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Comparison of Different Pruning Methods for Training Young Fernor Walnut Trees

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Abstract: The aim of the study is to determine the effects of minimal pruning (MP) and unpruned&unheaded (UP&UH) on young Fernor walnut cultivar trees concerning, tree growth, yield, nut quality and water use efficiency. The trees in the research orchard were planted in 2017. According to the data, a statistically significant difference was found between (MP) and (UP&UH) in terms of shoot length, canopy length, tree height, yield, kernel weight, kernel ratio and nut length. The two-year cumulative yield value was found to be 6036.28 g in (MP) and 9865.87 g in (UP&UH). The stem water potential values (GSP), (MPa) of (MP) and (UP&UH) were 7.65 and 7.50, respectively. Considering the significant difference in yield between two pruning methods, it has been observed that water use efficiency is higher in (UP&UH). If water deficiency and insufficient fertilization are combined with (UP&UH), significant losses in tree growth, yield and nut quality can occur. In this context, (UP&UH) should be considered along with other factors that will affect tree growth, yield and fruit quality.

Keywords: Pruning, Training, Yield, Fernor, Walnut.

Genç Fernor Ceviz Ağaçlarının Yetiştirilmesinde Farklı Budama Yöntemlerinin Karşılaştırılması

Öz: Çalışmanın amacı, minimum budama (MP) ve budamama&tepe vurmamanın (UP&UH) genç Fernor çeşidi ceviz ağaçlarında ağaç gelişimi, verim, meyve kalitesi ve su kullanım etkinliği üzerine etkilerini belirlemekti. Araştırma bahçesindeki ağaçlar 2017 yılında dikildi. Verilere göre (MP) ile (UP&UH) arasında sürgün uzunluğu, taç uzunluğu, ağaç yüksekliği, verim, iç ağırlığı, iç oranı ve meyve boyunda istatistiksel olarak önemli farklılık bulundu. İki yıllık kümülatif verim değeri (MP) ağaçlarında 6036,28 g, (UP&UH) ağaçlarında 9865,87 g olarak belirlendi. (MP) ve (UP&UH)'nin ortalama gün ortası gövde suyu potansiyeli değerleri (MPa) sırasıyla 7,65 ve 7,50'dır. İki budama yöntemi arasındaki verim farkı dikkate alındığında (MP) uygulamasında su kullanım etkinliğinin daha yüksek olduğu görüldü. Sulama suyu yetersizliği ve yetersiz bitki besleme&gübreleme yönetimi (MP&UH) ile birleştiğinde ağaç gelişiminde, verimde ve meyve kalitesinde önemli kayıplara neden olabilir. Bu bağlamda (MP&UH) uygulaması, ağaç gelişimi, verim ve meyve kalitesini etkileyecek diğer faktörlerle birlikte düşünülmelidir.

Anahtar Kelimeler: Budama, Şekil budaması, Verim, Fernor, Ceviz.

1.Introduction

Training and pruning of adult walnut trees serve purposes: controlling tree size balancing generative and vegetative development, enhancing nut size, promoting the formation of new fruit buds, ensuring proper light distribution within the tree, and eliminating old, dried, dead or overlapping branches. Additionally pruning aims to shape the tree suitably for harvesting and picking operations by machinery. Another objective of pruning in mature walnut trees is to stimulate strong spur development, encourage female flower formation, and facilitate the growth of large nuts. (Akça, 2014; Andersen, 1984).

In walnut varieties that bear fruit on terminal branches, pruning may delay yield. However, this effect is not seen in varieties that produce high amounts of fruit on lateral branches. Significant differences exist between varieties regarding development type, fruiting location, vigor and many other characteristics. If young fruit-bearing trees are minimally pruned, ıt can encourage sufficient shoot development to form fruit areas Conversely, heavy pruning of young trees may delay the onset of yielding.

Studies investigating the effects of different pruning methods on yield and fruit quality in walnut cultivation in Turkey are very limited (Argaç, 2021). According to our observations in walnut orchards where hard pruning is practiced in walnut cultivation, our initial results indicate that the shoots are susceptible to damage from the onset of yield is delayed, productivity is low and increased susceptibility to *Xaj* disease.

Similar research has been conducted on Ashley, Howard and Chandler varieties (Hasey et al., 1998; DeBuse et al., 2010). However, there is no study on the Fernor variety. Fernor trees have different morphological and physiological characteristics compared to the Ashley, Howard and Chandler varieties, their responses to different pruning systems remain unknown.

Walnut growers in Türkiye prefer very hard pruning over minimal pruning. There is a common belief that minimal or no pruning is not appropriate in walnut cultivation. Growers hold belief that with hard pruning, the tree will have a more uniform shape and stronger growth. In fact, it is widely believed that if young walnut trees are not pruned, their development will stagnate, leading to decreased yield and fruit quality due to early fruit bearing. Consequently , very hard pruning is performed on young trees. When we realized that we could not change habits with rhetoric, we realized that we needed applied research results. The only way to overcome resistance to paradigm change should be persuasion with results based on research.

It has been reported that young trees of the Howard, Chandler, Tulare, Forde, Solano and Livermore varieties do not require pruning to maintain growth and achieve adequate yield. Heavy pruning is not recommended for these cultivars as it leads to reduced yields and smaller trees. Pruning may delay yield on varieties that bear fruit on terminal branches, such as Hartley and Franquette. However, this effect is not observed in cultivars that bear fruit on lateral branches (Hasey et al., 1998).

There is no research investigating the effects of different pruning methods on yield in young Fernor walnut trees. The use of hard pruning methods, especially in the Fernor variety, delays the age at which trees start yielding. In our research, we investigated the effects of pruning management on tree development, yield, nut quality and water stress in young Fernor trees. Additionally, our study aimed to provide new results against the hypothesis that pruning reduces tree growth in varieties that bear fruit on lateral branches.

2.Material and Method

2.1. Material

2.1.1. Plant Material

In 2017, an extensive walnut orchard was planted in the Northeast of Turkey, in Lüleburgaz (41°18'44.08"N & 27°17'42.64"E), with trees spaced at 8m x 4m grafted 'Fernor' trees onto *J. regia* L. rootstock. Fernor walnut cultivar has the characteristics of late leafing and lateral bud fruitfulness. Fernor is the most used cultivar after the Chandler variety in walnut orchards established in Turkey in recent years.

2.1.2. Soil characteristics of the orchard where the research was conducted

Physical and chemical properties of the soils of the research orchard presented in Table 1.

Table 1. Physical and chemical properties of soils
Çizelge 1. *Toprakların fiziksel ve kimyasal özellikleri*

Parametre	Results	Unite	Metod
Soil pH	7.50	%	-
Eletrical conductivity (EC)	0.04	%	Saturation
CaCO3	1.95	%	Calsimetric
Saturation	74.80	%	Saturation
Organic matter (OM)	1.49	%	Walkey-Black
(N)	0.07	ppm	Kjeldahl
P	7.07	ppm	Olsen
K	191.54	ppm	Amonium Asetat - ICP
Ca	8 014.23	ppm	Amonium Asetat- ICP
Mg	740.81	ppm	Amonium Asetat- ICP
Fe	21.11	ppm	DTPA-ICP
Cu	1.68	ppm	DTPA-ICP
Zn	0.37	ppm	DTPA-ICP
Mn	8.50	ppm	DTPA-ICP

2.1.3. Devices used to measure length and weight

Meters were used to measure trunk length, shoot length, tree height, and canopy and tree width. An electronic caliper with 0.01 mm precision was used to determine fruit sizes from pomological characteristics, and an electronic scale with 0.01 g precision was used to determine nut and kernel weight.

2.2. Method

2.2.1. Preparation practices for different pruning methods on saplings

Fernor saplings were initially over 2.00 meters high and were pruned to 50 cm upon planting. Drip irrigation was installed from the outset, and orchard management followed standard practices appropriate to the environment and the age of the plantation. Before onset of the second growing season trees were cut at 1.80 m and 5-6 scaffold branches were selected, including the leading branch. Subsequently main shoot leader and scaffold branches were pruned during in the winter training, removing either 1/3 or 2/3 of their length depending on their annual growth, typically around a meter . During the three first year's stakes were used. The objective was to achieve 5 to 6 limbs, growing the first at 1.50 m from the soil. A minimal pruning system was applied to trial trees in 2018 and 2019 to establish a main branch system that would allow adequate light penetration (Ryugo et al., 1980). Trials of two different pruning systems started in 2020.

2.2.1.1.Minimum pruning method (MP)

In 2020 and 2021, the branches of the trees that will undergo (MP) were cut below 150 cm from the soil level, and 25% to 30% of the shoots from the previous year were cut and headed (Figure 1).

2.2.1.2.Unpruned/Unheaded training method (UP&UH)

In 2020, in (UP&UH), only narrow-angle and forked branches and branches below 150 cm on the trunk were removed. All other branches were unpruned and unheaded. Trees that were not crowned and cut under management UP&UH in 2020-2021 were released. However, during the summer pruning of these trees, only the forked branches, if any, were reduced to single branches (Figure 1).

2.2.2.Determination of the effects of pruning management on phenological, morphological, pomological and yield

In order to determine the effect of pruning management on phenological characteristics, data on bud burst time, leafing time, female flowering time, male flowering time, harvest time and leaf fall time were taken.

Figure 1. Crown shape of trees treated with (MP) (left) and (UP&UH) (right)
Resim 1. *(MP) (solda) ve (UP&UH) (sağda) uygulama yapılan ağaçların taç şekli*

In order to assess the act of pruning management on morphological characteristics, various parameters were examined, including tree trunk circumference, tree height, tree and canopy volume, canopy length and width.

In order to assess the act of pruning management on morphological characteristics, various parameters were examined, including tree trunk circumference, tree height, tree and canopy volume, canopy length and width.

Additionally, the yield per tree was determined to evaluate the effect of pruning methods on yield (g/tree). Tree volume (TV) and crown volume (CV) were calculated based on Equation 1 (Stehr, 2005) and Equation 2 (Argaç, 2001).

$$TV(m^3)=[(L+W)/4)^2.\pi.H]/2 \quad (1)$$

L: Crown height
W: Crown width
H: Tree height

$$CV(m^3)=\pi r^2 h/2 \quad (2)$$

h: Crown height
r: Crown width

In order to determine the effect of pruning methods on pomological characteristics, nut weight, kernel weight, kernel percentage shell thickness, nut dimensions (length, height, width) and kernel color characteristics were examined. Pomological characteristics were examined in 20 nuts randomly taken from 12 trees selected for each application.

2.2.3. Effect of pruning managements on plant water stress

Irrigation time, irrigation interval and irrigation water amount were determined using climate data obtained from the climate station in the research orchard. Soil moisture values obtained from digital soil moisture sensors.

To directly determine the effects of two different pruning treatments on plant water stress, stem water potential (GSP) values were measured with a pressure chamber device between in June and October. Measurements were conducted on Thursdays between 12.00 and 13.30 on three trees selected from each of the trial plots. Measurements were made on leaves selected from the south side of the tree. The leaf was wrapped with foil and the measurement was taken after waiting for 10 minutes. Leaf water potential was determined using a PMS 615 model device (Fulton et al., 2001).

2.2.4. Plant nutrition and leaf macro and micro element contents

Leaf and soil analyses were conducted to determine plant nutrition management, and yield served as a basis for adjustments. A total of 150 kg of nitrogen (N), 90 kg of potassium (K), and 40 kg of phosphorus (P) were applied per hectare. Additionally foliar fertilization with combined microelements was applied throughout the season. The effects of (MP) and (UP&UH) applications on leaf macro and micro element contents were examined.

2.2.5. Evaluation of data

The experiment was set up with 3 replications and 4 trees were included in each replication. The effects of two different pruning methods on morphology, pomology and yield were analyzed in the SAS statistical program and comparisons were made with the Duncan test.

3. Results and Discussion

The aim of the research is to determine the effects of (MP) and (UP&UH) on young trees of Fernor variety on the growth vigor, yield, nut quality and water use efficiency of the trees.

3.1. Effect of pruning methods on phenological characteristics

The leafing time, the time of receptive period in female flower and the time of pollen shedding of the trees that received (MP) were observed approximately 5 days earlier than the trees that (UP&UH). Harvest time and leaf fall dates were found to be similar for two pruning managements.

3.2. Effect of pruning methods on morphological characteristics

3.2.1. Tree trunk circumference

In (MP), the average tree trunk circumference values were measured between 37.70 cm and 43.50 cm, with and the average trunk circumference value calculated as 39.61±1.69 cm. In (UP&UH), the trunk circumference ranged from 35.50 cm to 41.80 cm with the average trunk circumference recorded as 38.46±1.89 cm (Table 2).

The effect of two pruning methods on the trunk circumference was found to be insignificant (Table 1). Similar results were reported in a study on Chandler walnut variety, where the effect of training systems on trunk circumference was also found to be insignificant. Moreover, investigations on four different pruning methods in Chandler cultivar revealed that trees without pruning exhibited the highest trunk circumference (126.90), while trees subjected to minimal pruning showed the lowest trunk diameter (Aletà et al., 2006). Like our research results, the effect of minimum pruning and non-pruning management on the tree trunk environment was found to be statistically insignificant (Argaç 2021). The effects of hard pruning, minimum-low level pruning, minimum pruning and unpruned/unheaded treatments on the trunk circumference were found to be insignificant in Gillet, Forde and Tulare varieties. However, in Chandler variety, a significant difference was found between severe pruning (27.9 cm) and other pruning methods (29.7-32.0 cm). Similar to our research results, the lowest trunk circumference value was found in the Tulare variety under unpruned/unheaded methods, and the highest value was found in the Gillet variety under minimum pruning (DeBuse et al., 2010).

Table 2. Effect of different pruning methods on morphological characteristics
Çizelge 2. *Farklı budama yöntemlerinin morfolojik özellikler üzerine etkisi*

Pruning Methods	Tree trunk circumference (cm)	Tree height (m)	Tree volume (m^3)	Canopy volume (m^3)	Canopy length (m)	Canopy width (m)	Shoot length (cm)
(MP)	39,61^a	5,50^b	36,32^a	18.28^a	4,00^b	4,17^a	90,28^a
(UP&UH)	38,46^a	5,71^a	41,12^a	20.91^a	4,23^a	4,29^a	33.32^b

*The difference between means shown with different letters in the same column is significant (P<0.05)

3.2.2. Tree height (m)

In (MP), tree height were measured between 4,90 and 5,76 m, and the average tree height was 5,50±0.27 m. In (UP&UH), tree height varied from 5.48 m to 6.35 m, and the average tree height was 5.71±0.22 m. A significant difference ($P<0.05$) was found in the tree heights between (MP) and (UP&UH) (Table 2). DeBuse et al. (2010) reported that different pruning systems had no significant effect on tree height, but they noted that the highest tree height was observed in minimally pruned trees. Similarly, in our research, the height of (UP&UH) trees was found to be higher than (MP) trees, consistent with the results of Argaç (2021).

3.2.3. Tree and canopy volume (m^3)

In (MP), tree volume (m^3) varied between 24,93 and 42,66 and the average canopy volume was 36,32±6,35. In (UP&UH), the tree volume (m^3) ranged from 33,12 to 52,17 and the average tree volume was 41,12±5,12 (Table 1). In (MP), canopy volume (m^3) varied between 11.62 -23.99 and the average canopy volume 18.28±3.67. In (UP&UH), the canopy volume ranged from 13.92 m^3-26.54 $m^3$and the average canopy volume was 20.91±3.20 m^3 (Table 2).

There was no statistical difference between the tree volume and canopy volume of trees with (MP) and (UP&UH) (Table 1). Similar to our research results, Argaç (2021) stated that the effect of non-pruning and minimum pruning on the tree and canopy volume is insignificant, but the tree and canopy volume values of trees without pruning are higher than the values of trees with minimum pruning.

3.2.4. Canopy length and width (m)

In (MP), the canopy length varied between 3.40 and 4.29 m, and the average canopy length was 4.00±0.27m. In (UP&UH), the canopy length was found between 3.98 and 4.85 m, and the average canopy length was 4.23±0.23 m. A statistically significant difference was found between the canopy length of trees with (MP) and (UP&UH) (Table 2).

In (MP), the canopy width varied between 3.45 and 4.80m, and the average canopy length was 4.17±0.35m. In (UP&UH), the canopy width was found between 3.58 and 4.95 m, and the average canopy width was 4.29±0.23 m. A statistically insignificant difference was found between the canopy width of trees with (MP) and (UP&UH) (Table 2).

3.2.5. Shoot length (cm)

In (MP), the shoot length varied between 79,6.0 to 106.0 cm and the average shoot length was 90,28±7,38 cm. In (UP&UH), the shoot length varied between 20,06 to 41,06 cm and the average shoot length was 33,32±6,38 cm. A statistically significant difference was found between the shoot length of trees with (MP) and (UP&UH) (Table 2).

Dalkılıç et al., (2005) compared the effects of six different intensities of pruning on vegetative growth in eight-year-old trees of Yalova-4 and Bilecik walnut varieties. They found significant effects of different pruning severities on vegetative development. Specifically, significant effects of pruning intensities on the number of annual shoots, total shoot length, and total number of buds were observed in the Yalova variety. The effect of different pruning intensities on the number of annual shoots in the Bilecik variety was found to be insignificant.

3.2.6. Average yield (g/tree)

According to two years, average yield values per tree in (MP) varied between 1707.28g and 4329.4g. The average yield of (UP&UH) management was between 2829.62 and 7036.25. The two-year cumulative yield was found 6036.68 g in (MP) and 9865.87 g in (UP&UH). The cumulative yield of (UP&UH) was determined to be 61.18% higher than that of (MP) management (Table 3). A significant difference ($P<0.05$) was observed between the yield values of (MP) and (UP&UH) (Table 3).

Table 3. Effect of different pruning methods on yield (g/tree)

***Çizelge 3**. Farklı budama yöntemlerinin verim üzerine etkisi (g/ağaç)*

Pruning Methods	Average Yield (g/ tree)		Cumulative y ield (g/tree)
	2021	2r022	
(MP)	1707.28 b	4329.4 b	6036,28b
(UP&UH)	2829.62 a *	7036.25 a *	9865,87a

* The difference between means shown with different letters in the same column is significant ($P<0.05$)

In the study comparing the yield values of Chandler trees with minimal pruning and no pruning in Bursa ecological conditions, it was reported that trees without pruning had a higher yield of 40.56% compared to trees with minimum pruning, which is consistent with our research results (Argaç 2021). Some studies have indicated that no significant difference in yield resulting from pruning walnut trees, but these results are from studies on mature trees. Observations conducted at UC Davis orchards and grower orchards in California have shown that walnut trees can grow without pruning even in the early years (Lampinen et al., 2015). The effects of hard pruning, minimum-low level pruning, minimum pruning and no-topping-no-pruning management on yield were found to be insignificant in Tulare variety and significant in other varieties (Chandler, Gillet, Forde). The highest yield value was found in unpruned and unheaded management, consist with our research results (DeBuse et al., 2010). Olsen et al., (1990), reported that the yield value of annually pruned trees did not significantly differ from that of unpruned trees, in contrast to our research results. According to Lampinen et al., (2015), the yield of trees that are not pruned except for removing branches that get in the way of shakers or tractors has a cumulative yield similar to trees that are pruned every year for the first 7 years.

3.2.8.Effect of pruning methods on pomological characteristics

The kernel colors of the trees applied to two different pruning methods were found to be similar (Figure 2).

In (MP), average nut weight was found as 11.57±0.59 (g), kernel weight was 5.09±0.40 (g), shell thickness (mm) was 1.85±0.15, nut length (mm) was 37.12±0.53, nut height (mm) was 32.55±0.51, nut width (mm) was 31.23±0.88 in 2021 year. In (MP), average nut weight was found as 13.30±0.42 (g), kernel weight was 6.37±0.36, shell thickness (mm) was 1.58±0.09, nut length (mm) was 40.75±0.95, nut height (mm) was 34.43±0.61, nut width (mm) was 32.56±0.54 in 2022 year (Table 4).

In (UP&UH), average nut weight was 12.01±0.48 (g), kernel weight was 5.32±0.27 (g), shell thickness (mm) was 1.79±0.16, nut length (mm) was 38.11±0.81, nut height (mm) was 33.84±1.15, nut width (mm) was 32.13±1.09 in 2021 year (Table 4).

In (UP&UH), average nut weight was found as 13.44±0.43 (g), kernel weight was 6.69±0.32 (g), shell thickness (mm) was 1.64±0.1, nut length 41.15±1.02 (mm) nut height was 34.35±0.45, nut width (mm) was 32.81±0.54in 2022 year (Table 4). According to 2021 data, there was a significant difference (P<0.05) between pruning methods in terms of nut length and nut height values . In the 2022 data, differences were found in terms of kernel weight and kernel percentage (Table 4). However, the effect of pruning methods on other pomological characteristics was found to be insignificant.

Nuts of (MP) | Nuts of (UP&UH)

Figure 2. The nuts of (MP) and (UP&UH)
Resim 2. *Minimum budama ve budamama&tepe vurmama uygulanan ağaçların meyveleri*

Table 4. Effect of different pruning methods on pomological characteristics
Çizelge 4. *Farklı budama yöntemlerinin pomolojik özellikler üzerine etkisi*

Pruning Methods	Nut weight (g)	Kernel weight (g)	Kernel percentage (%)	Shell thickness (mm)	Nut length (mm)	Nut height (mm)	Nut width (mm)
			2021				
(MP)	11.57 a	5.09 a	43.97 a	1.85 a	37.12 b	32.55 b	31.23 a
(UP&UH)	12.01 a	5.32 a	44.28 a	1.79 a	38.11 a*	33.84 a*	32.13 a
			2022				
(MP)	13.30 a	6.37 b	47.70 b	1.58 a	40.75 a	34.43 a	32.56 a
(UP&UH)	13.44 a	6.69 a*	50.57 a*	1.64 a	41.15 a	34.35 a	32.81 a

*The difference between means shown with different letters in the same column is significant (P<0.05)

Argaç (2021) reported no difference between nut width, shell thickness, kernel weight and kernel percentage of trees with minimal pruning and non-pruning trees. However there was a significant difference between management practices in terms of nut weight. Conversely , Olsen et al., (1990) stated that there were losses in nut size and nut quality in unpruned walnut orchards. The inconsistency of the results regarding the effects of pruning methods on nut quality may be attributed to variations in the walnut varieties used in the research, rootstock variety combinations, irrigation, plant nutrition, disease and pest management strategies.

3.2.9.Effect of pruning methods on midday stem water potential

In order to determine the effects of pruning management on plant water stress, leaf water potential values were measured and the results are presented in Table 5. Average stem water potential values were found as 7.65 in trees with (MP), and 7.5 in trees with (UP&UH). The difference between (MP) and (UP&UH) was found to be approximately 0.15 MPa (Table 5). There was no statistically significant difference between pruning management methods in terms of seasonal midday trunk water potential values (Table 5). Lampinen et al., (2015) reported that, similar to our research results, the effect of different pruning systems on water stress was minimal. They found no significant difference in average seasonal midday trunk water potential values between pruning treatments in 2005, 2006 and 2009. In 2007, 2008 and 2010, slight differences were detected (approximately 0.05 MPa). In our study, the irrigation management of the trees subjected to two different pruning methods was similar; thus, the amount of water provided to the trial trees and the irrigation timing were consistent.

Table 5. Midday trunk water potential values (MPa) of different pruning methods
Çizelge 5. *Farklı budama yöntemlerinin gövde su potansiyeli üzerine etkisi*

Date	(MP)	((UP&UH))
July 1, 2022	7.0	7.5
July 15, 2022	7.0	6.0
August 1, 2022	8.5	8.5
August 15, 2022	7.5	7.5
September 1, 2022	6.5	6.0
September 15, 2022	10.0	10.5
October 1, 2022	7.0	7.0
October 15, 2022	7.5	7.0
Average	7.65[a]	7.5[a]*

*The difference between means shown with different letters in the same column is significant (P<0.05)

Although the yield values (gr/tree) of no-pruning management were higher, the water use efficiency values of these trees were also observed to be high. As a matter of fact, similar to our research results, Lampinen et al., (2008) reported that unpruned trees had higher yield than pruned trees with the same amount of water.

3.2.10. The content of macro and micro elements of leaf

The content of macro and micro elements of leaf in (MP) and (UP&UH) are presented in Table 6. The macro and micro element contents of the leaves of the trees to which two different pruning methods were applied were found to be similar. The nitrogen, potassium and zinc contents of the leaves were found to be insufficient, but the copper content was high (Table 6)

Table 6. The content of macro and micro elements of leaf in MP and (UP&UH)
Çizelge 6. MP and (UP&UH) *budama yöntemlerinin yaprak makro ve mikro element içerikleri*

	(MP)	(UP&UH)	Unite	Metod
N	2.19	2.3	%	Kjeldahl
P	0.16	0.15	%	Wet digestion-ICP
K	1.15	1.04	%	" "
Ca	2.2	1.76	%	" "
Mg	0.65	0.49	%	" "
Fe	95	78	ppm	" "
Cu	130	118	ppm	" "
Zn	8	9.22	ppm	" "
Mn	85	97	ppm	" "

4. Conclusion

With (UP&UH), the costs of pruning labor and collecting pruning pieces are eliminated. Moreover, the earlier and higher yield of trees in the initial years positively impacts orchard management costs. In our study, unlike others , pruning management was started after a balanced and regular main skeleton was achieved on the canopy. It is estimated that this practice will increase light penetration into the trees canopy. According to preliminary results, the results contradict the hypothesis that young walnut trees that are not pruned will experience growth stagnation, crown fruit branches dying, and tree size reduction.

