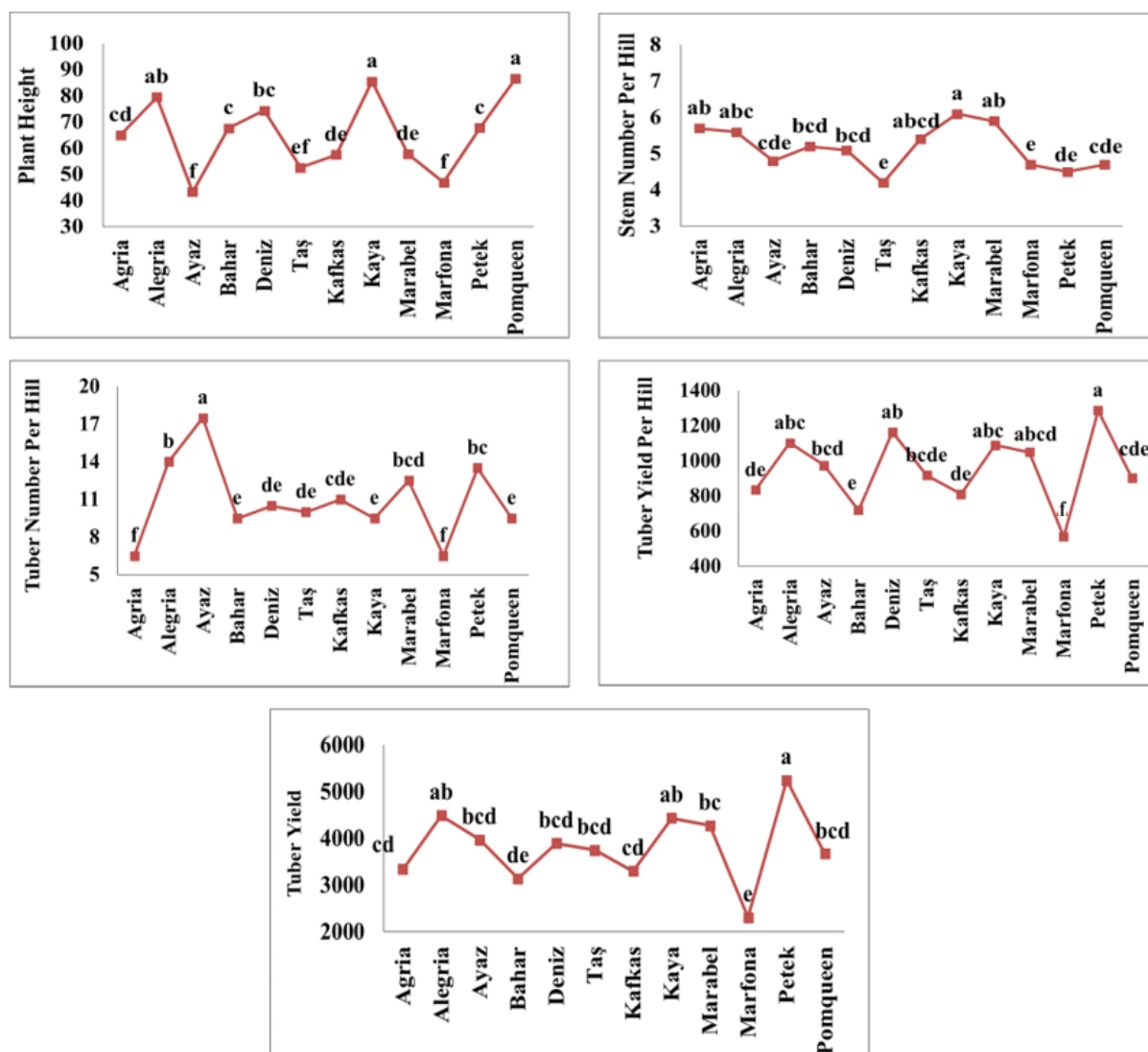
Figure 1. Influence of variety on plant height, stem and tuber number per hill, tuber yield per hill and total tuber yield of potato. Mean values with the same letter within the group are statistically similar at p<0.01.