For pruning management to be recommended despite its positive results, ecological conditions, plant nutrition and irrigation management must also be considered. If (UP&UH) management is combined with inadequate irrigation and suboptimal plant nutrition, significant setback in tree development, yield and nut quality may occur. Consequently , pruning management should be integrated with other factors influencing tree development, yield and fruit quality. It should not be implemented in walnut orchards without considering integrated orchard management. Additionally, for more definitive and consistent results, the research should be continued for at least 5 more years. Final decisions should be based on the findings obtained in the subsequent years.

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Effects of Girdling Treatments at Different Periods and Width on Grape Quality in Yalova Çekirdeksizi (*V. vinifera* L.) Grape Variety

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Abstract: This research was carried out in 2022 and 2023 to determine the effects of different width and single–double girdling treatments on grape quality in different phenologic periods of 'Yalova Çekirdeksizi' grape variety grown in the 'Table Grape Varieties Application and Research Vineyard' located in the 'ÇOMÜ Dardanos Campus, Faculty of Agriculture, Plant Production Research and Application Unit'. Within the scope of the research, a total of 9 treatments, including the control, were carried out in two different phenologic periods pre–bloom and post–berry set, followed by two different widths of single repeated girdling treatments, and double repeated girdling treatments during the veraison period. As a result; the numeral decrease in yield in the 5 mm girdling treatments (4.33 kg grapevine^{-1}) may be due to the removal of a wider bark+phloem layer, resulting in a later closure of the wound tissues compared to the 3 mm girdling treatments (4.95 kg grapevine^{-1}), and thus a decrease in assimilate products stored in the main root and old parts. Therefore, it was concluded that although 5 mm girdling treatments continue to give positive results in terms of grape quality in many parameters, it is not appropriate to repeat them more than one year in terms of average yield.

Keywords: Girdling, phenologic period, quality, *V. vinifera* L., yield.

Yalova Çekirdeksizi (*V. vinifera* L.) Üzüm Çeşidinde Farklı Dönem ve Genişliklerde Yapılan Bilezik Alma Uygulamalarının Üzüm Kalitesi Üzerindeki Etkileri

Öz: Bu araştırmada, 'ÇOMÜ Dardanos Yerleşkesi Ziraat Fakültesi Bitkisel Üretim Araştırma ve Uygulama Birimi'ndeki 'Sofralık Üzüm Çeşitleri Uygulama ve Araştırma Bağı'nda yetiştirilen Yalova Çekirdeksizi üzüm çeşidinde, farklı fenolojik dönemlerde, farklı genişlikte ve tek–çift bilezik alma uygulamalarının üzüm kalitesi üzerindeki etkilerinin belirlenmesi amacıyla, 2022 ve 2023 yıllarında yürütülmüştür. Araştırma kapsamında, çiçeklenme öncesi ve tane tutumu sonrası iki farklı fenolojik dönemde, iki farklı genişlikte tek tekrarlı bilezik alma uygulamalarının ardından, ben düşme döneminde çift tekrarlı bilezik alma uygulamaları da yapılarak, kontrol dâhil toplam 9 uygulama gerçekleştirilmiştir. Sonuç olarak; 5 mm bilezik alma uygulamalarındaki (4.33 kg omca^{-1}) rakamsal verim azalışının, daha kalın bir kabuk+floem tabakasının çıkartılması sonucunda 3 mm bilezik alma uygulamalarına (4.95 kg omca^{-1}) kıyasla yara yerlerinin daha geç kapanması neticesinde, ana kök ve yaşlı kısımlarda depolanan rezerv maddelerdeki azalışa bağlı olarak gerçekleşmiş olabileceği düşünülmektedir. Bu nedenle, 5 mm bilezik alma uygulamalarının birçok parametrede üzüm kalitesi yönünden olumlu sonuçlar vermeye devam etse de, ortalama verim açısından bir yıldan fazla tekrarının uygun olmadığı sonucuna varılmıştır.

Anahtar kelimeler: Bilezik alma, fenolojik dönem, kalite, verim, *V. vinifera* L.

1. Introduction

Grapes (*Vitis vinifera* L.), considered one of the most important commercial fruit crops of temperate and tropical regions, have gained popularity due to many factors such as its nutrient–rich value, containing compounds beneficial to human health and the diversity of its utilization. Türkiye, which is located in the temperate climate zone ideal for grape growing worldwide, is ranked among the important countries in the viticulture sector in the world with its yield values and grape production areas. In 2022, a total of 87.615.444 tons of grape production was realized in the world, and Türkiye has an important position in terms of grape cultivation, ranking fifth in the world after Spain, France, Italy and China with 384.537 ha of vineyard area and sixth in the world after China, Italy, France, Spain and the USA with 4.165.000 tons of grape production (FAO, 2024). Of the grape production, 50.42% is table grapes (2.099.859 tons), 40.38% is dried grapes (1.681.808 tons) and 9.20% is wine grapes (383.333 tons) (TUIK, 2024).

The aim of table grapes growing, which constitute half of the grape production in Türkiye, is to produce grapes with high yield and quality. The quality of table grapes consists of medium–sized clusters with uniformly sized berries, variety–specific skin color, aroma composition and other phytochemical contents. Consumers pay attention to the amount and content of phytochemicals that contribute to human health and come to the forefront in recent years, along with physical properties such as shape, color and form in table grapes (Kunter et al. 2013; Crupi et al., 2016; Cantürk et al., 2018a,b Nicolosi et al., 2018). For this purpose, some plant growth regulators and canopy management techniques such as crop load, shoot tipping and shoot topping removal, axillary shoot removal, cluster thinning, cluster tip cutting, etc. and some plant growth regulators are applied in vinestock to increase grape quality (Winkler, 1974; Türker & Dardeniz, 2014; Camcı & Çoban, 2016; Bahar et al., 2017; Korkutal et al., 2018; Şahin & Dardeniz, 2023). Another one of these cultural techniques to improve grape quality is girdling. Girdling is a technique for regulating phloem transport between grapevine canopy and roots by ensuring the distribution of photosynthesis products, plant regulators and nutrients. Girdling is carried out by removing the 3–6 mm width bark+phloem layer from both the trunk and the base internode of the canes of the grapevines with special clippers. In the parts where the girdling is taken, the phloem bridges take their previous form with the formation of callus, and the wound areas heal in approximately 3–6 weeks, but during the callus formation period, the nutrients that are expected to go to the roots remain in the canopy area and direct to the clusters, and their density increases. The effect of girdling varies depending on the period, environmental conditions and grape varieties (Carreño et al., 1998). Girdling treatments pre–bloom increases berry set (Jackson, 1985; Abu–Zahara, 2010) and grape yield, increases cluster sizes (Goren et al., 2004) and delays grape maturity (Rammings & Tarailo, 1998; Crupi et al., 2016). Girdling treatments post–berry set increases cluster–berry sizes (Abu–Zahara, 2010) and grape yield (Carreño et al., 1998; Gözcü & Dardeniz, 2022) and has positive effects on grape maturity (Keskin et al., 2013). During the veraison period, it increases the total soluble solids (TSS) ratio (Koshita et al., 2011) and enhanced grape maturity (Carreño et al., 1998; Çiftçi & Çelik, 2023), and improves the coloration of the berries in colored grape varieties by reducing acidity (Crupi et al., 2016). is improving (Carreño et al., 1998; Camcı & Çoban, 2016; Çiftçi & Çelik, 2023).

In the cultivation of grapes or a different species, the treatment period and repetition of the target appropriate girdling and the width of the bark+ phloem layer to be taken are of great importance. More research is needed on this subject based on grape varieties.

In this research, it was aimed to determine the effects of different width (3 mm and 5 mm) and single–double girdling treatments on grape quality in different phenologic periods (pre–bloom and post–berry set) in Yalova Çekirdeksizi grape variety grown in the 'Table Grape Varieties Application and Research Vineyard' located in the 'ÇOMÜ Dardanos Campus, Faculty of Agriculture Plant Production Research and Application Unit'.

2. Material and Method

This research was carried out in 2022 and 2023 on Yalova Çekirdeksizi grape variety grown in the 'Table Grape Varieties Application and Research Vineyard' located in the 'ÇOMÜ Dardanos Campus, Faculty of Agriculture Plant Production Research and Application Unit' located at 40° 4' 26.40" N latitude and 84 26° 21' 42.84" E longitude. The vineyard where the research was conducted was established with 2.0 da and 3.0 x 1.5 meter row spacing and intra–row spacing distances. The vines of Yalova Çekirdeksizi grape variety grafted on 5BB American grapevine rootstock have a Lenz–Moser bilateral fixed cordon system and are 19 years old as of the year the research was initiated.

In the Yalova Çekirdeksizi grape variety, a total of nine treatments, including the control, were carried out, following single repetitive girdling treatments in two different widths (3 mm ve 5 mm) in two different phenologic periods pre–bloom (EL–18; 24 May 2022; 30 May 2023) and post–berry set (EL–27; 9 June 2022; 20 June 2023), and double repetitive girdling treatments (EL–35; 19 July 2022; 24 July 2023) were made during the veraison period.

Winter pruning was carried out as cane pruning in March of the years in which the research was carried out. Within the scope of cane pruning, spurs (the base cane) was pruned from 2 nodes and canes (the upper cane) was pruned from 5 nodes. During the bloom period, all clusters on the summer shoots from the spurs were removed from the grapevine.

1. Control (CNT): No girdling treatment was carried out.

Single Repetitive Girdling Treatments

2. 3 mm Girdling Pre–Bloom (PB SR 3mm): 3–7 days pre–bloom (EL–18), the bark+floem layer between the base internode of the canes was removed with 3 mm wide double bladed girdling clippers.

3. 5 mm Girdling Pre–Bloom (PB SR 5mm): 3–7 days pre–bloom (EL–18), the bark+floem layer between the base internode of the canes was removed with 5 mm wide double bladed girdling clippers.

4. 3 mm Girdling Post–Berry Set (PBS SR 3mm): When the berries were 2 mm in size (EL–27), the bark+phloem layer between the base internode of the canes was removed with 3 mm wide double bladed girdling clippers.

5. 5 mm Girdling Post–Berry Set (PBS SR 5mm): When the berries were 2 mm in size (EL–27), the bark+phloem layer between the base internode of the canes was removed with 5 mm wide double bladed girdling clippers.

Double Repetitive Girdling Treatments

6. Double Repetitive 3 mm Girdling Pre–Bloom (PB DR 3mm): 3–7 days pre–bloom (EL–18), the bark+floem layer between the base internode of the canes was removed with 3 mm wide double bladed girdling clippers and the same process was carried out just below this point during the veraison period (EL–35) and double repetitive girdling were realized.

7. Double Repetitive 5 mm Girdling Pre–Bloom (PB DR 5mm): 3–7 days pre–bloom (EL–18), the bark+floem layer between the base internode of the canes was removed with 5 mm wide double bladed girdling clippers and the same process was carried out just below this point during the veraison period (EL–35) and double repetitive girdling were realized.

8. Double Repetitive 3 mm Girdling Post–Berry Set (PBS DR 3mm): When the berries were 2 mm in size (EL–27), the bark+phloem layer between the base internode of the canes was removed with 3 mm wide double bladed girdling clippers and the same process was carried out just below this point during the veraison period (EL–35) and double repetitive girdling were realized.

9. Double Repetitive 5 mm Girdling Post–Berry Set (PBS DR 5mm): When the berries were 2 mm in size (EL–27), the bark+phloem layer between the base internode of the canes was removed with 5 mm wide double bladed girdling clippers and the same process was carried out just below this point during the veraison period (EL–35) and double repetitive girdling were realized.

Within the scope of the summer pruning in the grapevines, the base leaves under the clusters and all the axillary shoots of the summer shoots were removed from the bottom during the thin unripe grape period (3–4 mm). The removal shoot tipping of spurs was carried out 20–25 cm above the second shoot tying wire. The removal shoot tipping of canes was carried out above four internodes the last cluster of the summer shoot in upper node of canes, so that all summer shoots were in the same level. In the spring period, mechanical tillage was carried out the inter–rows of the trial grapevines, and hoeing was realized on the intro–rows with hand hoe.

Average yield (kg grapevine^{-1}), whole cluster and berry characteristics and berry maturity parameters were analyzed in the grapes harvested and brought to ÇOMÜ Horticulture Pomology Laboratory. In article, the most important grape quality characteristics such as cluster width (cm cluster^{-1}), cluster compactness (1–9), cluster weight (g cluster^{-1}), berry width (mm berry^{-1}), berry weight (g berry^{-1}) (OIV, 2009), Hue (Keskin et al., 2017), TSS (%), pH (Cemeroğlu, 2007) and maturity index (TTS% acidity^{-1}) parameters were evaluated.

This research was planned on a total of 81 grapevines according to the split plots trial design, with 3 replications and 3 grapevines in each replication. The data obtained were subjected to analysis of variance in JMP ® Pro 17.0.0 version statistical programme. As a result of the analysis of variance, interaction effects of control, 3 mm and 5 mm girdling treatments, pre–bloom and post–berry set periods and single–double repetitive interaction effects (P*T*R), period main effect (PME), treatment main effect (TME), repetitive main effect (RME), period treatment (P*T), period repetitive (P*R), treatment repetitive (T*R) interactions were created. The data obtained were compared with $LSD_{0.05}$ multiple comparison test and statistical analyses were performed.

3. Results and Discussion

In this research, which aims to determine the effects of girdling treatments of different widths and different phenologic periods on grape quality in Yalova Çekirdeksizi grape variety, average yield, cluster width, cluster compactness, cluster weight, berry width, berry weight, Hue value, TSS, pH and maturity index are given in Table 1, Table 2, Table 3, Table 4, Table 5, Table 6, Table 7, Table 8, Table 9 and Table 10, respectively.

There was no significant difference at the $LSD_{0.05}$ level in the PME, RME, P*T*R, T*R and P*T interactions in terms of average yield value in Yalova Çekirdeksizi grape variety in 2022. In the P*R interaction, PBS DR girdling treatments (4.62 kg grapevine^{-1}) created the highest value and was in the first importance group, followed by PB SR (3.91 kg grapevine^{-1}) in the second importance group, and PBS SR (3.55 kg grapevine^{-1}) and PB DR (3.68 kg grapevine^{-1}) girdling treatments in the third importance group, respectively. In terms of the average yield value of 2023, 3mmME (5.36 kg grapevine^{-1}) had the highest value in the TME and was found more significant than CNTME (3.57 kg grapevine–1) and 5mmME (4.11 kg grapevine–1). There was no significant difference could be detected at the LSD0.05 level in the PME, RME, P*T*R, T*R, P*T and P*R interactions in 2023 and biennial average values. When the biennial average values were analyzed, it was determined that 3mmME (4.95 kg grapevine–1) and 5mmME (4.33 kg grapevine–1) were the highest values in TME, respectively, and were found to be significant compared to CNTME (3.15 kg grapevine–1) (Table 1).

It has been revealed in many scientific researches that girdling treatment increases the yield of the grapevines. Carreño et al. (1998) applied 4 mm girdling treatment during berry set and veraison in Italia grape variety, Şahan and Tangolar (2013) applied 4–5 mm girdling treatment in three different periods (berry set, 2 weeks after berry set, 4 weeks after berry set) in Alphonse Lavallée and Flame Seedless grape varieties, İşçi and Altındişli (2014) applied 3–6 mm girdling treatment on canes during veraison in Alphonse Lavallée grape variety, Crupi et al. (2016) applied 4–5 mm girdling treatment on canes when berries were 3–4 mm in size in Early Red Seedless grape variety, Fawzi et al. (2019) applied 2–3 mm girdling treatment on canes when berries were 2–3 mm in size in Thompson Seedless grape variety, Gündüz et al. (2020) applied 5 mm girdling treatment on canes when berries were 3–4 mm in size in Horoz Karası grape variety, Glisic et al. (2022) applied 2–4 mm girdling treatment post–berry set in Victoria grape variety and Gözcü and Dardeniz (2022) applied 3–5 mm girdling treatment from the base internode of the canes when berries were 4–5 mm in size in Yalova Çekirdeksizi grape variety, they concluded that girdling treatments increased grape yield compared to the control. These results in the literature are in harmony with the research findings we have obtained.

There was no significant difference at LSD0.05 level in RME, P*T*R, T*R, P*T and P*R interactions in terms of cluster width value of Yalova Çekirdeksizi grape variety in 2022. In PME, PBS girdling treatments (9.15 cm cluster–1) were in the first significance group by creating higher values compared to the PB girdling treatments (8.66 cm cluster–1). In 2023, a significant difference was detected only in RME at LSD0.05 level and it was determined that DR girdling treatments (9.06 cm cluster–1) were more significant compared to SR girdling treatments (8.60 cm cluster–1). When the biennial average cluster width values were analyzed, a difference at LSD0.05 level was detected in the P*T*R interaction and TME. In P*T*R interaction, the highest cluster width was obtained from PBS DR 3 mm girdling treatment (9.65 cm cluster–1) and was in the first importance group. This was followed by the PBS SR 5 mm (9.21 cm cluster–1), PB DR 5 mm (9.15 cm cluster–1), PB DR 3 mm (9.12 cm cluster–1) and PBS DR 5 mm girdling treatments (9.12 cm cluster–1) which are in the second importance group, respectively. CNT (8.50 cm cluster–1), PB DR 3 mm (8.65 cm cluster–1), PBS SR 3 mm (8.74 cm cluster–1) and PB SR 5 mm girdling treatments (8.79 cm cluster–1) were in the third importance group and gave the lowest cluster values. In TME, 5mmME (9.07 cm cluster–1) and 3mmMET (9.04 cm cluster–1) have the highest values, respectively, and are found more important than CNTME (8.50 cm cluster–1) (Table 2).

Çoban (2001) girdling treatment in the form of a girdling on the base internode of the canes during thin unripe grape period in the Yuvarlak Çekirdeksiz grape variety, Gündüz et al. (2020) applied 5 mm girdling treatment on canes when berries were 3–4 mm in size in Horoz Karası grape variety and Gözcü and Dardeniz (2022) applied 3–5 mm girdling treatment from the base internode of the canes when berries were 4–5 mm in size in Yalova Çekirdeksizi grape variety, it was revealed that the girdling treatment increased the cluster width and caused a significant difference compared to the control clusters. These results obtained from the literature are similar to the findings of our research.

In the P*T*R interaction, the highest cluster compactness value was obtained from PB SR 3 mm girdling treatment (6.29) and was in the first importance group. PBS SR 3 mm (6.22), PBS SR 5 mm (6.04), PB SR 5 mm (5.83), PBS SR 5 mm (5.81), PBS SR 3 mm (5.72), PB SR 3 mm (5.68) and PB SR 5 mm (5.41) created different intermediate groups. CNT (5.21) was in the last importance group with the lowest cluster compactness value. In the P*R interactions, PBS SR (5.78) and PBS DR girdling treatments (5.75) were in the first importance group with the highest cluster compactness value, followed by PBS SR (5.66) in the second importance group and PBS DR girdling treatment (5.43) in the third importance group.

Table 1. Average yield value of Yalova Çekirdeksizi grape variety (kg grapevine^{-1})

Çizelge 1. *Yalova Çekirdeksizi üzüm çeşidine ait ortalama verim değeri (kg omca^{-1})*

Period	Treatments	SR 2022	DR 2022	SR 2023	DR 2023	SR Average	DR Average	(P*T)* 2022	(P*T)* 2023	(P*T)* Average
PB	CNT (P*T*R)*	2.73	2.73	3.57	3.57	3.15	3.15	2.73	3.57	3.15
	3 mm (P*T*R)	4.73	4.01	5.54	4.95	5.13	4.48	4.37	5.25	4.81
	5 mm (P*T*R)	4.29	4.29	3.73	4.24	4.01	4.26	4.29	3.98	4.14
PBS	CNT (P*T*R)*	2.73	2.73	3.57	3.57	3.15	3.15	2.73	3.57	3.15
	3 mm (P*T*R)	3.77	5.66	4.99	5.94	4.38	5.80	4.72	5.47	5.09
	5 mm (P*T*R)	4.14	5.46	4.78	3.70	4.46	4.58	4.80	4.24	4.52
	$LSD_{0.05}$*	N.S.		N.S.		N.S.		N.S.	N.S.	N.S.
									PME (P)**	
PB (P*R)**		3.91 ab	3.68 b	4.28	4.25	4.10	3.96	3.80	4.27	4.03
PBS (P*R)		3.55 b	4.62 a	4.45	4.40	3.99	4.51	4.08	4.43	4.25
	$LSD_{0.05}$**	0.819		N.S.		N.S.		N.S.	N.S.	N.S.
									TME (T)***	
CNTME (T*R)***		2.73	2.73	3.57	3.57	3.15	3.15	2.73 b	3.57 b	3.15 b
3mmME (T*R)		4.25	4.84	5.26	5.45	4.76	5.14	4.54 a	5.36 a	4.95 a
5mmME (T*R)		4.21	4.87	4.26	3.97	4.24	4.42	4.54 a	4.11 b	4.33 a
	$LSD_{0.05}$***	N.S.		N.S.		N.S.		0.709	0.821	0.652
RME (R)****		3.73	4.15	4.36	4.33	4.05	4.24			
	$LSD_{0.05}$****	N.S.		N.S.		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

Table 2. Cluster width value of Yalova Çekirdeksizi grape variety (cm cluster^{-1})

Çizelge 2. *Yalova Çekirdeksizi üzüm çeşidine ait salkım eni değeri (cm salkım^{-1})*

Period	Treatments	SR 2022	DR 2022	SR 2023	DR 2023	SR Average	DR Average	(P*T)* 2022	(P*T)* 2023	(P*T)* Average
PB	CNT (P*T*R)*	8.25	8.25	8.74	8.74	8.50 b	8.50 b	8.25	8.74	8.50
	3 mm (P*T*R)	9.63	8.51	8.61	8.78	9.12 ab	8.65 b	9.07	8.70	8.88
	5 mm (P*T*R)	8.56	8.73	9.02	9.57	8.79 b	9.15 ab	8.65	9.30	8.97
PBS	CNT (P*T*R)*	8.25	8.25	8.74	8.74	8.50 b	8.50 b	8.25	8.74	8.50
	3 mm (P*T*R)	9.36	9.67	8.12	9.62	8.74 b	9.65 a	9.52	8.87	9.19
	5 mm (P*T*R)	10.02	9.37	8.39	8.87	9.21 ab	9.12 ab	9.70	8.63	9.16
	$LSD_{0.05}$*	N.S.		N.S.		0.758		N.S.	N.S.	N.S.
									PME (P)**	
PB (P*R)**		8.81	8.50	8.79	9.03	8.80	8.77	8.66 b	8.91	8.78
PBS (P*R)		9.21	9.10	8.42	9.08	8.81	9.09	9.15 a	8.75	8.95
	$LSD_{0.05}$**	N.S.		N.S.		N.S.		0.385	N.S.	N.S.
									TME (T)***	
CNTME (T*R)***		8.25	8.25	8.74	8.74	8.50	8.50	8.25 b	8.74	8.50 b
3mmME (T*R)		9.50	9.09	8.36	9.20	8.93	9.15	9.29 a	8.78	9.04 a
5mmME (T*R)		9.29	9.05	8.71	9.22	8.99	9.14	9.17 a	8.96	9.07 a
	$LSD_{0.05}$***	N.S.		N.S.		N.S.		0.472	N.S.	0.379
RME (R)****		9.01	8.80	8.60 b	9.06 a	8.81	8.93			
	$LSD_{0.05}$****	N.S.		0.355		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

Table 3. Cluster compactness value of Yalova Çekirdeksizi grape variety (1–9)
Çizelge 3. *Yalova Çekirdeksizi üzüm çeşidine ait salkım sıklığı değeri (1–9)*