The number of stems per hill was significantly affected by cultivar. The number of stems is related to the number of branches and leaves contributing to photosynthetic activity. An increase in the uptake of solar radiation can ensure higher photosynthesis and promote the synthesis and accumulation of carbohydrates, positively affecting the final tuber yield (White et al., 2007; Fantaw et al., 2019). Accordingly, the Kaya variety produced 6.1 stems per hill, higher than all other varieties. The lowest number of stems per hill was recorded for Marfona (4.0) and Taş (4.2) varieties (Table 3, Figure 1). This may be attributed to the natural genetic variation in the number of eyes on the tubers. Genetic differences among potato cultivars affect the number of shoots or eyes on tubers and, thus, the number of main stems (Struik & Wiersema, 1999; Getie et al., 2018). Potato varieties can show significant differences in plant height, number of primary branches, and tuber yield (Namugga et al., 2018). Increases in central stem density, which is affected by genetic structure, can lead to a rise in the number of tubers, and the size and weight of these tubers are determinants of potato

yield (Tsegaw, 2005; Zelalem et al., 2009). In other studies on the subject, it has been determined that both plant height and central stem number values of potato varieties show significant variability (Öner & Aytaç, 2016; Kaplan, 2018; Bekele, 2018; Çakır, 2019; Atasever, 2019; Özdemir & Arslanoğlu, 2021; Gül & Sefaoğlu, 2022; Asnake et al., 2023).

The number of tubers per hill of the varieties varied between 6.5 and 17.5 pieces. Ayaz variety ranked first in terms of the number of tubers per hill with 17.5 pieces, followed by Alegria (14.0 pieces), Petek (13.5 pieces), and Marabel (12.5 pieces) varieties. The lowest number of tubers per hill was found in Agria and Marfona varieties (6.5 pieces) (Table 3, Figure 1). When the cultivars used were compared, there was a significant difference in the number of tubers per hill in most of them. Genetic structure and environmental factors influence the formation of Stolon and tuber (Zheng et al., 2018). Asefa et al. (2016b) reported significant differences in the number of tubers per plant due to genetic variation among potato varieties.

Similar studies are reporting that different varieties have different values in terms of the maximum number of tubers per hill under the same and other ecological conditions and that the number of tubers per hill determined in each potato variety varies between 3.8 and 21.6 (Dede, 2004; Öztürk et al., 2008; Kahraman et al., 2017; Kaplan, 2018). In this study, it can be said that ecological differences affect the different tuber numbers of the varieties, as stated by Zheng et al. (2018) and Aliche et al. (2019).

The average tuber yield per hill of potato varieties was highest in local varieties Petek (1287.1 g) and Deniz (1163.3 g) and lowest in standard variety Marfona (567.9 g) and local Bahar variety (719.5 g). Considering that the tuber yield per hill is a joint function of the number of tubers per hill and average single tuber weight, it can be concluded that the factors affecting such characteristics will also be effective on the tuber yield per hill. In studies conducted in different regions on this subject (Şanlı & Karadoğan, 2012; Boydak & Kayantaş, 2017; Kaplan, 2018; Kavalcı, 2019; Özdemir & Arslanoğlu, 2021), it was observed that tuber yield per hill varied between 138.54-1171.0 g.

3.3. Mean Performance of Varieties for Total Tuber Yield

Varieties were found to affect total tuber yield (Table 2). The highest tuber yield was obtained from the local variety Petek (5251.5 kg da^{-1}), followed by Alegria (4497.5 kg da^{-1}), Kaya (4441.0 kg da^{-1}) and Marabel (4277.0 kg da^{-1}). The lowest yield was obtained from the registered foreign variety Marfona (2315.0 kg da^{-1}). In terms of yield, local cultivars such as Petek and Kaya, and other local cultivars, provided higher yields than many registered foreign cultivars, especially Marfona and Agria, which are regionally superior and preferred (Table 3, Figure 1). Although local cultivars such as Petek and Kaya had fewer tubers per plant, their total tuber yields were higher because they produced larger/heavier tubers which increased yield. These results align with studies conducted in different locations and varieties (Girma, 2012; Asefa et al., 2016b). On the other hand, it was also concluded that the increase in yield may be mainly due to the number of tubers per plant (Mehdi et al. 2008; Abubaker et al., 2011).

Therefore, these results suggest that environment and genetic variation significantly affect the differential expression of many of the traits examined, including yield, among potato cultivars. The effects of genotypic and phenotypic variation on total tuber yield, tuber weight, number of main stems, and plant height were reported to be high (>20%) (Shetty et al., 2023). Similar studies have also reported it (Dash et al., 2018; Ebrahim et al., 2018; Getie et al., 2018; Fantaw et al., 2019; Tessema et al., 2020; Asnake et al., 2023). According to the researchers, the improved varieties have higher yield capacity than the standard varieties, which is consistent with the results of the present study.

4. Conclusion and Recommendations

The results confirmed differences between local and standard potato varieties in yield and other relevant agronomic traits in the study area. The study revealed that the production of any of the improved local varieties, such as Petek and Kaya, as well as Ayaz, Deniz, and Taş, resulted in the highest total potato tuber yield compared to standard varieties, such as Marfona and Agria, which are well adapted, productive and of good quality in the region.

Therefore, considering the yield potential, farmers in areas with high altitudes and similar agroecology are advised to prefer local varieties Petek and Kaya, Ayaz, Deniz, and Taş over standard varieties.

Conflict of Interest

The authors declare that they have no conflict of interest.

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