Period	Treatments	SR	DR	SR	DR	SR	DR	(P*T)*		
		2022		2023		Average		2022	2023	Average
PB	CNT (P*T*R)*	5.21 e	5.21 e	5.54	5.54	5.37	5.37	5.21	5.54	5.37
	3 mm (P*T*R)	6.29 a	5.68 cd	5.63	5.63	5.96	5.65	5.99	5.63	5.81
	5 mm (P*T*R)	5.83 bc	5.41 de	5.71	5.71	5.77	5.56	5.62	5.71	5.66
PBS	CNT (P*T*R)*	5.21 e	5.21 e	5.54	5.54	5.37	5.37	5.21	5.54	5.37
	3 mm (P*T*R)	5.72 cd	6.22 ab	5.31	5.64	5.51	5.93	5.97	5.48	5.72
	5 mm (P*T*R)	6.04 abc	5.81 bcd	5.44	5.21	5.74	5.51	5.93	5.32	5.62
	$LSD_{0.05}$*	0.410		N.S.		N.S.		N.S.	N.S.	N.S.
								PME (P)**		
PB (P*R)**		5.78 a	5.43 b	5.62	5.63	5.70	5.53	5.61	5.62	5.61
PBS (P*R)		5.66 ab	5.75 a	5.43	5.46	5.54	5.60	5.70	5.45	5.57
	$LSD_{0.05}$**	0.237		N.S.		N.S.		N.S.	N.S.	N.S.
								TME (T)***		
CNTME (T*R)***		5.21	5.21	5.54	5.54	5.37	5.37	5.21 c	5.54	5.37 b
3mmME (T*R)		6.01	5.95	5.47	5.63	5.74	5.79	5.98 a	5.55	5.76 a
5mmME (T*R)		5.94	5.61	5.57	5.46	5.75	5.53	5.77 b	5.52	5.64 a
	$LSD_{0.05}$***	N.S.		N.S.		N.S.		0.205	N.S.	0.192
RME (R)****		5.72	5.59	5.53	5.54	5.62	5.57			
	$LSD_{0.05}$****	N.S.		N.S.		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

Table 4. Cluster weight value of Yalova Çekirdeksizi grape variety (g $cluster^{-1}$)
Çizelge 4. *Yalova Çekirdeksizi üzüm çeşidine ait salkım ağırlığı değeri (g $salkım^{-1}$)*

Period	Treatments	SR	DR	SR	DR	SR	DR	(P*T)*		
		2022		2023		Average		2022	2023	Average
PB	CNT (P*T*R)*	184.5 e	184.5 e	174.8 cd	174.8 cd	179.7 d	179.6 d	184.5	174.8	179.6
	3 mm (P*T*R)	279.2 ab	197.7 de	232.3 a	200.2 abc	255.7 ab	199.0 cd	238.4	216.3	227.4
	5 mm (P*T*R)	259.2 abc	232.3 cd	171.0 cd	204.1 abc	215.1 c	218.2 c	245.8	187.5	216.6
PBS	CNT (P*T*R)*	184.5 e	184.5 e	174.8 cd	174.8 cd	179.6 d	179.6 d	184.5	174.8	179.6
	3 mm (P*T*R)	249.6 bc	296.9 a	163.3 d	226.2 a	206.5 cd	261.6 a	273.3	194.8	234.0
	5 mm (P*T*R)	260.2 abc	241.3 bcd	189.7 bcd	177.3 cd	225.0 bc	209.3 cd	250.7	183.5	217.1
	$LSD_{0.05}$*	46.55		36.61		34.39		N.S.	N.S.	N.S.
								PME (P)**		
PB (P*R)**		240.9 a	204.8 b	192.7	193.0	216.8	198.9	222.9	192.9	207.9
PBS (P*R)		231.5 ab	240.9 a	175.9	192.8	203.7	216.8	236.2	184.3	210.3
	$LSD_{0.05}$**	26.88		N.S.		N.S.		N.S.	N.S.	N.S.
								TME (T)***		
CNTME (T*R)***		184.5	184.5	174.8	174.8	179.6	179.6	184.7 b	174.8 b	179.6 b
3mmME (T*R)		264.4	247.3	197.8	213.2	231.1	230.3	255.9 a	205.5 a	230.7 a
5mmME (T*R)		259.7	236.8	180.3	190.7	220.0	213.7	248.2 a	185.5 b	216.9 a
	$LSD_{0.05}$***	N.S.		N.S.		N.S.		23.28	18.30	17.22
RME (R)****		236.2	222.9	184.3	192.9	210.3	207.9			
	$LSD_{0.05}$****	N.S.		N.S.		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

A significant difference was determined at $LSD_{0.05}$ level in TME, and 3mmME (5.98) was in the first significance level with the highest cluster compactness value. It was followed by 5mmME (5.77) which was in the second importance level and CNTME (5.21) which was in the last importance group. In 2023, there was no significant difference at $LSD_{0.05}$ level in PME, RME, TME, P*T*R, T*R, P*T and P*R interactions. When the two–year average cluster compactness values were analyzed, a significant difference was determined only in TME at $LSD_{0.05}$ level, and 3mmME (5.76) and 5mmME (5.64) had the highest value and were found more significant than CNTME (5.37) (Table 3).

There was no significant difference at $LSD_{0.05}$ level in PME, RME, T*R and P*T interactions in terms of cluster weight value in Yalova Çekirdeksizi grape variety in 2022. In P*T*R interaction, the highest cluster weight was obtained from PBS DR 3 mm girdling treatment (296.9 g cluster^{-1}) and was in the first importance group. PB SR 3 mm (279.2 g cluster^{-1}), PBS SR 5 mm (260.2 g cluster^{-1}), PB SR 5 mm (259.2 g cluster^{-1}), PBS SR 3 mm (249.6 g cluster^{-1}), PBS DR 5 mm (241.3 g cluster^{-1}), PB DR 5 mm (232.3 g cluster^{-1}) and PB DR 3 mm girdling treatments (197.7 g cluster^{-1}) formed different intermediate groups. CNT (184.5 g cluster^{-1}) produced the lowest cluster weight value and was in the last importance group. In the P*R interactions, PB SR (240.9 g cluster^{-1}) and PBS DR girdling treatments (240.9 g cluster^{-1}) were in the first importance group with the highest cluster weight value, followed by PBS SR (231.5 kg cluster^{-1}) in the second importance group and PB DR girdling treatment (204.8 g cluster^{-1}) in the third importance group. There was a significant difference at $LSD_{0.05}$ level in TME and 3mmME (255.9 g cluster^{-1}) and 5mmME (248.2 g cluster^{-1}) had the highest cluster weight values, respectively, and were found more significant than CNTME (184.7 g cluster^{-1}). In 2023, no significant difference was detected in PME, RME, TME, T*R and P*T interactions at $LSD_{0.05}$ level. When the 2023 cluster width values were analyzed, it was determined that in the P*T*R interactions, PB SR 3 mm (232.3 g cluster^{-1}) and PBS DR 3 mm girdling treatments (226.2 g cluster^{-1}) were in the first importance group, respectively. These treatments were followed by PB DR 5 mm (204.1 g cluster^{-1}), PB DR 3 mm (200.2 g cluster^{-1}), PBS SR 5 mm (189.7 g cluster^{-1}), PBS DR 5 mm (177.3 g cluster^{-1}), CNT (174. 8 g cluster^{-1}) and PB SR 5 mm girdling treatments (171.0 g cluster^{-1}) which were in the different intermediate groups and PBS SR 3 mm girdling treatment (163.3 g cluster^{-1}), which was in the last importance group with the lowest cluster weight. In TME, 3mmME (205.5 g cluster^{-1}) had the highest cluster weight value, which was more significant than 5mmME (185.5 g cluster^{-1}) and CNTME (174.8 g cluster^{-1}), respectively. When two–year average cluster weight values were analyzed, no significant difference was detected at $LSD_{0.05}$ level in PME, RME, TME, T*R and P*T interactions. In the two–year average P*T*R interaction, PBS DR 5 mm girdling treatment (261.6 g cluster^{-1}) produced the highest cluster weight value and was in the first importance group. Different intermediate groups were formed by PB SR 3 mm (255.7 g cluster^{-1}), PBS SR 5 mm (225.0 g cluster^{-1}), PB DR 5 mm (218.2 g cluster^{-1}), PB SR 5 mm (215.1 g cluster^{-1}), PBS DR 5 mm (209.3 g cluster^{-1}), PBS SR 3 mm (206.5 g cluster^{-1}) and PB DR 3 mm (199.0 g cluster^{-1}). CNT (179.6 g cluster^{-1}) was in the last importance group with the lowest cluster weight value. In TME, 3mmME (230.7 g cluster^{-1}) and 5mmME (216.9 g cluster^{-1}) had the highest cluster weight values and were found more significant than CNTME (179.6 g cluster^{-1}), respectively (Table 4).

Çoban (2001) girdling treatment in the form of a girdling on the base internode of the canes during thin unripe grape period in the Yuvarlak Çekirdeksiz grape variety, Ahmad and Zargar (2005) applied 4 mm girdling treatment on trunk in post–berry set in the Perlette grape variety, Şahan and Tangolar (2013) applied 4–5 mm girdling treatment in three different periods (berry set, 2 weeks after berry set, 4 weeks after berry set) in Alphonse Lavallée and Flame Seedless grape varieties, Fawzi et al. (2019) applied 2–3 mm girdling treatment on canes when berries were 2–3 mm in size in Thompson Seedless grape variety, Glisic et al. (2022) applied 2–4 mm girdling treatment post–berry set in Victoria grape variety and Gözcü and Dardeniz (2022) applied 3–5 mm girdling treatment from the base internode of the canes when berries were 4–5 mm in size in Yalova Çekirdeksizi grape variety, and concluded that girdling application had an increasing effect on cluster weight. There is a similarity between these literature results and the findings of our research.

There was no significant difference at $LSD_{0.05}$ level in RME, P*T*R, T*R and P*R interactions in terms of berry width value in Yalova Çekirdeksizi grape variety in 2022, 2023 and two–year average. When 2022–year berry width values were analyzed, P*T interaction PBS 5 mm (17.65 mm berry^{-1}) and PBS 3 mm girdling treatments (17.47 mm berry^{-1}) produced the highest berry width value and was in the first importance group.

Table 5. Berry width value of Yalova Çekirdeksizi grape variety (mm berry^{-1})

Çizelge 5. *Yalova Çekirdeksizi üzüm çeşidine ait tane eni değeri (mm tane^{-1})*

Period	Treatments	SR	DR	SR	DR	SR	DR	(P*T)*		
		2022		2023		Average		2022	2023	Average
PB	CNT (P*T*R)*	15.72	15.72	12.88	12.88	14.30	14.30	15.72 bc	12.88 b	14.30 c
	3 mm (P*T*R)	15.76	14.26	13.58	13.30	14.67	13.78	15.01 c	13.44 b	14.22 c
	5 mm (P*T*R)	16.41	15.86	14.43	14.55	15.42	15.21	16.14 b	14.49 a	15.31 b
PBS	CNT (P*T*R)*	15.72	15.72	12.88	12.88	14.30	14.30	15.72 bc	12.88 b	14.30 c
	3 mm (P*T*R)	17.66	17.27	15.38	14.66	16.52	15.97	17.47 a	15.02 a	16.25 a
	5 mm (P*T*R)	17.44	17.86	15.75	14.65	16.60	16.26	17.65 a	15.20 a	16.43 a
	$LSD_{0.05}$*	N.S.		N.S.		N.S.		0.855	0.735	0.742
								PME (P)**		
PB (P*R)**		15.96	15.28	13.63	13.58	14.80	14.43	15.62 b	13.60 b	14.61 b
PBS (P*R)		16.94	16.95	14.67	14.06	15.81	15.51	16.95 a	14.37 a	15.66 a
	$LSD_{0.05}$**	N.S.		N.S.		N.S.		0.494	0.425	0.429
								TME (T)***		
CNTME (T*R)***		15.72	15.72	12.88	12.88	14.30	14.30	15.72 b	12.88 c	14.30 c
3mmME (T*R)		16.71	15.77	14.48	13.98	15.60	14.87	16.24 b	14.23 b	15.24 b
5mmME (T*R)		16.93	16.86	15.09	14.60	16.01	15.73	16.89 a	14.85 a	15.87 a
	$LSD_{0.05}$***	N.S.		N.S.		N.S.		0.605	0.520	0.525
RME (R)****		16.45	16.12	14.15	13.82	15.30	14.97			
	$LSD_{0.05}$****	N.S.		N.S.		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

Table 6. Berry weight value of Yalova Çekirdeksizi grape variety (g berry^{-1})

Çizelge 6. *Yalova Çekirdeksizi üzüm çeşidine ait tane ağırlığı değeri (g tane^{-1})*

Period	Treatments	SR	DR	SR	DR	SR	DR	(P*T)*		
		2022		2023		Average		2022	2023	Average
PB	CNT (P*T*R)*	2.96	2.96	2.04	2.04	2.50	2.50	2.96 bc	2.04 c	2.50 c
	3 mm (P*T*R)	3.26	2.25	2.28	2.35	2.77	2.30	2.75 c	2.31 b	2.53 c
	5 mm (P*T*R)	3.10	3.54	2.37	2.70	2.73	3.12	3.32 c	2.54 b	2.93 b
PBS	CNT (P*T*R)*	2.96	2.96	2.04	2.04	2.50	2.50	2.96 bc	2.04 c	2.50 c
	3 mm (P*T*R)	4.00	3.99	2.98	3.05	3.49	3.52	3.99 a	3.02 a	3.51 a
	5 mm (P*T*R)	4.10	4.24	2.93	2.72	3.51	3.48	4.17 a	2.82 a	3.50 a
	$LSD_{0.05}$*	N.S.		N.S.		N.S.		0.429	0.255	0.305
								PME (P)**		
PB (P*R)**		3.11	2.92	2.23	2.36	2.67	2.64	3.01 b	2.30 b	2.65 b
PBS (P*R)		3.69	3.73	2.65	2.61	3.17	3.17	3.71 a	2.63 a	3.17 a
	$LSD_{0.05}$**	N.S.		N.S.		N.S.		0.248	0.147	0.176
								TME (T)***		
CNTME (T*R)***		2.96 b	2.96 b	2.04	2.04	2.50	2.50	2.96 c	2.04 b	2.50 b
3mmME (T*R)		3.63 a	3.12 b	2.63	2.70	3.13	2.91	3.38 b	2.67 a	3.02 a
5mmME (T*R)		3.60 a	3.89 a	2.65	2.71	3.12	3.30	3.75 a	2.68 a	3.21 a
	$LSD_{0.05}$***	0.429		N.S.		N.S.		0.304	0.179	0.215
RME (R)****		3.40	3.32	2.44	2.49	2.92	2.91			
	$LSD_{0.05}$****	N.S.		N.S.		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

These applications were followed by PB 5 mm girdling treatment (16.14 mm berry^{-1}) and CNT (15.72 mm berry^{-1}), which were in different intermediate groups, and PB 3 mm girdling treatment (15.01 mm berry^{-1}), which was in the last importance group. In PME, PBS girdling treatments (16.95 mm berry^{-1}) were in the first importance group, creating a higher value than PB girdling treatments (15.62 mm berry^{-1}). In TME, 5mmME (16.89 mm berry^{-1}) had the highest berry width value and was found to be more important than 3mmME (16.24 mm berry^{-1}) and CNTME (15.72 mm berry^{-1}), respectively. When the P*T interaction of the berry width value of 2023 year is examined, PBS 5 mm (15.20 mm berry^{-1}), PBS 3 mm (15.02 mm berry^{-1}) and PB 5mm girdling treatments (14.49 mm berry^{-1}) have the highest berry width value, respectively and was placed in the first importance group. These applications were followed by PB 3 mm girdling treatment (13.44 mm berry^{-1}) and CNT (12.88 mm berry^{-1}), which are in the last importance group. In PME, PBS girdling treatments (14.37 mm berry^{-1}) were in the first importance group, creating a higher value than PB girdling treatments (13.60 mm berry^{-1}). In TME, 5mmME (14.85 mm berry^{-1}) is in the first importance group as having the highest berry width value. This was followed by 3mmME (14.23 mm berry^{-1}) in the second importance group and CNTME (12.88 mm berry^{-1}) in the last importance group. When the D*U interaction of the two–year average berry width value was examined, PBS 5 mm (16.43 mm berry^{-1}) and PBS 3 mm girdling treatments (16.25 mm berry^{-1}) were in the first importance group with the highest berry width value, respectively. These applications were followed by PB 5 mm (15.31 mm berry^{-1}), which is in the second importance group, and CNT (14.30 mm berry^{-1}) and PB 3 mm girdling treatment (14.22 mm berry^{-1}), which are in the last importance group. In PME, PBS girdling treatments (15.66 mm berry^{-1}) were in the first importance group, creating a higher value than PB girdling treatments (14.61 mm berry^{-1}). In TME, 5mmME (15.87 mm berry^{-1}) is in the first importance group as having the highest berry width value. This was followed by 3mmME (15.24 mm berry^{-1}) in the second importance group and CNTME (14.30 mm berry^{-1}) in the last importance group, respectively (Table 5).

Çoban (2001) girdling treatment in the form of a girdling on the base internode of the canes during thin unripe grape period in the Yuvarlak Çekirdeksiz grape variety, Şahan and Tangolar (2013) applied 4–5 mm girdling treatment in three different periods (berry set, 2 weeks after berry set, 4 weeks after berry set) in Alphonse Lavallée and Flame Seedless grape varieties, Fawzi et al. (2019) applied 2–3 mm girdling treatment on canes when berries were 2–3 mm in size in Thompson Seedless grape variety, Gündüz et al. (2020) applied 5 mm girdling treatment on canes when berries were 3–4 mm in size in Horoz Karası grape variety, Söyler et al. (2020) applied 3 mm girdling treatment on trunk when berries 4 mm in size in Mevlana grape variety, Glisic et al. (2022) applied 2–4 mm girdling treatment post–berry set in Victoria grape variety and Gözcü and Dardeniz (2022) applied 3–5 mm girdling treatment from the base internode of the canes when berries were 4–5 mm in size in Yalova Çekirdeksizi grape variety, Çiftçi ve Çelik (2023) applied girdling treatment on trunk at veraison period Samancı Çekirdeksizi and Alphonse Lavallée grape varieties, and concluded that girdling application had an increasing effect on berry width. It seems that these literature results are in harmony with the research findings we have conducted.

There was no significant difference at $LSD_{0.05}$ level in RME, P*T*R and P*R interactions in terms of berry weight value in Yalova Çekirdeksizi grape variety in 2022. In the T*R interaction, DR 5mmME (3.89 g berry^{-1}), SR 3mmME (3.63 g berry^{-1}) and SR 5mmME (3.60 g berry^{-1}) had the highest berry weight value and were found to be more important than DR 3mmME (3.12 g berry^{-1}) and CNTME (2.96 g berry^{-1}), respectively. When the P*T interaction was examined, PBS 5 mm (4.17 g berry^{-1}) and PBS 3 mm girdling treatments (3.99 g berry^{-1}) were in the first importance group with the highest berry weight value, respectively.

These treatments were followed by CNT (2.96 g berry–1), which constitutes the second importance group, and PB 5 mm (3.32 g berry–1) and PB 3 mm girdling treatments (2.75 g berry–1) which are in the last importance group. In PME, PBS girdling treatments (3.71 g berry–1) were in the first importance group, creating a higher value than PB girdling treatments (3.01 g berry–1) (Table 6).

In TME, 5mmME (3.75 g berry^{-1}) was in the first importance group as having the highest berry weight value. This was followed by 3mmME (3.38 g berry^{-1}) in the second importance group and CNTME (2.96 berry^{-1}) in the last importance group. When the P*T interaction of the berry weight value of 2023 was analyzed, PBS 3 mm (3.02 g berry^{-1}) and PBS 5 mm girdling treatments (2.82 g berry^{-1}), respectively, were in the first importance group with the highest berry weight value.

These applications were followed by PB 5 mm (2.54 g $berry^{-1}$) and PB 3 mm girdling treatments (2.31 g $berry^{-1}$), which constitute the second importance group, and CNT (2.04 g $berry^{-1}$), which is in the last importance group. In PME, PBS girdling treatments (2.63 g $berry^{-1}$) were in the first importance group, creating a higher value than PB girdling treatments (2.30 g $berry^{-1}$). In TME, 5mmME (2.68 g $berry^{-1}$) and 3mmME (2.67 g $berry^{-1}$) had the highest berry weight values, respectively, and were found to be more important than CNTME (2.04 g $berry^{-1}$). When the P*T interaction of the two–year average berry weight value was examined, PMS 3 mm (3.51 g $berry^{-1}$) and PBS 5 mm girdling treatments (3.50 g $berry^{-1}$) were in the first importance group with the highest berry weight value, respectively. These treatments were followed by PB 5 mm girdling treatment (2.93 g $berry^{-1}$), which constitutes the second importance group, and PB 3 mm girdling treatment (2.53 g $berry^{-1}$) and CNT (2.50 g $berry^{-1}$), which are in the last importance group. In PME, PBS girdling treatments (3.17 g $berry^{-1}$) were in the first importance group, creating a higher value than PB girdling treatments (2.65 g $berry^{-1}$). In TME, 5mmME (3.21 g $berry^{-1}$) and 3mmME (3.02 g $berry^{-1}$) had the highest berry weight values, respectively, and were found to be more important than CNTME (2.50 g $berry^{-1}$) (Table 6). Carreño et al. (1998) applied 4 mm girdling treatment during berry set and veraison in Italia grape variety, Çoban (2001) girdling treatment in the form of a girdling on the base internode of the canes during thin unripe grape period in the Yuvarlak Çekirdeksiz grape variety, Şahan and Tangolar (2013) applied 4–5 mm girdling treatment in three different periods (berry set, 2 weeks after berry set, 4 weeks after berry set) in Alphonse Lavallée and Flame Seedless grape varieties, Camcı and Çoban (2016) girdling treatment in the form of a girdling on trunk at veraison in the Superior Seedless grape variety, Crupi et al. (2016) applied 4–5 mm girdling treatment on canes when berries were 3–4 mm in size in Early Red Seedless grape variety, Soltekin et al. (2016) applied 4 mm girdling treatments on canes at post–berry set and the beginning of veraison Flame Seedless grape variety, Fawzi et al. (2019) applied 2–3 mm girdling treatment on canes when berries were 2–3 mm in size in Thompson Seedless grape variety, Gündüz et al. (2020) applied 5 mm girdling treatment on canes when berries were 3–4 mm in size in Horoz Karası grape variety, Gözcü and Dardeniz (2022) applied 3–5 mm girdling treatment from the base internode of the canes when berries were 4–5 mm in size in Yalova Çekirdeksizi grape variety, Tóth et al. (2022) applied 4 mm girdling treatment on canes at the beginning of veraison three different table grape varieties, Çiftçi ve Çelik (2023) applied girdling treatment on trunk at veraison period Samancı Çekirdeksizi and Alphonse Lavallée grape varieties, they emphasized that girdling increased the berry weight. These literature results are parallel to the research findings.

There was no significant difference at $LSD_{0.05}$ level in PME, RME, TME, P*T*R, T*R, P*T and P*R interactions in terms of Hue value in Yalova Çekirdeksizi grape variety in 2022. When the Hue value of 2023 was analyzed by TME, 5mmME (108.4) and 3mmME (108.1) were found to be more important than CNTME (107.3) with the highest Hue values, respectively. In the P*R interaction of the two–year average Hue value, PB DR girdling treatment (109.4) constituted the highest value and was in the first importance group, followed by PBS SR (109.0) and PB SR girdling treatments (108.7), which were in the second importance group, PBS DR girdling treatment (108.5), which is in the last importance group, respectively (Table 7).

There was no significant difference at $LSD_{0.05}$ level in PME, P*T*R, T*R, P*T and P*R interactions in terms of TSS value in Yalova Çekirdeksizi grape variety in 2022. It has been determined that the TSS value is more important in terms of RME in SR girdling treatments (17.58%) compared to DR girdling treatments (16.52%). In the TME, CNTME (17.62%) was in the first importance group with the highest TSS value. This was followed by 3mmME (17.22%) in the second importance group and 5mmME (16.31%) in the last importance group. There was no significant difference at $LSD_{0.05}$ level in PME, RME, TME, P*T*R, T*R, P*T and P*R interactions in terms of TSS value in Yalova Çekirdeksizi grape variety in 2023 and two–year average (Table 8).

There was no significant difference at $LSD_{0.05}$ level in PME, P*T*R, P*T and P*R interactions in terms of pH value in Yalova Çekirdeksizi grape variety in 2022. In the T*R interaction of 2022, SR 3mmME (4.11) was in the first importance group with the highest pH value. This was followed by CNTME (4.00), SR 5mmME (3.96) and DR 3mmME (3.92) in the second importance group, and DR 5mmME (3.85) in the last importance group. It has been determined that the pH value is more important in terms of RME in SR girdling treatments (4.02) than in DR girdling treatments (3.93). In the TME, 3mmME (4.02) and CNTME (4.00) had the highest pH values and were found to be more important than 5mmME (3.91).

Table 7. Hue value of Yalova Çekirdeksizi grape variety

Çizelge 7. *Yalova Çekirdeksizi üzüm çeşidine ait Hue değeri*

Period	Treatments	SR	DR	SR	DR	SR	DR	(P*T)*		
		2022		2023		Average		2022	2023	Average
PB	CNT (P*T*R)*	110.1	110.1	107.3	107.3	108.7	108.7	110.1	107.3	108.7
	3 mm (P*T*R)	109.3	111.4	107.1	109.1	108.2	110.2	110.4	108.1	109.2
	5 mm (P*T*R)	110.7	110.0	107.8	108.6	109.3	109.3	110.3	108.2	109.3
PBS	CNT (P*T*R)*	110.1	110.1	107.3	107.3	108.7	108.7	110.1	107.3	108.7
	3 mm (P*T*R)	109.1	108.3	108.2	108.0	108.6	108.2	108.7	108.1	108.4
	5 mm (P*T*R)	110.2	109.2	109.0	108.3	109.6	108.8	109.7	108.6	109.2
	LSD$_{0.05}$*	N.S.		N.S.		N.S.		N.S.	N.S.	N.S.
								PME (P)**		
PB (P*R)**		110.0	110.5	107.4	108.3	108.7 ab	109.4 a	110.3	107.9	109.1
PBS (P*R)		109.8	109.2	108.1	107.9	109.0 ab	108.5 b	109.5	108.0	108.7
	LSD$_{0.05}$**	N.S.		N.S.		0.779		N.S.	N.S.	N.S.
								TME (T)***		
CNTME (T*R)***		110.1	110.1	107.3	107.3	108.7	108.7	110.1	107.3 b	108.7
3mmME (T*R)		109.2	109.8	107.7	108.6	108.4	109.2	109.5	108.1 a	108.8
5mmME (T*R)		110.4	109.6	108.4	108.5	109.4	109.0	110.0	108.4 a	109.2
	LSD$_{0.05}$***	N.S.		N.S.		N.S.		N.S.	0.837	N.S.
RME (R)****		109.9	109.8	107.8	108.1	108.8	109.0			
	LSD$_{0.05}$****	N.S.		N.S.		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

Table 8. TSS value of Yalova Çekirdeksizi grape variety (%)

Çizelge 8. *Yalova Çekirdeksizi üzüm çeşidine ait SÇKM değeri (%)*

Period	Treatments	SR	DR	SR	DR	SR	DR	(P*T)*		
		2022		2023		Average		2022	2023	Average
PB	CNT (P*T*R)*	17.62	17.62	17.32	17.32	17.47	17.47	17.62	17.32	17.47
	3 mm (P*T*R)	19.07	15.06	17.20	16.94	18.13	16.00	17.06	17.07	17.07
	5 mm (P*T*R)	16.48	16.52	18.46	18.65	17.47	17.58	16.50	18.56	17.53
PBS	CNT (P*T*R)*	17.62	17.62	17.32	17.32	17.47	17.47	17.62	17.32	17.47
	3 mm (P*T*R)	17.72	17.05	17.44	17.45	17.58	17.25	17.38	17.45	17.41
	5 mm (P*T*R)	16.97	15.28	17.57	17.28	17.27	16.28	16.12	17.42	16.77
	LSD$_{0.05}$*	N.S.		N.S.		N.S.		N.S.	N.S.	N.S.
								PME (P)**		
PB (P*R)**		17.72	16.40	17.66	17.64	17.69	17.02	17.06	17.65	17.35
PBS (P*R)		17.43	16.65	17.44	17.35	17.44	16.99	17.04	17.39	17.22
	LSD$_{0.05}$**	N.S.		N.S.		N.S.		N.S.	N.S.	N.S.
								TME (T)***		
CNTME (T*R)***		17.62	17.62	17.32	17.32	17.47	17.47	17.62 a	17.32	17.47
3mmME (T*R)		18.39	16.06	17.32	17.20	17.86	16.63	17.22 ab	17.26	17.24
5mmME (T*R)		16.73	15.90	18.01	17.96	17.37	16.93	16.31 b	17.99	17.15
	LSD$_{0.05}$***	N.S.		N.S.		N.S.		1.038	N.S.	N.S.
RME (R)****		17.58 a	16.52 b	17.55	17.49	17.56	17.01			
	LSD$_{0.05}$****	0.847		N.S.		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

Table 9. pH value of Yalova Çekirdeksizi grape variety

Çizelge 9. *Yalova Çekirdeksizi üzüm çeşidine ait pH değeri*

Period	Treatments	SR	DR	SR	DR	SR	DR	(P*T)*		
		2022		2023		Average		2022	2023	Average
PB	CNT (P*T*R)*	4.00	4.00	3.47	3.47	3.74	3.74	4.00	3.47 b	3.74
	3 mm (P*T*R)	4.11	3.83	3.50	3.43	3.80	3.63	3.97	3.46 b	3.72
	5 mm (P*T*R)	3.94	3.84	3.62	3.54	3.78	3.69	3.89	3.58 a	3.74
PBS	CNT (P*T*R)*	4.00	4.00	3.47	3.47	3.74	3.74	4.00	3.47 b	3.74
	3 mm (P*T*R)	4.11	4.02	3.53	3.51	3.82	3.76	4.06	3.52 ab	3.79
	5 mm (P*T*R)	3.99	3.86	3.48	3.55	3.74	3.71	3.92	3.51 ab	3.72
	$LSD_{0.05}$*	N.S.		N.S.		N.S.		N.S.	0.065	N.S.
									PME (P)**	
PB (P*R)**		4.02	3.89	3.53	3.48	3.77	3.68	3.95	3.50	3.73
PBS (P*R)		4.03	3.96	3.49	3.51	3.76	3.74	3.99	3.50	3.75
	$LSD_{0.05}$**	N.S.		N.S.		N.S.		N.S.	N.S.	N.S.
									TME (T)***	
CNTME (T*R)***		4.00 b	4.00 b	3.47	3.47	3.74 bc	3.74 bc	4.00 a	3.47 b	3.74
3mmME (T*R)		4.11 a	3.92 bc	3.52	3.47	3.81 a	3.69 c	4.02 a	3.49 b	3.75
5mmME (T*R)		3.96 b	3.85 c	3.55	3.54	3.76 ab	3.70 bc	3.91 b	3.55 a	3.73
	$LSD_{0.05}$***	0.084		N.S.		0.059		0.059	0.046	N.S.
RME (R)****		4.02 a	3.93 b	3.51	3.49	3.77 a	3.71 b			
	$LSD_{0.05}$****	0.048		N.S.		0.034				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference.

Table 10. Maturity index value of Yalova Çekirdeksizi grape variety (TSS% acidity%$^{-1}$)

Çizelge 10. *Yalova Çekirdeksizi üzüm çeşidine ait olgunluk indisi değeri (%SÇKM %asitlik^{-1})*

Period	Treatments	SR	DR	SR	DR	SR	DR	(P*T)*		
		2022		2023		Average		2022	2023	Average
PB	CNT (P*T*R)*	35.55	35.55	35.24	35.24	35.40	35.40	35.55	35.24 b	35.40
	3 mm (P*T*R)	40.31	26.15	38.55	36.67	39.43	31.41	33.23	37.61 b	35.42
	5 mm (P*T*R)	29.52	29.61	52.09	43.51	40.80	36.56	29.56	47.80 a	38.68
PBS	CNT (P*T*R)*	35.55	35.55	35.24	35.24	35.40	35.40	35.55	35.24 b	35.40
	3 mm (P*T*R)	38.24	37.60	35.43	36.85	36.83	37.22	37.92	36.14 b	37.03
	5 mm (P*T*R)	31.61	25.20	40.46	36.76	36.03	30.98	28.41	38.61 b	33.51
	$LSD_{0.05}$*	N.S.		N.S.		N.S.		N.S.	4.269	N.S.
									PME (P)**	
PB (P*R)**		35.13	30.44	41.96	38.47	38.54	34.46	32.78	40.22 a	36.50
PBS (P*R)		35.14	32.78	37.04	36.28	36.09	34.53	33.96	36.66 b	35.31
	$LSD_{0.05}$**	N.S.		N.S.		N.S.		N.S.	2.464	N.S.
									TME (T)***	
CNTME (T*R)***		35.55	35.55	35.24	35.24	35.40	35.40	35.55	35.24 b	35.40
3mmME (T*R)		39.28	31.88	36.99	36.76	38.13	34.32	35.58	36.87 a	36.23
5mmME (T*R)		30.56	27.40	46.27	40.14	38.42	33.77	28.98	43.20 a	36.09
	$LSD_{0.05}$***	N.S.		N.S.		N.S.		N.S.	2.463	N.S.
RME (R)****		35.13	31.61	39.50	37.38	37.32	34.49			
	$LSD_{0.05}$****	N.S.		N.S.		N.S.				

PB: Pre–Bloom, PBS: Post–Berry Set, CNT: Control, CNTME: Control Main Effect, 3mmME: 3 mm Main Effect, 5mmME: 5 mm Main Effect, RME: Repetitive Main Effect, TME: Treatment Main Effect, PME: Period Main Effect, P: Period, T: Treatment, R: Repetitive, SR: Single Repetitive, DR: Double Repetitive, LSD: Least Significant Difference

When the P*T interaction of the pH value of 2023 was examined, PB 5 mm girdling treatment (3.58) was in the first importance group with the highest pH value. This was followed by PBS 3 mm (3.52) and PBS 5 mm girdling treatments (3.51), which constitute the second importance group, and CNT (3.47) and PB 3 mm girdling treatments (3.46), which are in the last importance group. In TME, 5mmME (3.55) had the highest pH value and was found to be more important than 3mmME (3.49) and CNTME (3.47). In the T*R interaction of the two–year average pH value, SR 3mmME (3.81) was in the first importance group with the highest pH value. This was followed by SR 5mmME (3.76), CNTME (3.74) and DR 5mmME (3.70) in the second importance group, and DR 3mmME (3.69) in the last importance group. It has been determined that SR girdling treatments (3.77) are more important in terms of RME compared to DR girdling treatments (3.71) (Table 9).

There was no significant difference at $LSD_{0.05}$ level in PME, RME, TME, P*T*R, T*R, P*T and P*R interactions in terms of maturity index value in Yalova Çekirdeksizi grape variety in 2022 and two–year average. When the D*U interaction of 2023 was analyzed, PB 5 mm (47.80) was in the first importance group with the highest maturity index value. Respectively, CNT (35.24), PBS 3 mm (36.14), PB 3 mm (37.61) and PBS 5 mm girdling treatments (38.61) were found in the last importance group, creating the lowest maturity index value. In PME, PB girdling treatments (40.22) were in the first importance group, creating a higher value than PBS girdling treatments (36.66). In TME, 5mmME (43.20) and 3mmME (36.87) had the highest maturity index values, respectively, and were found to be more important than CNTME (35.24) (Table 10).

Carreño et al. (1998) applied 4 mm girdling treatment during berry set and veraison in Italia grape variety, Fawzi et al. (2019) applied 2–3 mm girdling treatment on canes when berries were 2–3 mm in size in Thompson Seedless grape variety, Gündüz et al. (2020) applied 5 mm girdling treatment on canes when berries were 3–4 mm in size in Horoz Karası grape variety, it is emphasized that girdling has a positive effect on the maturity index. The results of this literature and the findings of the research conducted in 2023 are similar.

5. Conclusion

In this research, in which the effects of girdling treatments at different periods and widths on grape quality in Yalova Çekirdeksizi (*V. vinifera* L.) grape variety were investigated, when the two–year average findings were evaluated; it was determined that 3mmME and 5mmME produced higher values in TME compared to CNTME in terms of average yield, cluster width, cluster compactness, cluster weight and berry weight values. According to the P*T*R interactions, it was concluded that the PBS DR 3 mm girdling treatment had the highest values in cluster width and cluster weight values compared to the other treatments. According to the P*T interaction, it was determined that PBS 3 mm and PBS 5 mm girdling treatments increased berry width and berry weight compared to other periods and treatments. When PME was analyzed, significant increases were determined in berry width and berry weight values obtained from PBS girdling treatments.

In this research, when the two–year average findings were evaluated; the highest berry width value in TME was realized by 5mmME. When the maturity index value was analyzed, a significant difference was detected only in PME, TME and P*T interaction in 2023, but no significant difference was detected in the two–year average findings.

As a result; in Yalova Çekirdeksizi grape variety grown under arid conditions and standard summer pruning was performed, it was determined that 3 mm and 5 mm girdling treatments produced the same average yield value in 2022, while 3 mm girdling treatments continued to increase the average yield value in 2023, while 5 mm girdling treatments slightly decreased the average yield value. The decrease in yield in the 5 mm girdling treatments may be due to the removal of a wider bark+phloem layer, resulting in a later closure of the wound tissues compared to the 3 mm girdling treatments, and thus a decrease in assimilate products to the main root and old parts. Therefore, it was concluded that although 5 mm girdling treatments continue to give positive results in terms of grape quality in many parameters, it is not appropriate to repeat them two years in a row in terms of average yield.

Note: This article was compiled from a part of Esra Şahin's PhD thesis titled 'The Effects of Canopy Management and Girdling Applications on Grape Quality and Biochemical Properties of Table Grape Varieties (*Vitis vinifera* L.) at Different Periods'.

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Conflicts of Interest

The authors declare no conflict of interest.

Contribution Rate Statement Summary

The authors declare that they have contributed equally to the article.

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Diversity of Drosophilidae (Diptera) in Strawberry Orchards:Discovery of some new species in Konya, Türkiye

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Abstract: Drosophilidae, a diverse and widespread fly family, has recently received increasing attention due to the proliferation of its agricultural pest species, especially in fruit-growing regions. This study aimed to assess the diversity of Drosophilidae in strawberry fields within the Konya province from April to December 2022. A total of 10 species belonging to 4 genera (*Drosophila* Fallén, *Hirtodrosophila* Duda, *Zaprionus* Coquillett, *Phortica* Schiner) were identified in the Drosophilidae family. This study reported the presence of *Drosophila busckii* Coquillett, *Drosophila hydei* (Sturtevant), *Drosophila obscura* (Fallén), *Drosophila subobscura* (Collin), *Drosophila simulans* (Sturtevant), *Drosophila suzukii* (Matsumura), *Hirtodrosophila cameraria* (Haliday), *Phortica variegata* (Fallén) and *Zaprionus indianus* (Gupta) in Konya (Türkiye) for the first time. Additionally, *Drosophila obscura, Drosophila simulans, Hirtodrosophila cameraria* and *Phortica variegata* were determined for the first time in Türkiye's strawberry fields.

Keywords: *Drosophila*, Drosophilidae, Strawberry, Vinegar fly, *Zaprionus*

Çilek Bahçelerinde Drosophilidae Çeşitliliği: Konya'da (Türkiye) Bazı Yeni Türlerin Keşfi

Öz: Drosophilidae, çok çeşitli ve yaygın bir sinek familyasıdır. Özellikle meyve yetiştiriciliği yapılan bölgelerde tarımsal zararlı türlerinin çoğalması nedeniyle son zamanlarda artan bir ilgi görmüştür. Bu çalışmada, Nisan-Aralık 2022 tarihleri arasında Konya ili çilek tarlalarında bulunan Drosophilidae tür çeşitliliğininin belirlenmesi amaçlanmıştır. Drosophilidae familyasında 4 cinse (*Drosophila* Fallén, *Hirtodrosophila* Duda, *Zaprionus* Coquillett, *Phortica* Schiner) ait toplam 10 tür tespit edilmiştir. Bu çalışma, *Drosophila busckii* Coquillett, *Drosophila hydei* (Sturtevant), *Drosophila obscura* (Fallén), *Drosophila subobscura* (Collin), Drosophila simulans (Sturtevant), *Drosophila suzukii* (Matsumura), *Hirtodrosophila cameraria* (Haliday), *Phortica variegata* (Fallén) ve *Zaprionus indianus* (Gupta) türlerinin Konya'da (Türkiye) ilk kez tespit edildiğini rapor etmektedir. Ayrıca *Drosophila obscura*, *Drosophila simulans, Hirtodrosophila cameraria* ve *Phortica variegata* Türkiye'deki çilek tarlalarında ilk kez belirlenmiştir.

Anahtar kelimeler: *Drosophila*, Drosophilidae, Çilek, Sirke sineği, *Zaprionus*

1. Introduction

Strawberry, which can be grown economically in different climate and soil conditions, has become increasingly important in the world and in Türkiye. China, Mexico, the United States of America (USA), Spain and Türkiye are the countries with the highest strawberry production (Simpson, 2018). In recent years, Konya has emerged as the fourth-largest strawberry producer in Türkiye, owing to the surge in strawberry cultivation (Anonymous, 2022). The long strawberry production season and the fact that it can be grown at different altitudes provide different environments for pests to cause problems. One of these problems is *Drosophila* species (Drosophilidae: Diptera), which have spread in many countries in recent years and threaten fruit production in places where they are infected. Most *Drosophila* lay eggs in decaying organic matter materials such as compost, rotting produce; others grow in living or decaying fungi, in the slime or sap of trees or flowering plants (Bächli et al., 2004; O'Grady & Markow, 2009). Some species, such as *Drosophila suzukii* (Matsumura), also lay eggs in healthy unripe fruit (Fartyal et al., 2014). Due to the high reproductive potential of *Drosophila* species, their populations can increase rapidly. The Drosophilidae family, commonly known as vinegar or small fruit flies, comprises 73 extant genera and 3 extinct genera, over 3950 species Drosophilidae. It has mainly two subfamilies Drosophilinae (~3500 species) and Steganinae (~700 species) (Brake & Baechli, 2008; NCBI, 2023). While numerous species within the Drosophilinae subfamily (43 genera), the Steganinae subfamily (30 genera) still lacks comprehensive understanding. The genus *Drosophila* contains more than half of the family, with about 2000 species (O'Grady & DeSalle, 2018). The last published species catalog of the *Drosophila* family in Türkiye lists 36 species (Koçak & Kemal, 2013). However, the online TaxoDros database (http://www.taxodros.uzh.ch/) currently documents 52 species of Drosophilidae (Taxodros, 2023). There are probably more undescribed species waiting to be discovered in Türkiye. The majority of *Drosophila* species have been ignored because they are known to be saprophytic, but in recent years, studies have accelerated in Türkiye with the identification of invasive species that can cause damage to fruits (Başpınar et al., 2022; Çatal et al., 2021; Efil, 2018; Ögür et al., 2018; Zengin, 2020). There is a record of the existence of *Drosophila melanogaster* (Beyşehir district) (Özsoy, 2007) and *Drosophila pallida* (Cihanbeyli and Sille district) (Máca, 1999) species in Konya. Also, the presence of *D. suzukii* in Karaman, Central Anatolia has been reported (Ögür et al., 2018). However, no studies have been conducted in Konya other than these records.

Relying on morphological characteristics for taxonomic identification proves to be an efficient method for recognizing numerous drosophilid species. (Yuzuki & Tidon, 2020). However, accessible identification tools for non-experts remain scarce in this field. We provide photographic descriptions of 10 species recorded in Türkiye to make up for this deficiency.

Since information on Drosophilidae in Konya (Turkey) is very limited, this study aimed to investigate their presence and diversity. Through comprehensive morphological characterization and detailed photography, ten drosophilid species were identified and introduced. Notably, nine of these species represent new records for Konya, Türkiye. Additionally, due to the identification of discrepancies in species names and reference errors in the TaxoDros online database, a revised checklist of *Drosophila* species in Türkiye is presented.

2. Material and Methods

The study was conducted between April and November 2022 in Derbent (38° 06' 88" S, 32˚57′55″ W), Konya, Türkiye. *Drosophila* adults were collected using apple cider vinegar traps from the strawberry fruit samples. A trap was fashioned using a sturdy 500 ml plastic bottle, baited with 100 ml of apple cider vinegar. Eight holes with a diameter of 5 mm were drilled in the top of the bottle to facilitate the entry of the attracted flies. The traps were suspended on garden stakes positioned at an angle near the edges of elevated strawberry beds, with three traps allocated per strawberry garden. They were positioned in a manner where the trap's bottom hung slightly below the top of the strawberry leaves (Renkema et al., 2018). The traps were changed regularly weekly. The collected *Drosophila* species were identified by us based on the morphological criteria described by Bächli et al. (2004), Markow & O'Grady (2006) and Miller et al. (2017) and are deposited in the Department of Plant Protection, Faculty of Agriculture, Selçuk University, Konya, Türkiye.

3. Results and Discussion

1. *Drosophila busckii* Coquillett, 1901

The *busckii* species typically display longitudinally striped pleurae and lack preapical setae on the second and third tibiae (Markow & O'Grady, 2006). The scutum appears yellowish with three distinct dark stripes, the median of which forks in its posterior half (Figure 3). Additionally, the pleura is yellowish and bears two prominent dark horizontal markings (Figure 3). The eyes are round and broader than long (Figure 2). The abdominal tergites are yellowish, each adorned with approximately four more or less isolated dark spots (Figure 1). These are slender flies belonging to the subgenus *Dorsilopha* (Bächli et al., 2004). The *busckii* has transparent wings with no markings (Figure 4) (Miller et al., 2017).

Kocatepe (2019) reported that *D. busckii* was detected in Strawberry orchards in Marmaris (western Türkiye). The same author reported that *D.suzukii* damages and prefers fresh fruits, while *D. busckii* damages rotten fruits.

3.1. *Drosophila hydei* Sturtevant, 1921

This particular species might be sizable and dark but deviates from the previously described characteristics (Figure 5). The mesonotum lacks a bluish hue in its ground color. The frons shows a distinct V-shaped pattern of setae, differing from the previous descriptions. Additionally, the coxae of the forelegs share a similar color with the rest of the leg. The lateral areas of abdominal segments are almost entirely covered by extensions of apical bands, displaying a uniform color without interruptions or lighter regions (Figure 5). The arista features three branches above, excluding the terminal fork (Figure 7). Wing crossveins in this species lack clouding (Figure 8). The mesonotum appears grayish and distinctly spotted, while the second oral vein is either absent or approximately half the length of the first (Markow & O'Grady, 2006). *Drosophila hydei* can be further identified by the presence of prominent fine setae on the inner side of the fore tarsus (Figure 6). Additionally, the apex of the first costal wing section is typically pale in this species (Bächli et al., 2004).

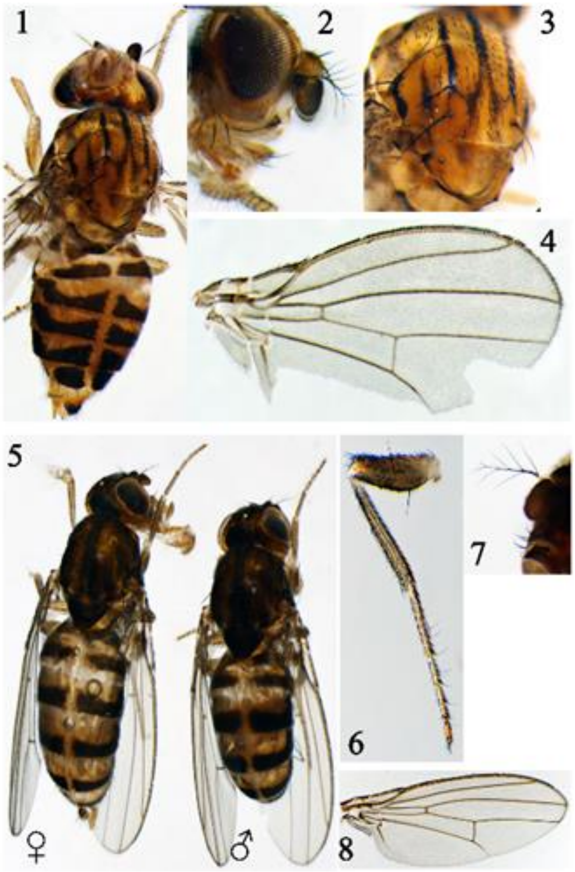

Figure 1-8. Characters of *Drosophila busckii* 1–4: (1) female body, (2) female eyes, (3) female thorax, (4) female wing. Characters of *Drosophila hydei* 5-8: (5) female and male body, (6) male foreleg, (7) male arista, (8) male wing

Şekil 1-8. *Drosophila busckii' nin karakterleri 1–4: (1) dişinin vücudu, (2) dişide gözler, (3) dişide toraks, (4) dişide kanat. Drosophila hydei' nin karakterleri 5–8: (5) dişi ve erkek vücudu, (6) erkekte ön bacak, (7) erkek arista, (8) erkekte kanat.*

3.2. *Drosophila obscura* Fallén, 1823

The *obscura* is dark-colored flies (Figure 9). The acrostichal setulae are organized in eight rows, and sex combs are evident on the first and second tarsal segments (Figure 10-12). The basitarsus notably exceeds the length of the second tarsal segment, and the sex combs themselves are comparatively shorter (Figure 12). The apical of the aedeagus is slender, roundish and the paraphyses are broadened (Figure 11) (Markow & O'Grady, 2006). The outer paraphysis is broad and does not taper towards the tip; instead, it has a distinctly blunt tip (Figure 11) (Bächli et al., 2004).

3.3. *Drosophila subobscura* Collin, 1936

Acrostichal setulae are arranged in eight rows; the proximal sex comb typically has more than ten teeth, while the distal sex comb varies between nine to thirteen teeth. Moreover, the first tarsomere equals the length of the second tarsal segment (Figure 14-15). Both the pleura and abdomen exhibit an overall dark coloration (Figure 13), and the wings display a faint darkening along the costal fringe (Figure 18).

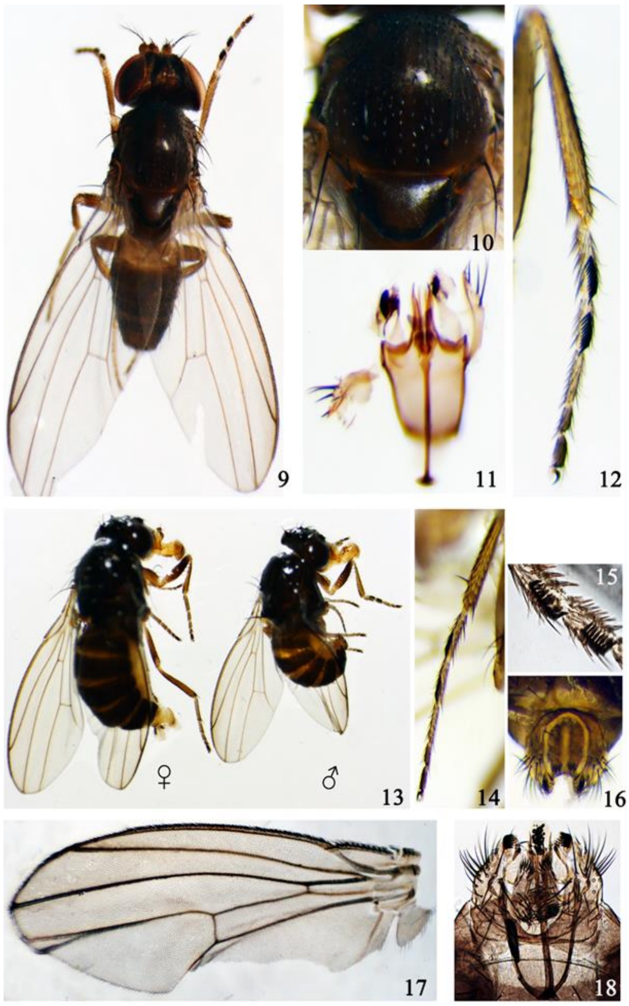

Figure 9–18. Characters of *Drosophila obscura* 9–12: (9) male body, (10) thorax, (11) aedeagus, (12) foreleg. Characters of *Drosophila subobscura* 13–18: (13) female and male body, (14) male foreleg, (15) male sex comb, (16) male cercus, (17) male wing, (18) male ventral view of cercus

Şekil 9-18. *Drosophila obscura' nın karakterleri 9–12: (9) erkek vücudu, (10) toraks, (11) erkek genital organ, (12) ön bacak. Drosophila subobscura' nın karakterleri 13–18: (13) dişi ve erkek vücudu, (14) erkek ön bacak, (15) erkek seks tarağı, (16) erkekte sersi, (17) erkekte kanat, (18) erkekte sersinin alttan görünümü.*

On the ventral margin of the cercus, there's a cluster of short, dense setulae (Figure 16-17). The external process of the epandrium is rounded and bulging at the base, extending into a thin projection. Additionally, the surstylus is substantial and cup-shaped, laterally compressed, housing a very short, square-like comb containing six to eight setae (Markow & O'Grady, 2006).

3.4. *Drosophila melanogaster* Meigen, 1830

The *melanogaster*'s sex combs are exclusively located on the basitarsus (Figure 20). There are no hooked setae on the mid-leg of males, and only primary claspers are observable. The aedeagus exhibits lateral expansions, the epandrium has a posterior expansion, and the anal plates lack both ventral processes and teeth (Figure 22). The epandrial expansion is broader than it is long. The expansion on the epandrium is trapezoidal in shape (Markow & O'Grady, 2006). In both males and females, the gena is relatively broad, approximately 1/10 of the diameter of the large eye (Figure 19). In male specimens, the dorsal branch of the ventral epandrial lobe appears small, nearly triangular in shape, and exhibits a pale coloration when observed laterally (Bächli et al., 2004). The arista first ventral branch has small ray but excluding terminal fork (Figure 21).

Kocatepe (2019) reported that *D. melanogaster* was detected in Strawberry orchards in Marmaris (western Türkiye).

3.5. *Drosophila simulans* Sturtevant, 1919

The *simulans'* sex combs are found only on the basitarsus (Figure 24). On the mid-leg of the male, there are no hooked setae. Solely primary claspers are observable in this region. The aedeagus displays lateral expansions, the epandrium exhibits a posterior expansion, and the anal plates lack both ventral processes and teeth (Figure 26 and 27). The epandrial expansion is wider than long. The expansion on the epandrium is substantial, semicircular, and distinctly visible from a lateral perspective (Markow & O'Grady, 2006). The gena appears relatively narrow, roughly 1/20 the diameter of the large eye (Figure 23). In males, the dorsal branch of the ventral epandrial lobe is notably large, rounded in shape, and presents an amber hue when observed laterally (Bächli et al., 2004). The arista first ventral branch has not small ray but has terminal fork (Figure 25).

3.6. *Drosophila suzukii* (Matsumura, 1931)

In *suzukii* males, tergites 2-4 are pale yellow with narrow, unbroken dark posterior bands, while tergites 5 and 6 are entirely darkened (Figure 28). Females have pale yellow tergites with narrow, unbroken dark

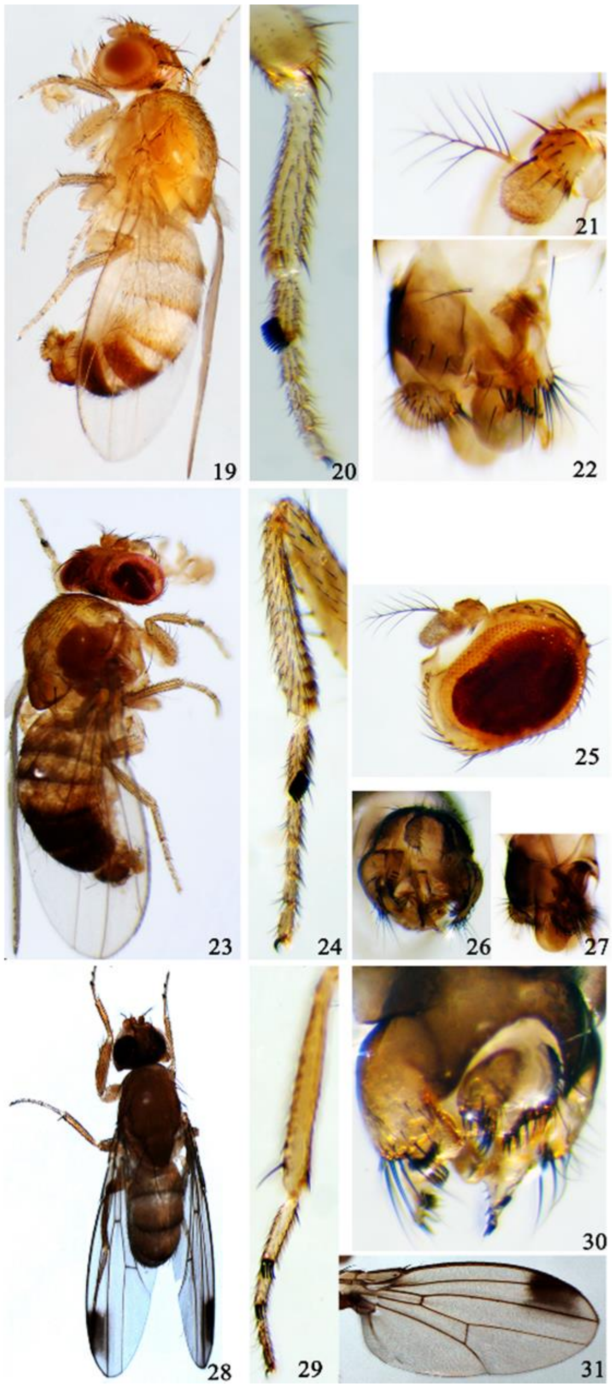

Figure 19–31. Characters of *Drosophila melanogaster* (19–22): (19) male body, (20) male fore tarsus, (21) male arista, (22) male cerci. Characters of *Drosophila simulans* (23–27): (23) male body, (24) male fore tarsus, (25) male arista, (26–27) male cerci. Characters of *Drosophila suzukii* (28–31): (28) male body, (29) male fore tarsus, (30) male cerci, (31) male wing.

Şekil 19-31. *Drosophila melanogaster' in karakterleri (19–22): (19) erkek vücudu, (20) erkekte ön tarsus, (21) erkekte arista, (22) erkekte sersi.Drosophila simulans'ın karakterleri (23–27): (23) erkek vücudu, (24) erkek ön tarsus, (25) erkek arista, (26–27) erkek cercus.Drosophila suzukii'nin karakterleri (28–31): (28) erkek vücut, (29) erkekte tarsus, (30) erkekte sersi, (31) erkekte kanat.*

posterior bands. Males do not possess a dorsal branch of the epandrial ventral lobe (Figure 30). The male fore tarsus features a comb on the first and second fore tarsomeres, with the comb on the first fore tarsomere composed of 4-6 teeth, and the comb on the second fore tarsomere composed of 2-3 teeth (Figure 29). Male wings commonly display infuscation at the apices of wing veins R_{4+5} and R_{2+3}, though sometimes this feature may be absent (Figure 31) (Miller et al., 2017).

Drosophila suzukii was detected for the first time in our country in strawberry fruit in Erzurum (eastern Türkiye) in 2014 (Orhan et al. 2016). Then, in 2019, it was reported that it was detected in Marmaris (western Türkiye) and was harmful throughout the entire fruit season from fruit formation to the end of harvest (Kocatepe, 2019).

3.7. *Hirtodrosophila cameraria* (Haliday, 1833)

The description you provided from Bächli et al. (2004) regarding the diagnosis of *H. cameraria* species (Figure 32) is as follows: The mesonotum showcases a wide, diffuse dark brown median stripe (Figure 33).

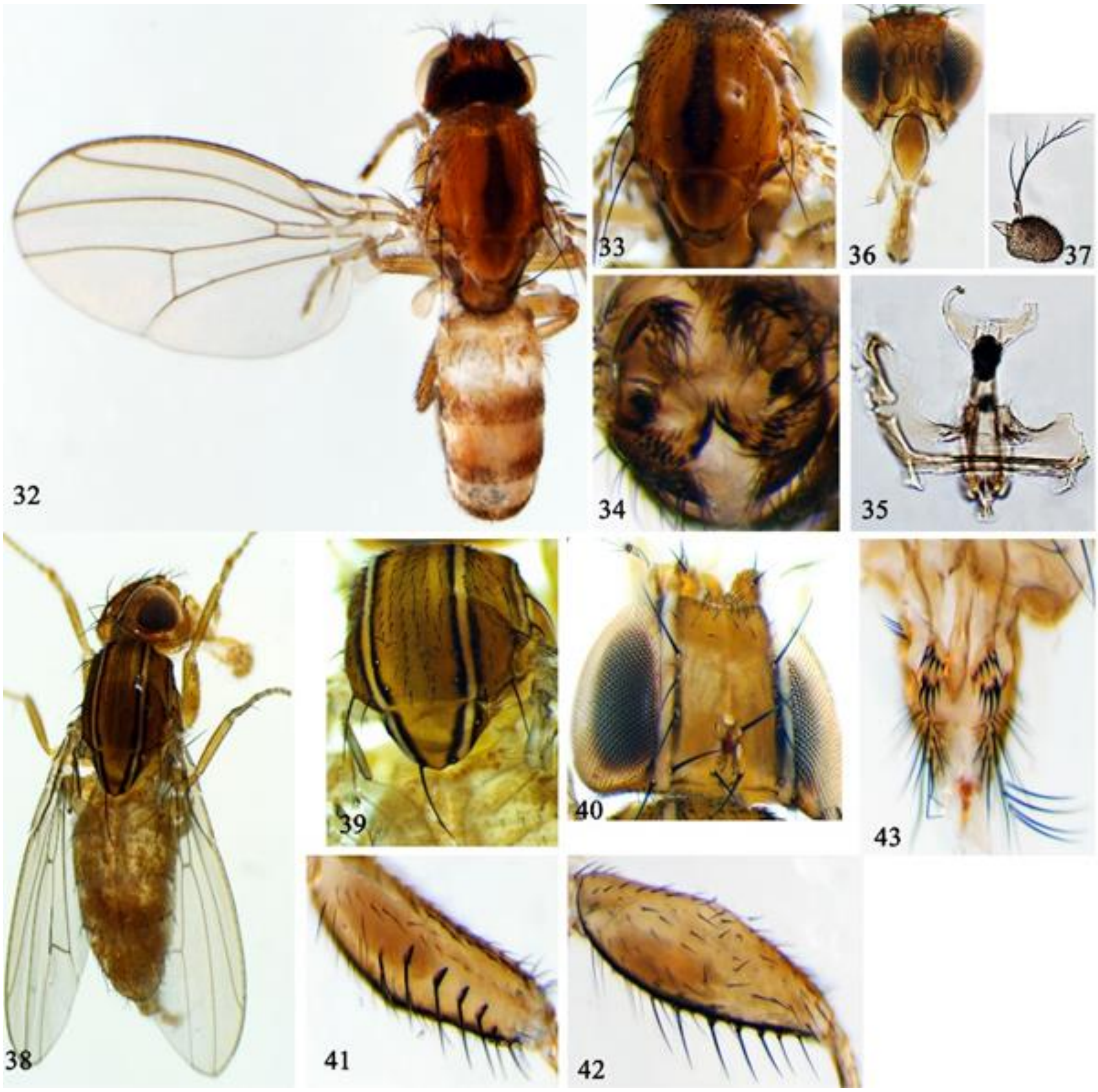

Figure 32–43. Characters of *Hirtodrosophila cameraria* (32–35): (32) male body, (33) male metanotum, (34) male cerci, (35) male aedeagus, (36) male carina, (37) male arista. Characters of *Zaprionus indianus* (38–43): (38) male body, (39) male metanotum, (40) male head, (41–42) male fore femur, (43) male cerci.

Şekil 32-43. *Hirtodrosophila cameraria'nın karakterleri (32–37): (32) erkek vücudu, (33) erkekte metanotum, (34) erkekte sersi, (35) erkek genital organ, (36) erkekte carina, (37) erkekte arista. Zaprionus indianus'un karakterleri (38–43): (38) erkek vücudu, (39) erkekte metanotum, (40) erkekte baş, (41–42) erkekte ön femur, (43) erkekte sersi.*

The cercus displays lateral ventral expansion and features a row of approximately 15 peg-like setae, more prominent at the inner corner (Figure 34). The aedeagus bifurcates apically into two horn-shaped branches, long, slender, scaly, curving dorsally in lateral view and inward in posterior view (Figure 35). The carina appears narrow and relatively small (Figure 36). Antennae exhibit a yellowish hue, with the first flagellomere displaying a diffuse brownish margin. Notably, it features a short lower branch positioned just behind the terminal fork, approximately five relatively long inner branches, and a small terminal fork (Figure 37).

3.8.*Zaprionus indianus* Gupta, 1970

Commonly known as the 'African fig fly,' it belongs to the armatus group in the *Zaprionus* genus and is originally from the African tropics (Gupta, 1970). The fore femur is characterized by a row of approximately five strong setae, each of which is positioned adjacent to and divergent with a stiff setula (Figure 41 and 42). These setae and setula arise from a small tubercle (Bächli et al., 2004). There's a faint, almost transparent dark stripe noticeable along the apical margin of tergites 2-5. Additionally, the sub-apical setae on tergites 4-5 emerge from a dark spot (Figure 39) (Castrezana, 2007). The aedeagus flap is exceptionally smooth at the tip, and the oviscape features six peg-like ovi sensilla (Figure 43) (Yassin & David, 2010).

3.9. *Phortica variegata* (Fallén, 1823)

The homotypic synonym of *Phortica variegata* species is *Amiota variegata*, identified as *Drosophila variegata* according to NCBI (2023). Each antenna arista exhibited between three to six short dorsal branches, gradually decreasing in length towards the tip (Figure 46). The eyes were red in color, encircled by a pale white ring (Figure 47). The scutum showcased a pattern of merging dark spots (Figure 48). Additionally, there was an extra cross-vein separating the discal and second basal wing cells (arrow), a distinctive trait of Drosophilidae, along with two interruptions of the costal vein. The abdomen displayed a yellow and brown design, featuring three transversal bands and one longitudinal dark band against a lighter background (Figure 44–45). Meanwhile, the legs presented three distinct dark bands encircling the tibia. The coxae and femur appeared dark in color, with the latter displaying a lighter base and apex. The female final tergite is conical with an epiproct covered in several short hairs, and the cerci, which are non-sclerotized and hairy, converge at the base (Figure 49) (Otranto et al., 2006).

Although there are studies conducted in fruit orchards such as cherry and peach in Türkiye, research related to strawberries is extremely limited. *Drosophila suzukii* was first determined on strawberry orchards in Erzurum (Türkiye) by Orhan et al. (2016). Kocatepe (2019) were determined *Drosophila suzukii, Drosophila melanogaster* and *Drosophila busckii* species in the strawberry orchards of Marmaris (Muğla-Türkiye). It was determined that among these species, *D. suzukii* was harmful throughout the entire fruit season, from the formation of the fruits until the end of the harvest. The other species, namely *D. melanogaster* and *D. busckii*, were found to be pests in ripened and rotting fruits damaged by *D. suzukii*. The predominant focus on *D. suzukii* among investigations into Drosophilidae species in strawberry fruit can be attributed to its classification as an agricultural pest (Goodhue et al., 2011; Dean et al., 2013; Gong et al., 2016; Bernardi et al., 2017; Efil, 2018; Živković et al.,2019; Baena et al., 2022). To further elucidate the Drosophilidae fauna associated with strawberries, the present study undertook a comprehensive examination of Drosophilidae species in strawberry fruit.

The gena, located on the lower margin of the eye or cheeks, and its width in proportion to the eye, is at times used to distinguish closely related species like *Drosophila melanogaster* and *D. simulans*. In *D. simulans*, the expansion on the epandrium is notably large, semicircular, and clearly visible from a lateral perspective, while in *D. melanogaster*, it tends to assume a trapezoidal shape. (Markow & O'Grady, 2006). While *D. melanogaster* has been previously reported in Konya (Özsoy, 2007), this study provides the first record of the other nine Drosophilidae species identified Konya.

Drosophila obscura can be distinguished from *D. bifasciata* Pomini by the characteristics you mentioned. In *D. obscura*, the outer paraphysis is broad and has a blunt tip, and the aedeagus has a roundish tip. These differences in the outer paraphysis and aedeagus tip are key distinguishing features between the two species. Moreover, in *D. obscura*, tergites 2-3 commonly display a small pale area at the lateroventral corners. The oviscapt valve might also possess distinct characteristics aiding in its differentiation from other species (Bächli et al., 2004).

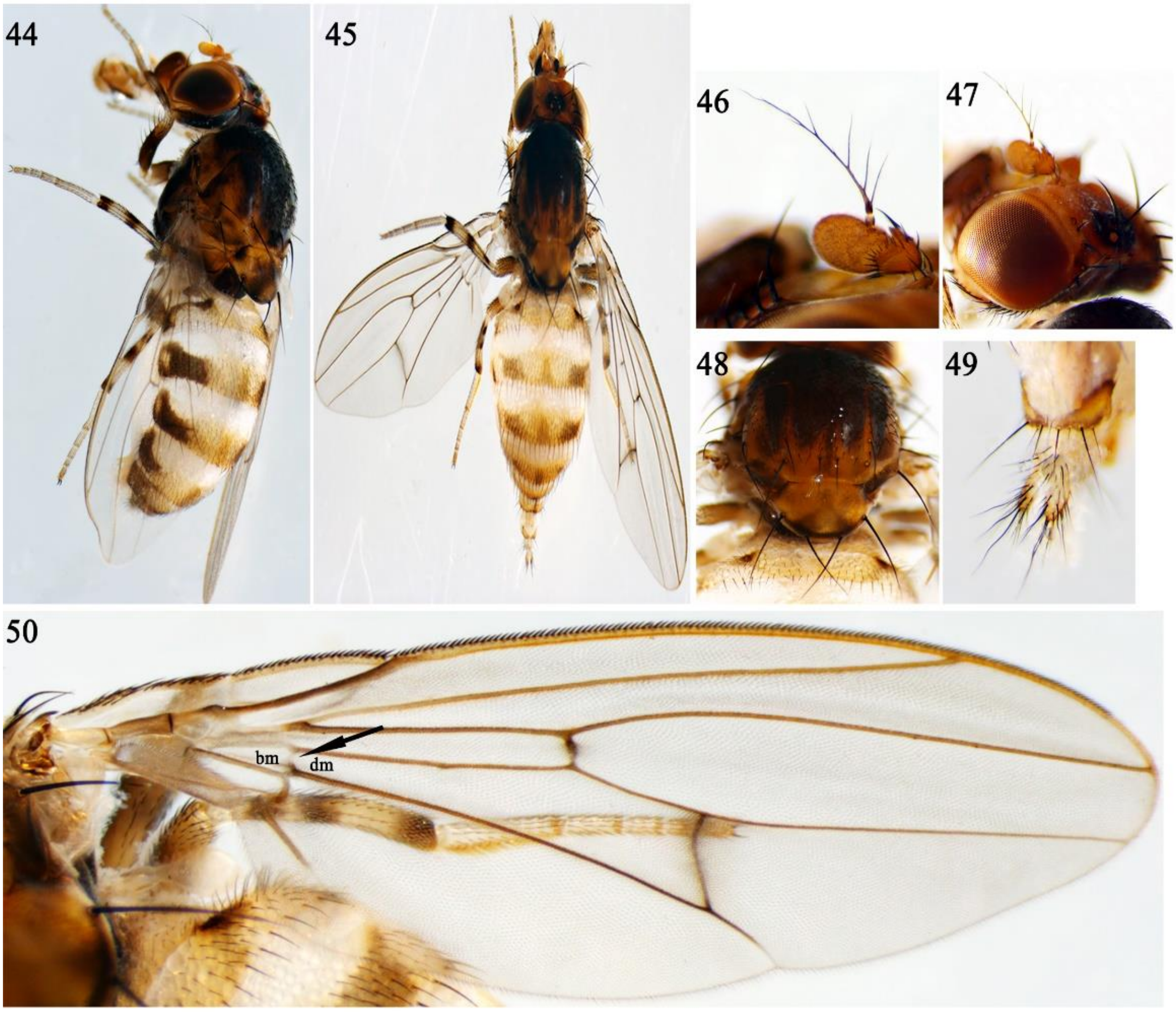

Figure 44–50. Characters of *Phortica variegata* (44–50): (44) male body, (45) female body, (46) male carina, (47) a pale white ring of male eyes, (48) male metanotum, (49) female terminalia organ, (50) male wing.

Şekil 44-50. *Phortica variegata'nın karakterleri (44–50): (44) erkek vücudu, (45) dişi vücudu, (46) erkekte carina, (47) erkekte gözler cevresinde soluk beyaz halka, (48) erkekte metanotum, (49) dişi terminal organ, (50) erkekte kanat.*

The list of species belonging to the Drosophilidae family in Türkiye is presented as 36 species by (Koçak & Kemal, 2013). The online TaxoDros database now lists 52 species of Drosophilidae (Taxodros, 2023). In addition, it appears that some species names published in the TaxoDros online database contain errors and some are given repeatedly. Consequently, this study necessitated the meticulous verification and correction of these discrepancies, leading to the compilation of a comprehensive checklist of Drosophilidae species in Türkiye.

4. Conclusion

Konya has become a very important location for strawberry cultivation in recent years. This study was to determine the *Drosophila* species that cause data loss in fruits. This is the first report on the occurrence of *Drosophila busckii*, *Drosophila hydei*, *Drosophila obscura*, *Drosophila subobscura*, *Drosophila simulans*, *Drosophila suzukii*, *Hirtodrosophila cameraria, ,* *Phortica variegata* and *Zaprionus indianus* in strawberry orchards in Konya, Türkiye. Additionally, *Drosophila obscura, Drosophila simulans, Hirtodrosophila cameraria* and *Phortica variegata* were determined for the first time in Türkiye's strawberry fields. Most of these species are invasive and threaten fruit species.

These results emphasize the importance of regional biodiversity by contributing to the Drosophilidae fauna of Türkiye. In addition, detailed descriptions and photographs of the morphological characteristics of the ten species determined in this study are included in order to facilitate identification. Due to their potential for rapid reproduction and spread, further studies are required to determine the population dynamics and distributions of these species, as well as their host plants and feeding habits.

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CHECKLIST OF DROSOPHILIDAE (DIPTERA) OF TURKIYE
ORDER: DIPTERA
SUBORDER: BRACHYCERA
SUPER FAMILY: EPHYDROIDEA

Sub Family: Steganinae Herdel, 1917
Genus *Acletoxenus* Von Frauenfeld
Acletoxenus formosus (Loew 1864)
Genus *Amiota* Loew
Amiota allemandi Bachli, Vilela & Haring, 2002
Amiota goetzi Maca,1987
Amiota subtusradiata Duda, 1934
III. Genus: *Phortica* Schiner 1862
Phortica semivirgo Maca,1977
Phortica variegata Fallén,1823
Genus: *Chymomyza* Czerny
Chymomyza amoena Loew,1862
Chymomyza procnemoides (Wheeler, 1952)
Genus: *Gitona*
Gitona distigma Meigen,1830
Genus: *Leucophenga* Mik, 1886
Leucophenga acutipennis Malloch, 1926
Leucophenga maculata Dufour, 1839
Leucophenga helvetica Bachli, Vilela & Haring, 2002
Leucophenga obscuripennis Loew
Subfamily Drosophilinae Rondani, 1856
Genus: *Drosophila* Fallén, 1823
Sub genus: *Drosophila (Dorsilopha*) Sturtevant 1942
Drosophila (Dorsilopha) busckii Coquillett, 1901
Sub genus: *Drosophila (Drosophila*) Fallen
Drosophila buzzatii Patterson & Wheeler,1942
Drosophila flavicauda Toda,1991
Drosophila funebris Fabricius, 1787
Drosophila histrio Meigen, 1830
Drosophila hydei Sturtevant, 1921
Drosophila immigrans Sturtevant, 1921
Drosophila kuntzei Duda, 1924
Drosophila littoralis Meigen, 1830
Drosophila nigrosparsa Strobl, 1898
Drosophila phalerata Meigen, 1830
Drosophila picta Zetterstedt, 1847
Drosophila repleta Wollaston, 1858
Drosophila schachti Baechli, Vilela & Haring 2002
Drosophila testacea Roser, 1840
Drosophila transversa Fallen, 1823
Drosophila virilis Sturtevant,1916
Sub genus: *Drosophila (Sophophora*) Sturtevant, 1939
Drosophila (Sophophora) ambigua Pomini,1940
32. *Drosophila glabra* Chen & Gao, 2015
33.*Drosophila (Sophophora) melanogaster* Meigen, 1830
34. *Drosophila (Sophophora) obscura* Fallén, 1823
35. *Drosophila (Sophophora) simulans* Sturtevant, 1919
36. *Drosophila (Sophophora) subobscura* Collin, 1936
37. *Drosophila (Sophophora) suzukii* (Matsumura, 1931)
38. *Drosophila tristis* Fallen 1823
Sub genus: *Drosophila (Lordiphosa*) Basden
39.*Drosophila (Lordiphosa) andalusiaca* Strobl
40. *Drosophila (Lordiphosa) fenestrarum* Fallén, 1823
Genus: *Hirtodrosophila* Duda, 1923
41. *Hirtodrosophila cameraria* (Haliday, 1833)
42. *Drosophila (Hirtodrosophila) confusa* Staeger,1844
Genus: Scaptodrosophila Duda
43. *Drosophila (Scaptodrosophila) deflexa* Duda,1924
44. *Drosophila (Scaptodrosophila) rufifrons* Loew,1873
Genus: Scaptomyza Hardy, 1850
45. *Scaptomyza adusta* Loew,1862
46. *Scaptomyza flava* Fallén,1823
47. *Scaptomyza graminum* Fallen, 1823
48. *Scaptomyza griseola* (Zetterstedt 1847)
49. *Scaptomyza pallida* (Zetterstedt,1847)
Genus: *Zaprionus* Coquillett, 1901
50. *Zaprionus indianus* Gupta, 1970
51. *Zaprionus tuberculatus* Malloch
52. *Zaprionus ghesquierei* Collart, 1937

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Determination of CO_2 Emission Based on Fuel Consumption in Wheat and Corn Production in TR 62 Region and Projection Estimate

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Abstract: This study aims to determine the carbon dioxide emission based on fuel consumption in wheat and corn production of Adana and Mersin provinces covering TR62 Region between 2014 and 2023 years and to estimate the projection for future years. The average CO_2 emission, specific fuel consumption and specific CO_2 emission values of TR62 Region based on fuel consumption in wheat production are 87.19 ktCO_2, 30.80 g_{fuel} $kg_{product}^{-1}$ and 105. 29 gCO_2 $kg_{product}^{-1}$, while in corn production between 2014 and 2023 years, the average CO_2 emission, specific fuel consumption and specific CO_2 emission values for the same years were determined as 37.13 ktCO_2, 100.97 g_{fuel} kg_{crop}^{-1} and 345.19 gCO_2 $kg_{product}^{-1}$ respectively. Total CO_2 emissions are expected to decrease, while specific fuel consumption and specific CO_2 emission values are expected to increase according to the future projections calculated between 2024 and 2033 years in wheat and corn production in TR62 Region.

Keywords: Total CO_2 emission, specific fuel consumption, specific CO_2 emission, projection coefficient

TR 62 Bölgesinin Buğday ve Mısır Üretiminde Yakıt Tüketimine Dayalı CO_2 Emisyonunun Belirlenmesi ve Projeksiyon Tahmini

Özet: Bu çalışma, TR62 Bölgesini kapsayan Adana ve Mersin illerinin 2014-2023 yılları arasındaki buğday ve mısır üretimindeki yakıt tüketimine dayalı karbondioksit emisyonunu belirlemeyi ve gelecek yıllara ait projeksiyon tahminini amaçlamıştır. TR62 Bölgesinin 2014-2023 yılları arasındaki buğday üretimindeki yakıt tüketimine dayalı olarak meydana gelen ortalama CO_2 emisyonu, özgül yakıt tüketimi ve özgül CO_2 emisyonu değerleri sırasıyla 87.19 ktCO_2, 30.80 $g_{yakıt}$ $kg_{ürün}^{-1}$ ve 105.29 gCO_2 $kg_{ürün}^{-1}$ olarak belirlenirken, mısır üretiminde ise aynı yıllara ait ortalama CO_2 emisyonu, özgül yakıt tüketimi ve özgül CO_2 emisyonu değerleri de sırasıyla 37.13 ktCO_2, 100.97 $g_{yakıt}$ $kg_{ürün}^{-1}$ ve 345.19 gCO_2 $kg_{ürün}^{-1}$ olarak belirlenmiştir. TR62 Bölgesinin buğday ve mısır üretiminde 2024-2033 yıllarına ait hesaplanan gelecek projeksiyon tahminlerine göre, toplam CO_2 emisyonunun düşme eğiliminde, buna karşın özgül yakıt tüketimi ve özgül CO_2 emisyon değerlerinin ise artma eğiliminde olacağı öngörülmektedir.

Anahtar Kelimeler: Toplam CO_2 emisyonu, özgül yakıt tüketimi, özgül CO_2 emisyonu, projeksiyon katsayısı

1. Introduction

The agricultural production sector is not only affected by climate change but also causes climate change. Although the industrial agriculture and food production system is profitable and efficient, it also causes many health, environmental and economic problems. The tillage, sowing-planting-maintenance, irrigation, spraying, harvesting and post-harvest operations cause climate change and thus the formation of greenhouse gas emissions in the agricultural production sector (Lal, 2004; Massey et al., 2019). The productivity and profitability are evaluated together with the energy consumed in agricultural production. There is an increase in exhaust emissions and greenhouse gas emissions from fuel and engine oil consumed by tractors and agricultural machinery due to the change and development in agricultural production technologies (Küsek, 2018).

The total greenhouse gas emission for 2015 was 475 MtCO_2 equivalent and it is explained that 72% (340 MtCO_2 equivalent) of the total emission was from energy, 13% (61 MtCO_2 equivalent) from industrial processes, 12% (57 MtCO_2 equivalent) from agriculture and 3% (17 MtCO_2 equivalent) from waste according to the national inventory report submitted by Turkey in

2017 under the Framework Convention on Climate Change (Ağaçayak and Öztürk, 2017).

According to TURKSTAT 2020 greenhouse gas inventory results, total greenhouse gas emissions were calculated as 523.9 $MtCO_2$ equivalent with an increase of 3.1% compared to the previous year, and total greenhouse gas emissions per capita were 4 tons of CO_2 equivalent in 1990 and 6 tons in 2019, 2 tons of CO_2 equivalent in 1990 and 6.3 tons of CO_2 equivalent in 2020, and greenhouse gas emissions in the agriculture sector were calculated as 73.2 $MtCO_2$ equivalent in 2020, increasing by 58.8% compared to 1990 and 7.5% compared to the previous year (TURKSTAT, 2020).

Natural disasters such as changes in precipitation regime, temperature increases, desertification as a result of drought, floods and storms due to climate change threaten agricultural production efficiency, ecosystem and indirect economy at a significant level (Hayaloğlu, 2018). Turkey is one of the countries most vulnerable to the impacts of climatic change. According to the International Panel on Climate Change (IPCC, 2013), Turkey is expected to have a drier and warmer climate in the coming decades, as well as a more uncertain climate structure in terms of precipitation. Since Turkey, with its existing agro-ecosystems, is being negatively affected by climatic changes to a great extent, it is not possible to achieve sustainable development without eliminating the causes of these changes (Smagulova et al., 2017; Aydın and Aktuz, 2023).

According to the International Panel on Climate Change report, 95% of climate change is human-induced and therefore, greenhouse gas emissions should be reduced to take measures to prevent the negative effects of climate change (IPCC, 2013). All countries must make high-level investments to reduce greenhouse gas emissions (Aydın, 2023). Öztürk and Vulkan (2017) emphasize that the use of fuel and engine oil as energy sources in agricultural production, the incompatibility of the power balance of tractors and agricultural machinery, and the use of overloaded engines result in the release of smoke, toxic and harmful substances into the atmosphere with exhaust gas emissions, and in this respect, it is necessary to reduce greenhouse gas emissions that cause global warming for safe and healthy food production.

For sustainable agriculture, using energy more effectively in all production periods, reducing fossil fuel consumption, consuming this type of fuel at a minimum level, reducing greenhouse gas emissions and developing more effective agricultural systems (Öztürk, 2017). For this purpose, although many provincial, regional and national studies are being carried out to control and minimize the increase in greenhouse gas emissions, these studies should be increased (Gołasa et al., 2021). Studies and possible new studies in this direction are of great importance in terms of determining emissions by IPCC methods for the province, region and country in general and making accurate predictions for the future.

Çukurova region has a very important place in Turkey in terms of agricultural production. Ecologically, more tractors and agricultural machinery are also needed at the point where many crops can be produced in every period. In their study, Gül et al. (2022) examined the changes in the number of tillage tools and machines and sowing machines, number of tractors, agricultural mechanization level (kW, kW ha^{-1}, tractor 1000 ha^{-1}, ha $tractor^{-1}$) of Adana and Mersin provinces within the TR62 Region for the years 2013-2022 and made projection estimates for the years 2023-2032 by chain index method. As a result of the study, they emphasized that the number and power of tractors will increase for Adana and Mersin provinces within the TR62 Region, and the mechanization level criteria will also increase.

Recently, the studies on the determination of greenhouse gas emissions occurring in the production of different crops in many provinces, regions and countries (Bilgili et al. (2018) for olive production in the Eastern Mediterranean Region, Kuzu et al. (2024) for wheat and corn production in the Eastern Mediterranean Region, Küsek (2018) for lentil production in the Southeastern Anatolia Region, (Öztürk and Vulkan, 2017) for wheat and corn at the Turkish scale, (Öztürk et al. (2017) for cotton production at the Turkish scale, etc.) have been carried out. In addition, the precautions and new plans to be taken for the results of these studies have also been emphasized. In this study, it was aimed to determine the carbon dioxide emission values based on fossil fuel consumption in wheat and corn production, which are intensively produced in TR62 Region, and it tried to make estimates of emission projections for the coming years.

2. Material and Method

TR62 Region includes Adana and Mersin provinces. Adana province is located on both sides of the Seyhan River at latitude 35 north and longitude 34°-36° east. Mersin province is between 36-37° north latitude and 33-35° east longitude. While 30% of Turkey's land is agricultural, 38% of Adana's and 25% of Mersin's land is agricultural.

Figure 1. Location of TR62 Region on the map of Türkiye.
Şekil 1. *Türkiye haritasındaki Tr62 Bölgesinin konumu.*

TR62 region is a region where the use of tractors and agricultural machinery is intensive in the agricultural sector and can easily adapt to new technologies with its fertile soils (Anonymous, 2017). The location of TR62 Region on the map of Türkiye are given in Figure 1.

The total agricultural areas in TR62 Region are 831 965 ha. Adana province of the TR62 Region is the province where wheat, which accounts for 69% of Turkey's cereal cultivation area and ranks first, is harvested the earliest. Adana province accounts for approximately 5% of Turkey's wheat production and 17.5% of total corn production, according to 2023 statistics (Anonim, 2024). In 2020, 234 835 tons of wheat and 89 705 tons of corn were harvested from 85 571 ha of wheat and 8217 ha of corn cultivation areas in Mersin province within the TR62 Region (TURKSTAT, 2020).

The cultivated area, production amount and yield values for the main crops wheat and corn production for the TR62 Region between 2014 and 2023 years are taken from the Turkish Statistical Institute (TURKSTAT) agricultural statistics (TURKSTAT, 2024) and given in Table 1 and Table 2.

There was a 35.28% decrease in wheat cultivated areas and a 27.98% decrease in the amount of production, while an increase of 11.27% was recorded in yield values between 2014 and 2023 years (Table 1).

The decreases of 7.26%, 9.19% and 2.08% were recorded in corn cultivated areas, production amounts and yield values between 2014 and 2023 years, respectively, according to Table 2.

Kuzu et al. (2024) explained that the fuel values consumed per unit area for wheat and corn were used in

Table 1. Wheat cultivated area, amount of product produced and product yield values in TR62 Region (TURKSTAT, 2024).
Çizelge 1. *TR62 Bölgesindeki buğday ekili alanı, üretilen ürün miktarı ve ürün verim değerleri (TÜİK, 2024).*

Year	Cultivated area (ha)	Crop production (ton)	Yield (ton ha^{-1})
2014	321 890	833 739	2.59
2015	301 138	1 002 320	3.33
2016	285 042	855 883	3.00
2017	275 467	926 480	3.36
2018	265 202	915 240	3.45
2019	217 828	722 292	3.32
2020	244 274	856 684	3.51
2021	236 326	932 600	3.95
2022	214 729	739 371	3.44
2023	208 336	600 447	2.88
Mean	***257 023***	***838 506***	***3.28***

Table 2. Corn cultivated area, amount of product produced and product yield values in TR62 Region (TURKSTAT, 2024).
Çizelge 2. *TR62 Bölgesindeki mısır ekili alanı, üretilen ürün miktarı ve ürün verim değerleri (TÜİK, 2024).*

Year	Cultivated area (ha)	Crop production (ton)	Yield (ton ha^{-1})
2014	110 658	1 241 290	11.22
2015	114 121	1 215 688	10.65
2016	115 755	1 307 130	11.29
2017	111 115	1 236 917	11.13
2018	87 250	995 804	11.41
2019	75 751	809 566	10.69
2020	78 372	909 683	11.61
2021	78 193	922 639	11.80
2022	96 996	988 970	10.20
2023	102 624	1 127 193	10.98
Mean	**97 084**	**1 075 490**	11.22

carbon dioxide emission calculations as 115.16 l ha^{-1} and 133.17 l ha^{-1}, respectively, for the fuel consumption values for wheat and corn production in TR62 Region. The oil consumption values for wheat and corn production in TR62 Region were calculated as 4.5% of total fuel consumption and the total amount of oil consumed for wheat and corn production was determined as 5.18 l ha^{-1} and 5.99 l ha^{-1}, respectively (Hacıoğlu et al., 2024).

The heating values and CO_2 emission factors of fuel and oil used in wheat and corn production for TR62 Region are given in Table 3 (IPCC, 1996; Öztürk et al., 2017; Bilgili and Aybek, 2018; Küsek, 2018).

The calculations of total CO_2 emissions from fuel and oil as described in the Intergovernmental Panel on Climate Change (IPCC, 1996) were taken into consideration, while calculating the CO_2 emissions from fossil (fuel and oil) sources for wheat and corn production in TR62 Region, (Öztürk et al., 2017; Bilgili and Aybek 2018; Küsek, 2018). The equations used in the calculation of CO_2 emissions based on fossil fuel and oil consumption are given in Table 3 and Table 4.

Table 3. The heating values and CO_2 emission factors of diesel fuel and engine oil used in wheat and corn production in TR62 Region.
Çizelge 3. *TR62 Bölgesindeki buğday ve mısır üretiminde tüketilen dizel yakıt ve kullanılan motor yağının ısıl değerleri ve CO_2 emisyon faktörleri.*

Fuel	Fuel heating value *($GJ\ l^{-1}$)*	CO_2 emission factor *($kg_{CO2}\ GJ^{-1}$)*
Diesel	0.0371	74.01
Engine oil	0.0382	73.28

Table 4. Equations used in the calculations for the determination of CO_2 emissions from fossil (fuel and oil) sources ((IPCC, 1996).
Çizelge 4. *Fosil (yakıt ve yağ) kaynaklı meydana gelen CO_2 emisyonlarının tespitine ait yapılan hesaplamalarda kullanılan eşitlikler ((IPCC, 1996).*

ToE= FCE + OCE [(Total CO_2 emissions = CO_2 emissions from fuel + CO_2 emissions from oil use)]
FCE= TD x FHV x FE [(Fuel-based CO_2 emissions ($kgCO_2$) = Total amount of diesel consumed (l) x Fuel lower heating value (0.0371 GJ l^{-1}) x Fuel emission factor (74.01 kg CO_2 G J^{-1})]
OCE= TO x OHV x OE [CO_2 emissions from oil use (kg CO_2) = Total amount of oil consumed (l) x Oil lower heating value (0.0382 GJ l^{-1}) x Oil emission factor (73.28 kg CO_2 G J^{-1})]

The following equations were used for specific fuel consumption (fuel consumption per amount of production) and specific CO_2 emissions (CO_2 emissions per amount of production) in determining crop-based fuel consumption and crop-based CO_2 emissions for wheat and corn production for the TR62 Region (Öztürk et al., 2017; Bilgili and Aybek 2018; Küsek, 2018). The density value of diesel fuel was taken into account as 0.84 g cm^{-3}, when converting the total amount of fuel consumed into grams (Beşergil, 2009).

$$SFC=ToE/CP \quad (1)$$

Where;
SFC : Specific fuel consumption ($g_{fuel}\ kg_{product}^{-1}$),
ToE : Amount of fuel consumed (g_{fuel})
CP : Amount of production ($kg_{product}^{-1}$).

$$SCE=ToE/CP \quad (2)$$

Where;
SCE : Specific CO_2 emission ($gCO_2\ kg_{product}^{-1}$),
ToE : Total CO_2 emissions (gCO_2),
CP : Amount of production ($kg_{product}^{-1}$).

The increase and decrease of values for total CO_2 emission (ToE), specific fuel consumption (SFC) and specific CO_2 emission (SCE) were calculated as percentages and the average coefficients of the percentages were obtained in wheat and maize production in TR62 Region between 2014 and 2023 years. A positive (+) projection coefficient indicates that the values for total CO_2 emission (ToE), specific fuel consumption (SFC) and specific CO_2 emission (SCE) have increased, while a negative (-) coefficient indicates that the ToE, SFC and SCE have decreased (Demir et al., 2013).

Calculations for the rate of change for the sample two years can be formulated as follows:

$$CR= [(Y_1 - Y_0) / Y_0] * 100 \quad (3)$$

Where:
CR: Rate of change (%) for the two sample years
Y_1= Values for ToE, SFC and SCE in the current year
Y_0= Values for ToE, SFC and SCE in the previous year

The values for ToE, SFC and SCE in the previous year was multiplied by the coefficient related to this ToE, SFC and SCE values, and in line with the decrease and increase of the coefficients, the 10-year projections of the ToE, SFC and SCE in wheat and corn production for the TR62 Region until 2033 were determined (Demir et al., 2013; Altuntaş, 2020).

3. Results and Discussion

Total CO_2 emissions, specific fuel consumption and specific CO_2 emissions calculated based on greenhouse gas emissions for TR62 Region are affected by wheat and corn production area, total production amount and yield values of the Region (Table 1 and Table 2). Accordingly, the changes in the cultivated area, production amount and yield values for the years 2014-2023 in wheat and corn production for the TR62 Region are given in Figure 2 and Figure 3.

Figure 2. Changes in wheat production values for TR62 Region by years.
Şekil 2. *TR62 Bölgesi için buğday üretim değerlerindeki yıllara göre değişim.*

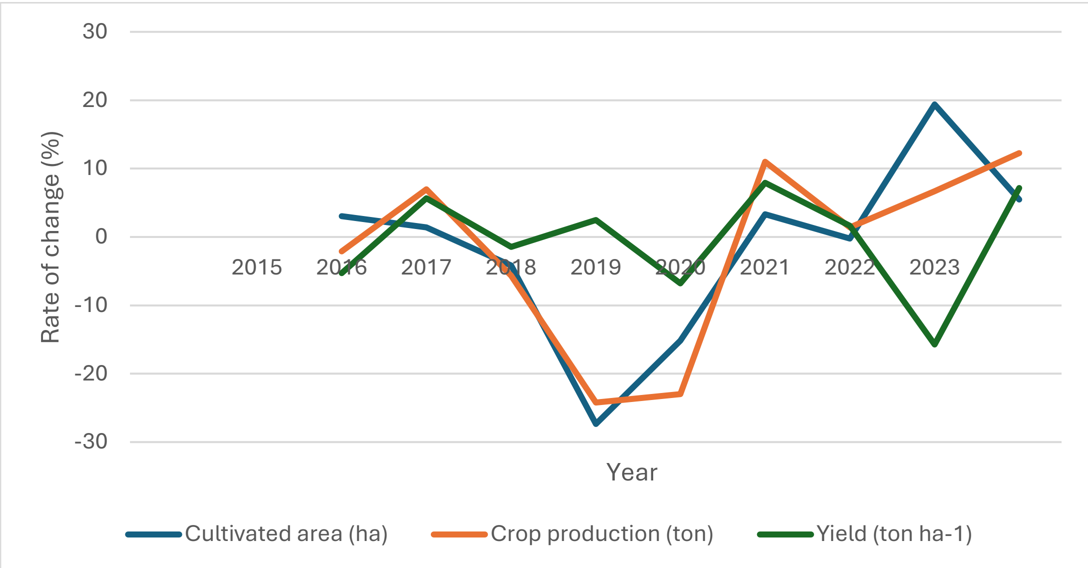

Figure 3. Changes in corn production values for TR62 Region by years.
Şekil 3. *TR62 Bölgesi için mısır üretim değerlerindeki yıllara göre değişim.*

The average values of total CO_2 emission (ToE), specific fuel consumption (SFC) and specific CO_2 emission (SCE) calculated in wheat production in TR62 Region for the last 10 years between 2014 and 2023 years are given in Table 5.

The average total CO_2 emission from fossil fuel (diesel+oil) in wheat production between 2014 and 2023 years was 87.19 $ktCO_2$, the average specific fuel consumption was 30.80 g_{fuel} $kg_{product}^{-1}$ and the average specific CO_2 emission was 105.29 gCO_2 $kg_{product}^{-1}$ for the TR62 Region, (Table 5).

According to Table 6, while there is a 7.25% decrease in total CO_2 emissions in corn production between 2014 and 2023, there is a 9.73% and 9.01%

increase for specific fuel consumption and specific CO_2 emissions, respectively. Figure 4 shows the change in total CO_2 emissions and specific CO_2 emissions for the previous year in wheat production for TR62 Region between 2014 and 2023 years.

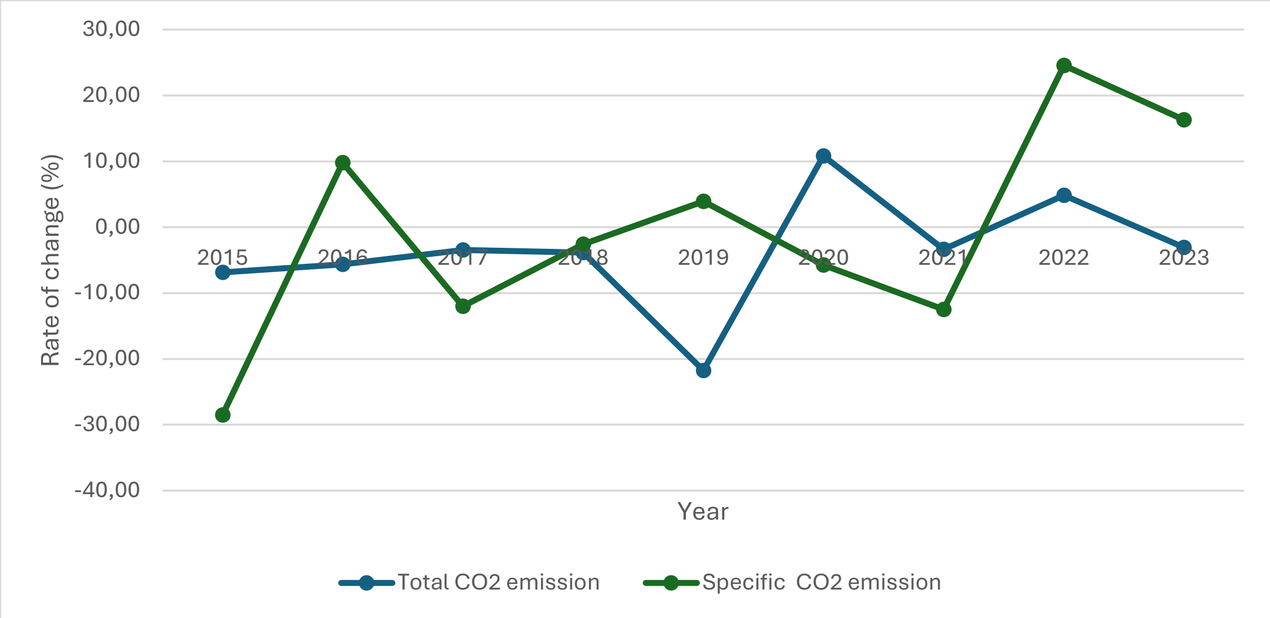

Figure 4. Changes in the values of total CO_2 emissions and specific CO_2 emissions in wheat production for TR62 Region by years.
Şekil 4. *TR62 Bölgesi için buğday üretimindeki toplam CO_2 emisyonu ve özgül CO_2 emisyonunun değerlerindeki yıllara göre değişim.*

Table 5. Changes in total CO_2 emissions, specific fuel consumption and specific CO_2 emissions in wheat production for TR62 Region between 2014 and 2023 years.
Çizelge 5. *TR62 Bölgesi için buğday üretiminde 2014-2023 yılları arasındaki toplam CO_2 emisyonu, özgül yakıt tüketimi ve özgül CO_2 emisyonunun değişimi.*

Year	ToE (Total CO_2 emissions, $ktCO_2$)	SFC (Specific fuel consumption, g_{fuel} $kg_{product}^{-1}$)	SCE (Specific CO_2 emission (gCO_2 $kg_{product}^{-1}$)
2014	106.45	37.35	127.68
2015	99.59	29.06	99.36
2016	94.27	32.22	110.14
2017	91.10	28.76	98.33
2018	87.70	28.03	95.83
2019	72.04	29.17	99.73
2020	80.78	27.58	94.30
2021	78.16	24.51	83.80
2022	82.12	32.49	111.07
2023	79.67	38.81	132.69
Mean	***87.19***	***30.80***	***105.29***

Table 6. Changes in total CO_2 emissions, specific fuel consumption and specific CO_2 emissions in corn production for TR62 Region between 2014 and 2023 years.
Çizelge 6. *TR62 Bölgesi için mısır üretiminde 2014-2023 yılları arasındaki toplam CO_2 emisyonu, özgül yakıt tüketimi ve özgül CO_2 emisyonunun değişimi.*

Year	ToE (Total CO_2 emissions, $ktCO_2$)	SFC (Specific fuel consumption, g_{fuel} $kg_{product}^{-1}$)	SCE (Specific CO_2 emission (gCO_2 $kg_{product}^{-1}$)
2014	42.32	9.97	34.09
2015	43.64	10.50	35.90
2016	44.27	9.91	33.87
2017	42.49	10.05	34.35
2018	33.37	9.80	33.51
2019	28.97	10.47	35.78
2020	29.97	9.64	32.95
2021	29.90	9.48	32.41
2022	37.09	10.97	37.51
2023	39.25	10.18	34.82
Mean	***37.13***	***10.10***	***34.52***

The average total CO_2 emission from fossil fuel (diesel+oil) of corn production was determined as 37.13 $ktCO_2$, the average specific fuel consumption as 10.10 g_{fuel} $kg_{product}^{-1}$ and the average specific CO_2 emission as 34.52 gCO_2 $kg_{product}^{-1}$ between 2014 and 2023 years for the TR62 Region (Table 6). According to Table 6, there is a 2.13% increase in both specific fuel consumption and specific CO_2 emissions while there is a 7.26% decrease in total CO_2 emissions in corn production between 2014 and 2023 years.

Figure 5 shows the change in total CO_2 emissions and specific CO_2 emissions for the previous year between 2014 and 2023 years in corn production for TR62 Region. The lowest total CO_2 emission in the last 10 years was 72.04 kt CO_2 in 2019, the lowest specific fuel consumption was 24.51 g_{fuel} $kg_{product}^{-1}$ in 2021 and the lowest specific CO_2 emission was 83.80 gCO_2 $kg_{product}^{-1}$ in 2021 in wheat production for the TR62 Region. In wheat production for TR62 Region, the highest values of ToE, SFC, and SCE in the last 10 years were determined to be 106.45 $ktCO_2$ in 2014, 38.81 g_{fuel} $kg_{product}^{-1}$ in 2023, and 132.69 gCO_2 $kg_{product}^{-1}$ in 2023, respectively.

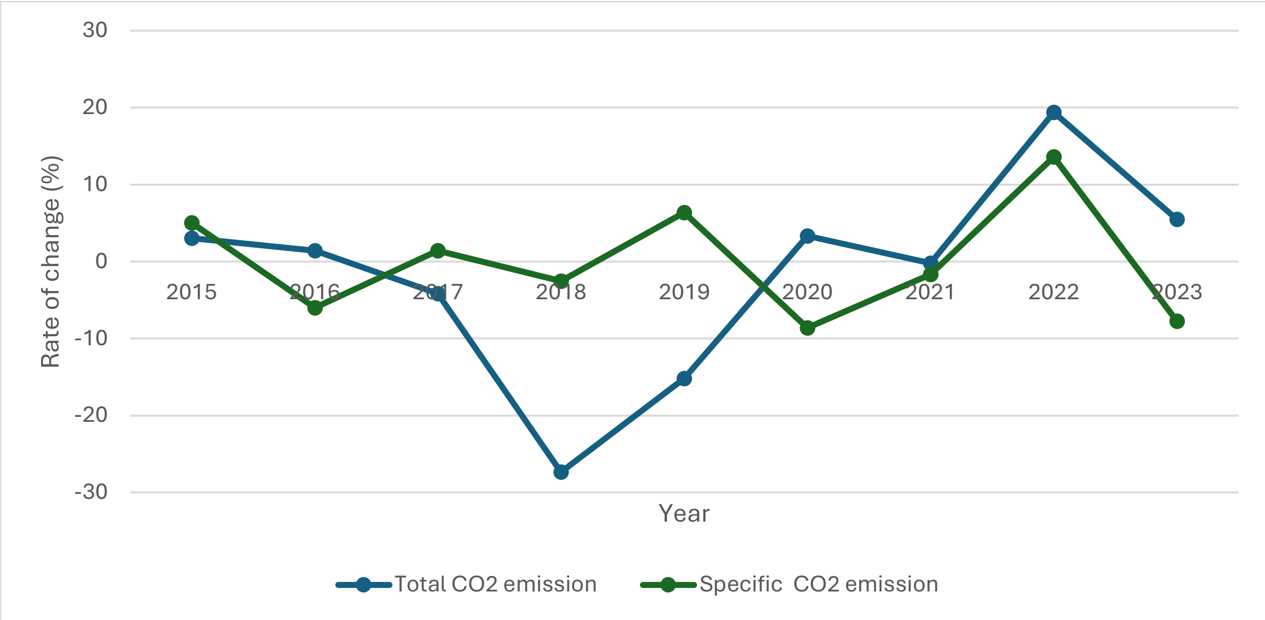

Figure 5. Changes in total CO_2 emission and specific CO_2 emission values in corn production for TR62 Region by years.
Şekil 5. *TR62 Bölgesi için mısır üretimindeki toplam CO_2 emisyonu ve özgül CO_2 emisyonunun değerlerindeki yıllara göre değişim.*

The lowest total CO_2 emission in the last 10 years was 28.97 $ktCO_2$ in 2019, the lowest specific fuel consumption was 9.48 g_{fuel} $kg_{product}^{-1}$ in 2021 and the lowest specific CO_2 emission was 32.41 gCO_2 $kg_{product}^{-1}$ in 2021 in corn production for the TR62 Region. the highest values of ToE, CVE, and CVE in the last 10 years were determined to be 44.27 $ktCO_2$ in 2016, 10.97 g_{fuel} $kg_{product}^{-1}$ in 2022, and 37.51 gCO_2 $kg_{product}^{-1}$ in 2022, respectively, in corn production for TR62 Region.

It is seen that specific CO_2 emission increases as wheat and corn production amounts decrease when specific CO_2 emission and wheat and corn production amounts are evaluated together.

Kuzu et al. (2024) reported that the averages of CO_2 emission values, specific fuel consumption and specific CO_2 emission values for wheat production in the Eastern Mediterranean Region for the years 2018-2022 were 154.50 $ktCO_2$, 26.60 g_{fuel} $kg_{product}^{-1}$ and 90.49 gCO_2 $kg_{product}^{-1}$, respectively, while the averages of CO_2 emission, specific fuel consumption and specific CO_2 emission values for wheat production in the TR62 Region for the years 2014-2023 were 87. 13 $ktCO_2$, 30.80 g_{fuel} $kg_{product}^{-1}$ and 105.29 gCO_2 $kg_{product}^{-1}$, respectively, and the values found in the TR62 Region were 77.33% lower than the Eastern Mediterranean Region, while the specific fuel consumption and specific CO_2 emissions gave similar values according to the total CO_2 emission values. Öztürk and Vulkan (2017) reported that the average CO2 emission in wheat production was 1.4 $MtCO_2$ $_{year}^{-1}$, specific fuel consumption was 20.7 g_{fuel} $kg_{product}^{-1}$ and specific CO_2 emission was 67.7 g_{CO2} $kg_{product}^{-1}$ in wheat production between 2010 and 2015 years for Turkey as a whole.

Projection estimates between 2024 and 2033 years are given in Table 7 and Table 8 by calculating the projections between 2024 and 2033 years by taking into account the values of ToE, SFC and SCE between 2014 and 2023 years in wheat and corn production for TR62 Region.

When the projection coefficients obtained according to the rates of change in the past years for the years 2024-2033 considering the CO_2 emission values, specific fuel consumption and specific CO_2 emission values between 2014 and 2023 years in wheat production for TR62 Region are examined, it is seen that it is -2.862 for ToE and 1.656 for the parameters SFC and SCE (Table 7).

It is estimated that as a parameter in the coming years, total CO_2 emissions will show a decrease, while specific fuel consumption and specific CO_2 emission values will show a loss of increase. The ToE, SFC and SCE values produced for the TR62 Region in 2033 are predicted to be 38.80 $ktCO_2$, 106.94 g_{fuel} $kg_{product}^{-1}$ and 365.60 gCO_2 $kg_{product}^{-1}$, respectively.

It is estimated that total CO_2 emission will show a decreasing trend, while specific fuel consumption and specific CO_2 emission values will show an increasing trend for each parameter in the coming years. In 2033, it is predicted that ToE, SFC and SCE values for wheat production for TR62 Region will be 59.59 $ktCO_2$, 45.74 g_{fuel} $kg_{product}^{-1}$ and 156.37 gCO_2 $kg_{product}^{-1}$, respectively.

Table 7. Projection estimates of wheat production in TR62 Region for the years 2024-2033 based on ToE, SFC and SCE values for the years 2014-2023.
Çizelge 7. *TR62 Bölgesi için buğday üretiminde 2014-2023 yıllarına ait ToE, ÖzY ve ÖzE değerleri dikkate alınarak 2024- 2033 yıllarına ait projeksiyon tahminleri.*

Year	**ToE** (Total CO_2 emissions, $ktCO_2$)	**SFC** (Specific fuel consumption, (g_{fuel} $kg_{product}^{-1}$)	**SCE** (Specific CO2 emission (gCO_2 $kg_{product}^{-1}$)
2014	106.45	37.35	127.68
2015	99.59	29.06	99.36
2016	94.27	32.22	110.14
2017	91.10	28.76	98.33
2018	87.70	28.03	95.83
2019	72.04	29.17	99.73
2020	80.78	27.58	94.30
2021	78.16	24.51	83.80
2022	82.12	32.49	111.07
2023	79.67	38.81	132.69
Projection coefficient	-2.862	1.656	1.656
2024	77.39	39.46	134.89
2025	75.18	40.11	137.12
2026	73.03	40.77	139.39
2027	70.94	41.45	141.70
2028	68.91	42.13	144.05
2029	66.93	42.83	146.43
2030	65.02	43.54	148.86
2031	63.16	44.26	151.32
2032	61.35	44.99	153.83
2033	59.59	45.74	156.37

Table 8. Projection estimates for corn production in TR62 Region for the years 2024-2033, taking into account the values of ToE, SFC and SCE for the years 2014-2023.
Çizelge 8. *TR62 Bölgesi için mısır üretiminde 2014-2023 yıllarına ait ToE, ÖzY ve ÖzE değerleri dikkate alınarak 2024- 2033 yıllarına ait projeksiyon tahminleri.*

Year	**ToE** (Total CO_2 emissions, $ktCO_2$)	**SFC** (Specific fuel consumption, (g_{fuel} $kg_{product}^{-1}$)	**SCE** (Specific CO2 emission (gCO_2 $kg_{product}^{-1}$)
2014	42.32	9.97	34.09
2015	43.64	10.50	35.90
2016	44.27	9.91	33.87
2017	42.49	10.05	34.35
2018	33.37	9.80	33.51
2019	28.97	10.47	35.78
2020	29.97	9.64	32.95
2021	29.90	9.48	32.41
2022	37.09	10.97	37.51
2023	39.25	10.18	34.82
Projection coefficent	-0.114	0.490	0.490
2024	39.20	10.23	34.99
2025	39.16	10.28	35.16
2026	39.11	10.33	35.33
2027	39.07	10.39	35.50
2028	39.02	10.44	35.68
2029	38.98	10.49	35.85
2030	38.94	10.54	36.03
2031	38.89	10.59	36.20
2032	38.85	10.64	36.38
2033	38.80	10.69	36.56

In Table 8, corn product production for the TR62 Region, CO_2 emission values, specific fuel consumption and specific CO_2 emission values for the years 2014-2023 are recorded. Emission coefficients obtained according to the change rates compared to previous years for the years 2024-2033, -0.114 for ToE, SFC and SCE was found to be 0.490.

It is estimated that as a parameter in the coming years, total CO_2 emissions will show a decrease, while specific fuel consumption and specific CO_2 emission values will show a loss of increase. The ToE, SFC and SCE values produced for the TR62 Region in 2033 are predicted to be 38.80 $ktCO_2$, 10.69 g_{fuel} $kg_{product}^{-1}$ and 36.56 gCO_2 $kg_{product}^{-1}$, respectively.

4. Conclusion

It was determined that the emissions of wheat and corn production in the TR62 Region were based on fossil fuel consumption. it is seen that specific CO_2 emissions are observed as the wheat and corn production amounts decrease when the specific CO_2 emission and wheat and corn production amounts are evaluated together and the changes over the years are examined.

ToE, SFC and SCE values of wheat and corn production for the TR62 Region between 2014 and 2023 years are recorded and the projections between 2024 and 2033 are produced. total CO_2 emission decreases and specific fuel consumption and specific CO_2 values increase as each parameter. It is expected to be recorded, according to the future years.

It is possible to reduce fossil fuel consumption by using energy more efficiently in all production periods for sustainable agriculture. Minimal consumption of fossil fuels will lead to a reduction in greenhouse gas emissions and the development of more efficient sustainable agricultural systems. The practices related to the use of protected tillage systems and increasing parcel sizes instead of conventional tillage systems in agricultural production, which cause the highest fuel consumption; limiting the use of chemical fertilizers and pesticide inputs required for crop production will ensure more efficient use and reduce greenhouse gas emissions. In addition, reducing the inputs of tractors and agricultural machinery in the production system, selecting the tractor and agricultural tools and machinery in harmony with the tractor and agricultural machinery at the point of ensuring power and machine harmony, and using the engine at optimal load will reduce the release of smoke, toxic and harmful substances into the atmosphere in exhaust emissions. Appropriate matching of agricultural field size and machine field capacity, new environmentally friendly production planning and switching from conventional tillage to conservation tillage will reduce fossil fuel consumption and improve the environmental profile.

In this context, it is important that the Organizations of the Ministry of Agriculture and Forestry, Universities and Farmers' Associations, under a common roof, carry out more serious studies and planning to increase production and quality and to take the mentioned measures to reduce carbon dioxide emissions under a common roof in order to reduce fuel consumption in fossil fuel-based production of CO_2 emissions on a provincial, regional, regional and national basis.

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Prediction of the Potential Spatial Distribution of the *Vitis vinifera* (Narince cv) Species Under Current and Future Climate Change Scenarios Using the MaxENT Model

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Abstract: The negative impacts of climate change, such as increasing competition among species, habitat loss, and spatial shifts, have a strong influence on species distribution areas. Species distribution models are an important tool for protecting species from the adverse effects of climate change and for conserving biodiversity. In this study, the spatial impact of climate change on the distribution of the Narince (*Vitis vinifera)* cultivar, which has a natural distribution in Turkey, was predicted using the MaxEnt model. To determine how the species' distribution areas would be affected by climate change, the HadGEM3-GC31-LL climate model, developed based on the principles of the Coupled Model Intercomparison Project Phase 6 (CMIP6), was used. The potential distribution of the Narince cultivar for the periods 2041–2060 and 2081–2100 was modeled under SSP 2.45 and SSP 5.85 scenarios. The results of the study show that the predicted current distribution area of Narince (*Vitis vinifera)* cultivar corresponds to its present geographic distribution. Under current climate conditions, suitable areas for the Narince cultivar are calculated as 21.837 km², highly suitable areas as 5.169 km², and the total suitable area as 27.006 km². For the future climate change scenarios, under the SSP 2.45 scenario, the total suitable areas are predicted to be 22,877 km² for the 2041–2060 period and 24,225 km² for the 2081–2100 period. Under the SSP 5.85 scenario, the total suitable areas are estimated to be 23.908 km² for the 2041–2060 period and 22.781 km² for the 2081–2100 period. According to our findings, it has been determined that the spatial distribution of the species will change under future climate change scenarios, and its distribution in Turkey is expected to decrease.

Keywords: MaxEnt, Species Distribution Modeling, Climate Change Scenarios, GIS

Mevcut ve Gelecekteki İklim Değişikliği Senaryoları Altında *Vitis vinifera* (Narince cv) Türlerinin Potansiyel Mekansal Dağılımının MaxENT Modeli Kullanılarak Tahmini

Öz: İklim değişikliğinin türler üzerindeki rekabeti artırma, yaşam alanı kaybı ve mekânsal değişikliği gibi olumsuz etkileri, türlerin dağılım alanları üzerinde güçlü bir etkiye sahiptir. Türleri iklim değişikliğinin olumsuz etkilerinden korumada ve biyolojik çeşitliliğin korunmasında tür dağılım modelleri önemli bir araçtır. Çalışmada Türkiye'de doğal yayılış alanı gösteren Narince üzüm (*Vitis vinifera*) çeşidinin iklim değişikliğinden mekânsal olarak nasıl etkileneceği MaxEnt model ile tahmin edilmiştir. Türün yayılış alanlarının iklim değişiminden nasıl etkileneceğini belirlemek için Birleştirilmiş Model Karşılaştırma Projesi Aşama 6 (CMIP6) ilkelerine göre geliştirilen HadGEM3-GC31-LL iklim modelinin SSP 2.45 ve SSP 5.85 senaryolarına göre türün 2041-2060 ve 2081–2100 yıllarındaki potansiyel yayılış alanı modellenmiştir. Çalışma sonucunda Narince çeşidinin (*Vitis vinifera)* günümüz tahmini yayılış alanı mevcut coğrafi dağılımı ile uyuşmaktadır. Günümüz iklim koşullarında çeşide uygun alanlar 21.837km^2, çok uygun alanlar 5.169 km^2 ve toplam uygun alan 27.006 km^2 olarak hesaplanmıştır. Gelecek iklim değişikliği senaryolarından SSP2.45 senaryosunda 2041-2060 yılları arasında toplam uygun alanların 22.877km^2, 2081-2100 yılları arasında 24.225km^2 olacağı tahmin edilmektedir. SSP 5.85 senaryosunda 2041-2060 yılları arasında toplam uygun alanların 23.908km^2, 2081-2100 yılları arasında 22.781 km^2 olacağı tahmin edilmektedir. Bu sonuçlarımıza göre gelecek iklim değişikliği senaryoları altında çeşidin mekânsal dağılışının değişeceği ve türün dağılımının Türkiye'de azalacağı tespit edilmiştir.

Anahtar Kelimeler: CBS, İklim Değişikliği Senaryoları, MaxEnt, Tür Dağılım Modelleme

1.Introduction

Biodiversity forms the foundation of life-support systems necessary for the continuation of life on Earth and humanity. Natural balances, food resources, industry, and economy are directly related to biodiversity. From this perspective, biodiversity is an important source of wealth and power for every country (Uyanık et al., 2012). The *Vitis vinifera* species plays a significant role in the ecosystem by supporting biodiversity (Cangi & Yağcı, 2017). It prevents soil erosion, provides habitat for insect and bird species, and supports biodiversity. In addition to its support for biodiversity, this species holds great value in Turkey due to its economic significance, genetic diversity, and potential health benefits. It can be utilized in various forms such as table grapes, raisins, verjuice, molasses, fruit leather, and vine leaves (Soltekin et al., 2021). These alternative uses contribute to the economy by supporting rural development (Uysal et al., 2021).

Ecological factors such as climate and geographical location have a direct impact on grape cultivation. Among the climatic characteristics, temperature is a critical parameter that determines whether viticulture can be practiced in a particular region (Daler et al., 2024). In addition to temperature, annual rainfall and its seasonal distribution are of critical importance for vine growth. High soil moisture during bud break, shoot, and flowering development, followed by dry and stable atmospheric conditions from flowering to fruit ripening, are essential factors for vine cultivation. Excessive rainfall can cause waterlogging in the grapes, and excessive humidity can negatively affect the plant in terms of disease and pests (Doğan & Uyak, 2021). Another climatic factor affecting viticulture is wind, particularly its intensity and direction. The most detrimental winds for viticulture are the spring winds coming from the north, which lower temperatures and cause breakage in young shoots. Additionally, the moist southern winds in spring can promote diseases. Strong winds near harvest time can also cause damage to grape berries, especially in table grape-growing regions, leading to product and quality loss (Ataş et al., 2021).

Climate change significantly impacts biodiversity by reshaping global ecosystems and species distributions, leading to ecosystem degradation, a decline in biodiversity, and species extinction (Biju Kumar & Kekeç & Kadıoğlu, 2020). These effects pose a significant threat to biodiversity (Gür, 2023). Monitoring the spatial distribution of species is crucial in combating climate change and conserving biodiversity (Miller, 2010). In this context, spatial modeling of the Narince grape cultivar is necessary for its conservation and to ensure sustainable grape production in Turkey.

MaxEnt is used in various fields such as species distribution modeling, spatial ecology studies, decision-making, and quantum mechanics (Elith et al., 2011). In the field of species distribution modeling, MaxEnt helps to understand the impact of climate change on the spatial distributions of species by using global climate model outputs as environmental predictors. Studies have proven that MaxEnt performs well in predicting training data and classifying out-of-bag data, making it a practical tool for handling imbalanced and biased data in species distribution models (Elith et al., 2011; Merow et al., 2013).

This study aims to highlight the role of *Vitis vinifera* (Narince cv) species in supporting biodiversity within ecosystems and its economic importance, while evaluating the potential effects of climate change on this significant species. Accordingly, the study seeks to model the impacts of climate change on the population dynamics and habitat suitability of *Vitis vinifera* (Narince cv) species with the goal of informing sustainable conservation strategies for the species.

2. Materials and Methods

2.1. Study area

The study area is defined as the natural distribution range of *Vitis vinifera* (Narince cv) species within Turkey's borders. Turkey, located between 36° 35′ and 42° 02′ north latitudes and 26° 04′ and 44° 07′ east longitudes, has a total area of 783.562 km². The average elevation of the study area is 1132 meters, which is higher than the average elevation of 700 meters of the Earth's landmasses. Elevation, aspect, and mountain structures in Turkey affect the distribution of precipitation, temperature, and relative humidity, leading to regional climatic variations (Atalay, 2017).

2.2. Spatial modeling

The study employed the Maximum Entropy Algorithm. The reliability of the model outputs was assessed using the AUC (Area Under the ROC Curve) value derived from ROC (Receiver Operating Characteristic) analysis (Phillips et al., 2006). The closer the AUC test value is to 1, the more sensitive and descriptive the model is (Akyol & Örücü, 2019). To determine the impact level of environmental variables, the Jackknife test was used. This method allows for the determination of the importance levels of each independent variable in model construction (Phillips et al., 2006).

2.3. Environmental variables and species occurrence data

Sample points for the Narince grape (*Vitis vinifera)* cultivar included in the MaxEnt model were downloaded from the GBIF (Global Biodiversity Information Facility) website (Figure 2). 70% of the sample points were randomly selected as training data, while the remaining 30% were used as test data. The aspect parameter included in the model was derived from the 12.5-meter resolution DEM (Digital Elevation

Model) data provided by the ALOS-PALSAR satellite, which includes elevation data. Organic Carbon and Clay maps were produced using data downloaded from the SoilGrids site. To predict the current distribution of the species, bioclimatic variables with a spatial resolution of 30 arc-seconds (~1 km) were downloaded from the WorldClim site and maps (Table 1) were generated using ArcGIS 10.5 software.

2.4. Climate model

To determine how climate change will affect the species' distribution areas, the HadGEM3-GC31-LL climate model, developed according to the principles of the Coupled Model Intercomparison Project Phase 6 (CMIP6), was used. Both the intermediate mitigation

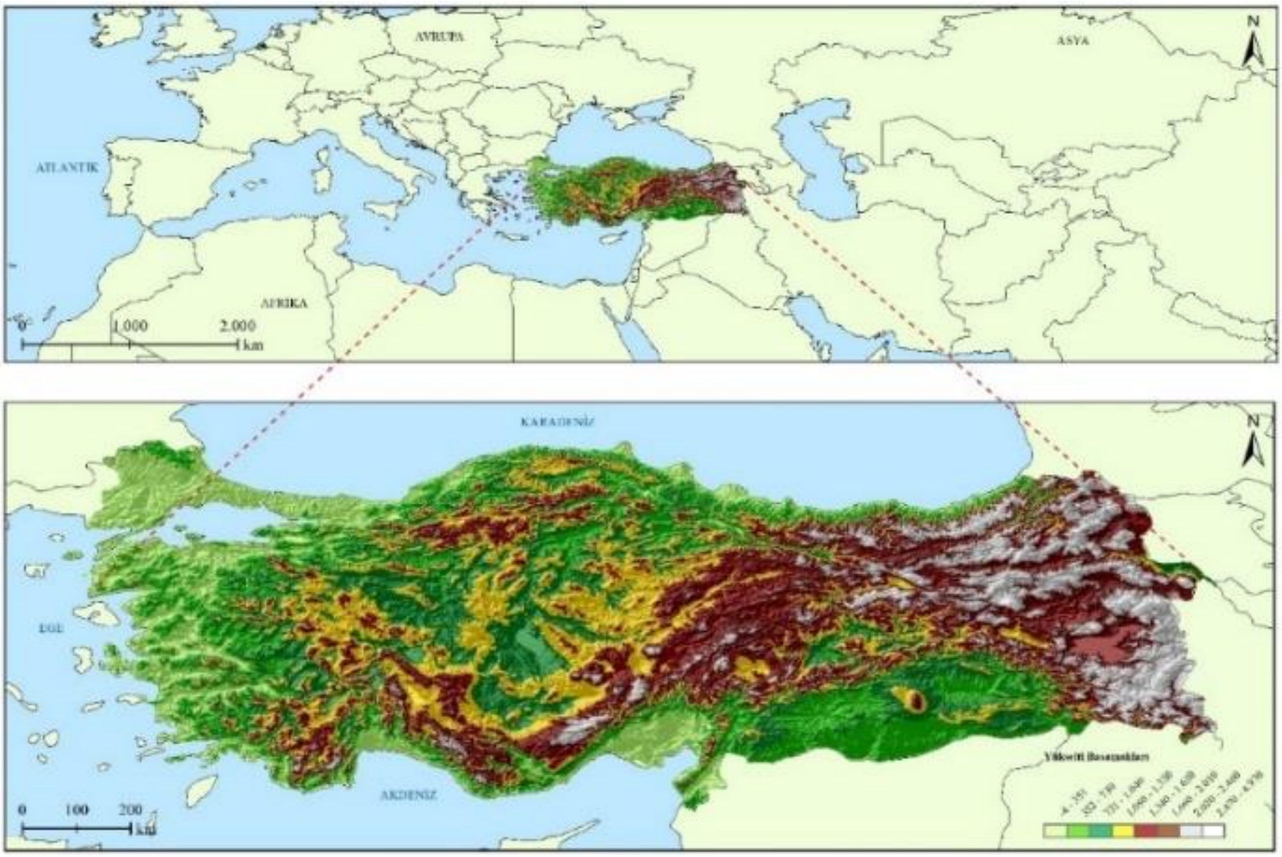

Figure 1. Location Map of the Study Area
Şekil 1. *Çalışma alanı haritası.*

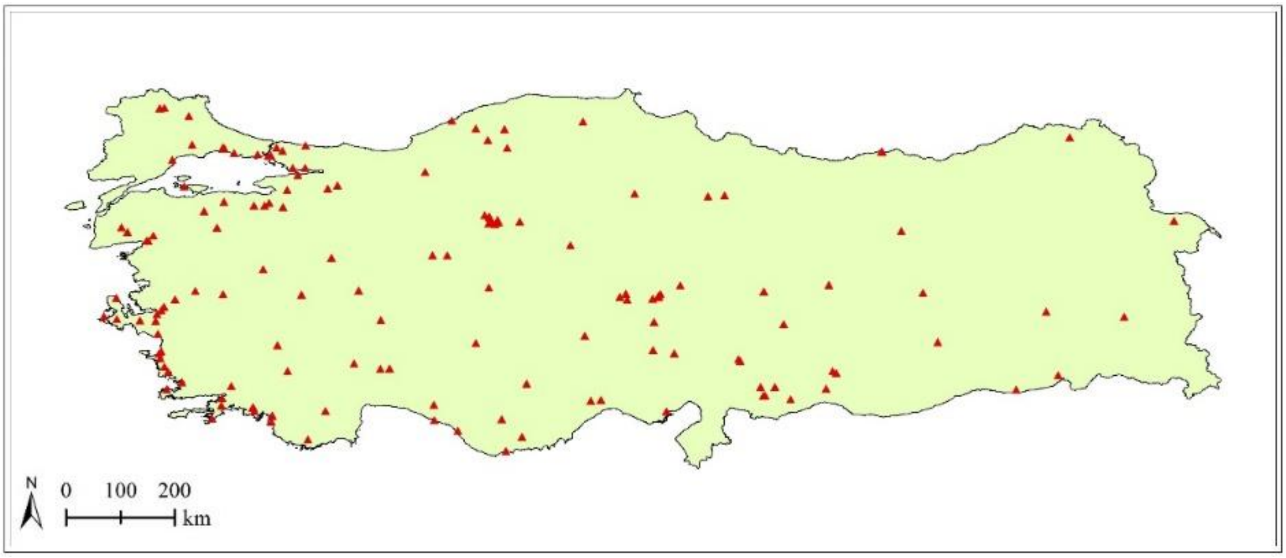

Figure 2. Species Distribution Points.
Şekil 2. *Türün noktasal dağılım haritası.*

Table 1: Environmental Variables Used in the Modeling.
Çizelge 1. *Modellemede kullanılan çevresel değişkenler.*

Code	Environmental Variable	Unit
Bio1	Annual mean temperature	°C
Bio2	Average of daily maximum and minimum temperatures	°C
Bio3	Isothermality (Equal temperature)	°C
Bio4	Seasonal temperature	°C
Bio5	Maximum temperature of the warmest month	°C
Bio6	Minimum temperature of the coldest month	°C
Bio7	Annual temperature range (Bio 5 – Bio 6)	°C
Bio8	Mean temperature of the wettest three months	°C
Bio9	Mean temperature of the driest three months	°C
Bio10	Mean temperature of the hottest three months	°C
Bio11	Mean temperature of the coldest three months	°C
Bio12	Annual precipitation	mm
Bio13	Precipitation of the wettest month	mm
Bio14	Precipitation of the driest month	mm
Bio15	Seasonal precipitation	mm
Bio16	Precipitation of the wettest three months	mm
Bio17	Precipitation of the driest three months	mm
Bio18	Precipitation of the hottest three months	mm
Bio19	Precipitation of the coldest three months	mm
Orgcar	Organic carbon	dg/kg
Clay	Clay content	percent
Aspect	Aspect	degree
cec	Cation exchange capacity	dg/kg
slope	Slope	degree
ph	pH watrer	pH*10
silt	Silt	degree

scenario (SSP 2-4.5) and the high mitigation scenario (SSP 5-8.5) were applied. Climate model data were downloaded from the WorldClim site in GeoTIFF format, processed using ArcGIS 10.5, and maps were generated.

3. Results and Discussion

3.1. Determination of the parameters to be used in the model

The presence of high multicollinearity among the parameters used in the MaxEnt model can lead to overfitting, thereby negatively affecting the accuracy of model predictions (Elith et al., 2011). To assess the multicollinearity between variables, Pearson correlation analysis was performed on the parameters listed in Table 1. According to Özcan (2024) and Esringü et al. (2021), parameters with a correlation value exceeding 0.70 contribute to multicollinearity. Consequently, variables such as bio 5, bio 6, bio 7, bio 13, bio 16, bio 17, bio 18, bio 19, cec, slope, pH, and silt were excluded from the model to prevent high variance inflation due to their significant multicollinearity.

The accuracy of the obtained model was evaluated using AUC assessment. According to this assessment, an AUC value of 0.50 indicates poor performance, an AUC value between 0.50 and 0.70 indicates average performance, and an AUC value above 0.70 indicates excellent performance (Li et al., 2024). In this context, the model's AUC value of 0.80 demonstrates good performance.

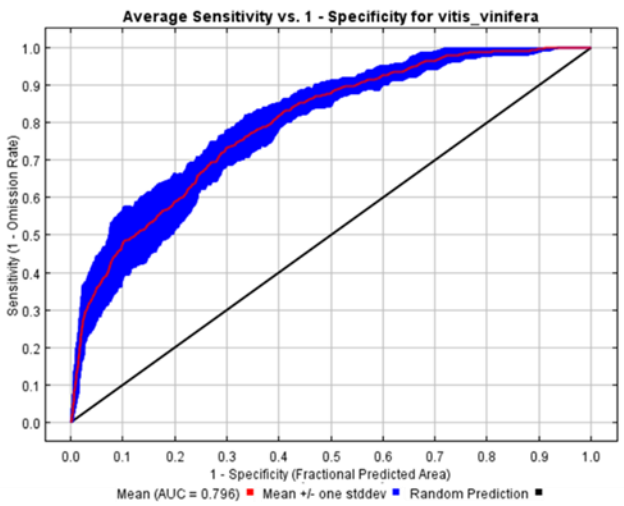

Figure 3. AUC Value Graph of the Model
Şekil 3. *Modelin AUC grafiği*

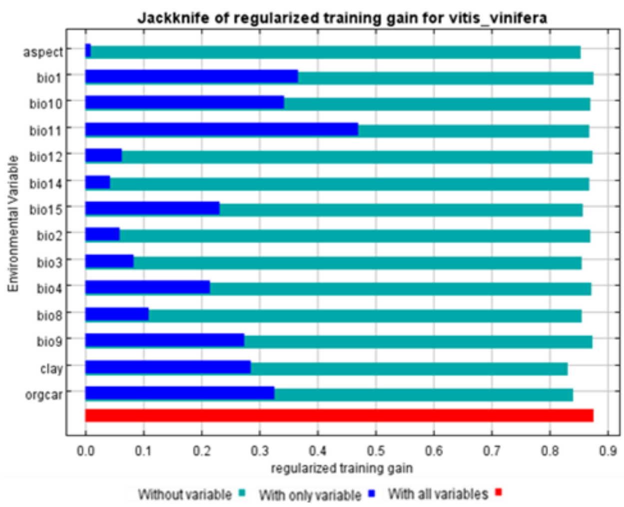

Figure 4. Jackknife Test Graph of the Model.
Şekil 4. *Modelin Jackknife Test Grafiği*

The Jackknife test is used to evaluate the effects of environmental variables on the model. It measures the contribution of each variable to the model (Li et al., 2024; Meena et al., 2024). The Jackknife test results for the Narince (*Vitis vinifera)* cultivar are presented in Figure 7. According to the graph, the parameter for the mean temperature of the coldest three months (bio11) is the environmental variable that provides the highest gain when used alone in the model. This is followed by Bio1, Bio10, and Organic Carbon. The environmental variable that most significantly reduces the gain when omitted is clay, suggesting it has the most unique information not provided by other variables. Zhang et al. (2021) used the MaxEnt model to analyze the distribution of grape species in Schuman by modeling their ranges between 1980 and 2019. According to the model results, the most suitable parameters for determining the potential distribution of grape species were, in order, accumulated active temperature, annual temperature, annual sunshine duration, and annual precipitation. These results align with our model outputs, where the annual precipitation parameter (bio1), which ranks second among the effective parameters in determining distribution, shows a notable similarity.

The response curves used to examine the importance of environmental variables are shown in Figure 5. Upon reviewing the response curves, it can be observed that parameters providing the most gain to the model include the Mean Temperature of the Coldest Three Months (bio11), the Mean Temperature of the Hottest Three Months (bio10), Seasonal Precipitation (bio15), Precipitation of the Driest Month (bio14), and the Mean Temperature of the Wettest Three Months (bio8). These parameters exhibit a positive relationship with the model. Conversely, Annual Mean Temperature (Bio1), Seasonal Temperature (bio4), Annual Precipitation (bio12), soil organic carbon, and clay parameters show a negative relationship with the model.

The MaxEnt model defines the probability of a species' presence within a given area using values ranging from 0 to 1 (Elith et al., 2011). In this study, threshold values for the spatial distribution of Narince (*Vitis vinifera)* cultivar were set as follows: 0-0.20 as unsuitable, 0.20-0.40 as not suitable, 0.40-0.60 as marginally suitable, 0.60-0.80 as suitable, and 0.80-1.0 as highly suitable. The map of the current potential spatial distribution areas of *Vitis vinifera* is shown in Figure 9.

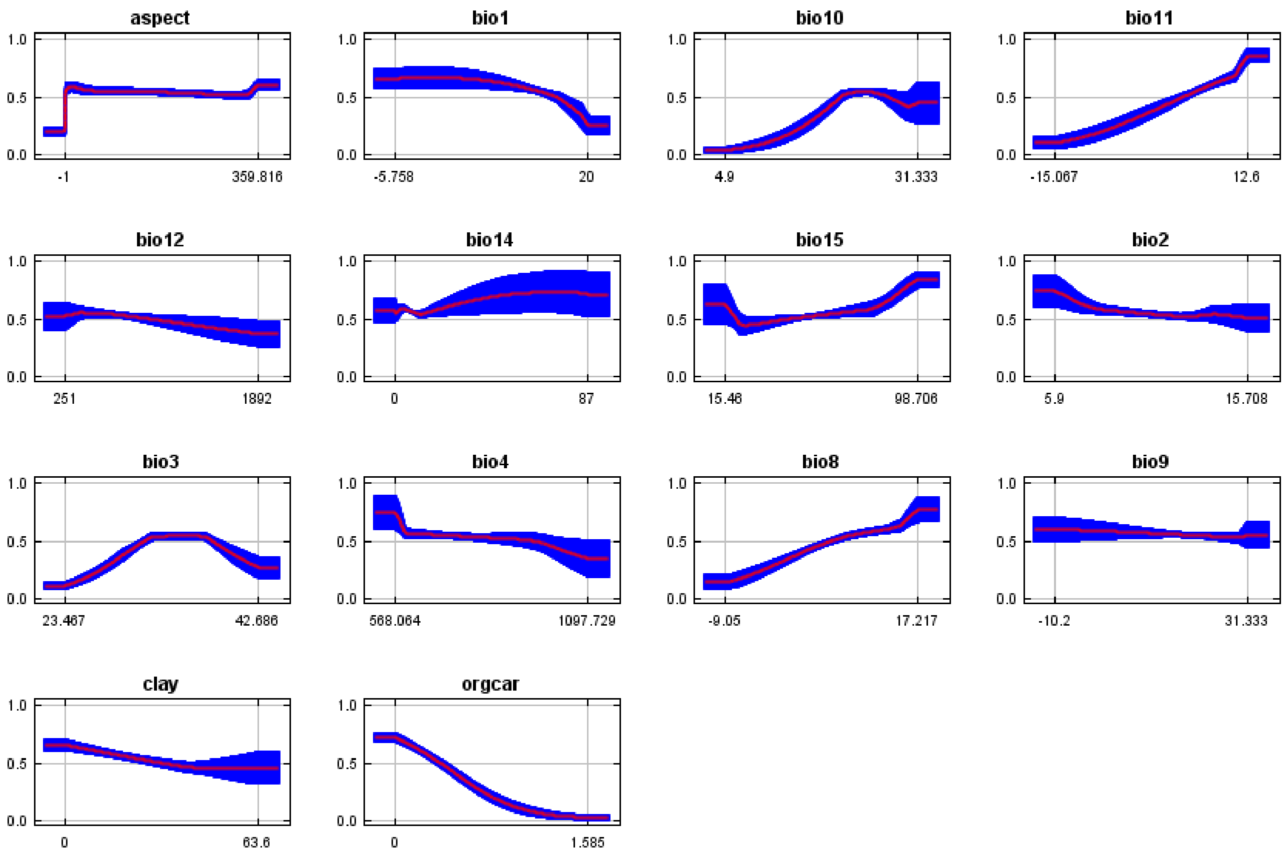

Figure 5. Response Curves of the Variables Included in the Model.
Şekil 5*. Modele Dahil Edilen Değişkenlerin Yanıt Eğrileri.*

The models created for Narince (*Vitis vinifera)* cultivar were converted to raster data in ArcGIS, threshold values were defined using the reclassify command, and area calculations were performed. The numerical data related to these calculated areas are provided in Table 2.

Table 2. Suitability levels of narince (*Vitis vinifera)* cultivar under different climate scenarios
Çizelge 2. *Narince (Vitis vinifera) çeşidinin farklı iklim senaryoları altında uygunluk düzeyleri*

Narince *(Vitis vinifera)* cultivar		SSP 2.45		SSP 5.85	
Suitability Level	**Current**	**2041- 2060**	**2081-2100**	**2041-2060**	**2081-2100**
Not Suitable	427.606	434.045	438.749	439.252	441.638
Unsuitable	246.424	248.154	243.092	234.367	231.110
Marginally Suitable	82.523	78.483	77.493	86.032	88.029
Suitable	21.837	16.735	17.406	18.687	17.203
Highly Suitable	5.169	6.142	6.819	5.221	5.578
Total (km²)	783.562	783.562	783.562	783.562	783.562

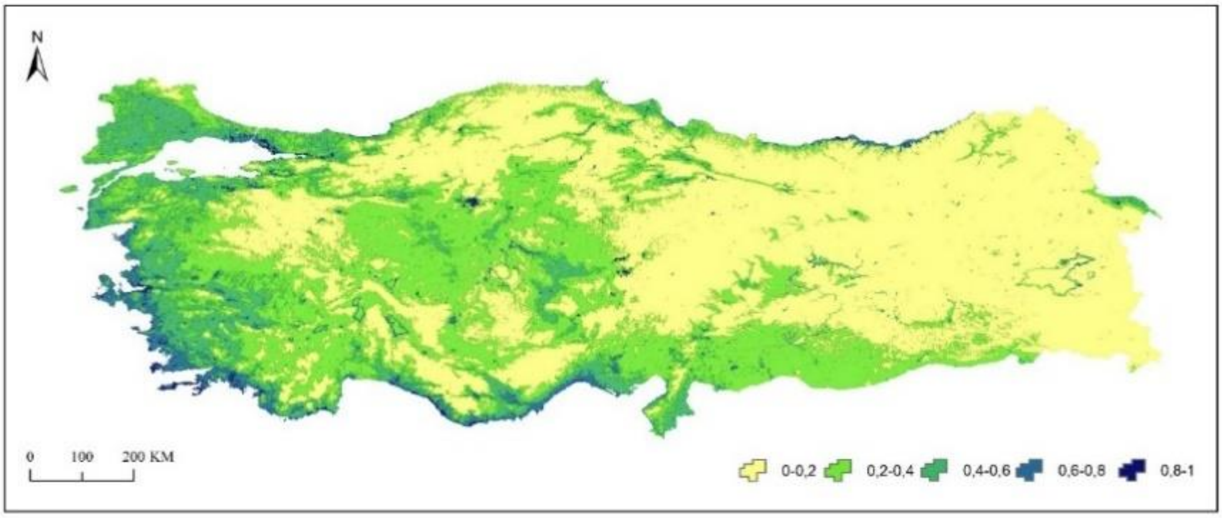

Figure 6. Predicted spatial distribution areas of *Vitis vinifera* (narince cv) species under current conditions.
Şekil 6. *Mevcut koşullar altında Vitis vinifera (narince çeşidi) türünün tahmin edilen mekansal dağılım alanları.*

Examining the map of the predicted spatial distribution areas under current conditions, it is observed that *Vitis vinifera* (Narince cv) species has very suitable growth conditions throughout the Aegean and Mediterranean coastal regions, the southern part of the Marmara Basin, the northern part of the Eastern Black Sea Basin, and the Tokat-Erbaa region. Suitable growing conditions are found in the Meriç, Susurluk, Gediz, and Büyük Menderes Basins. Conversely, the Aras, Çoruh, and Dicle Basins show conditions that are not suitable at all.

Climate change is having significant effects on ecosystems and biodiversity worldwide. In this context, understanding the future distribution and suitability changes for *Vitis Vinifera* (Narince cv) species is critical for assessing the impacts of climate change on agricultural production and biodiversity.

3.2. Potential spatial distribution under medium and high emission scenarios

Under the medium emission scenario, Narince (*Vitis vinifera)* cultivar maintains its density along the Mediterranean and Aegean coasts (Figure 7). These areas continue to provide suitable conditions for the species despite climate change. However, there are losses in marginally suitable areas in the Meriç, Marmara, Konya Closed, Sakarya, and Asi Basins. This can be attributed to microclimatic changes and reductions in water resources in these regions. In the southern part of the Dicle Basin, there is a shift from marginally suitable to suitable areas, indicating that the climate conditions in the region have become more favorable for the species. According to the numerical data in Table 2, the highly suitable areas increase from 5.169 km² to 6.142 km² between 2041-2060, while the suitable areas decrease from 21.837 km² to 16.735 km². This indicates an overall reduction in suitable areas despite an increase in highly suitable areas. In the period 2081-2100, highly suitable areas are projected to rise to 6.819 km², suitable areas to drop to 17.406 km², and unsuitable areas to increase to 438.749 km².

Under the high emission scenario, Narince (*Vitis vinifera)* cultivar continues to maintain its density along the Mediterranean and Aegean coasts. However, a transition from unsuitable to marginally suitable areas is observed in the Susurluk, Gediz, and Büyük Menderes Basins. This may indicate that the impacts of climate change are less severe in these regions, or that adaptation processes are occurring more rapidly. According to the numerical data in Table 2, highly suitable areas increase from 5,169 km² to 5.221 km² between 2041-2060, suitable areas decrease from 21.837 km² to 18.687 km², and unsuitable areas rise from 427.606 km² to 439.252 km². In the period 2081-2100, highly suitable areas are expected to increase to 5.578 km², suitable areas to decrease to 17.203 km², and unsuitable areas to rise to 441.638 km².

The results of this study highlight significant insights into how climate change may impact the spatial distribution of the Narince (*Vitis vinifera)* cultivar in Turkey. Using the MaxEnt model, we have projected potential changes in the habitat suitability of this key agricultural and biodiversity species under varying climate scenarios. The MaxEnt model demonstrated a reliable performance with an AUC value of 0.80, indicating that the model's predictions are robust and trustworthy. Jackknife analysis identified that the variable Average Temperature of the Coldest Three Months (Bio11) has the highest impact on the model, followed by Average Temperature of the Hottest Three Months (Bio10) and "Seasonal Precipitation (Bio15). These findings underscore the importance of temperature and precipitation variables in shaping the habitat suitability for Narince (*Vitis vinifera)* cultivar. Conversely, the "Clay" variable, when omitted, led to the most significant decrease in model performance, suggesting its critical role in the habitat modeling.

The current spatial distribution maps show that Narince cultivar thrives in the Aegean and Mediterranean coastal regions, southern Marmara Basin, northern Eastern Black Sea Basin, and Tokat-Erbaa region under present conditions. However, the impact of climate change scenarios (SSP 2.45 and SSP 5.85) reveals a troubling trend: potential suitable areas for Narince cultivar are expected to decline significantly in the future. Under the medium emission scenario (SSP 2.45), suitable areas are projected to decrease from 21.837 km² to 16,735 km² by 2041-2060, and further to 17.406 km² by 2081-2100 (Figure 7a, 7b). For the high emission scenario (SSP 5.85), suitable areas are expected to decrease from 21.837 km² to 18.687 km² by 2041-2060, and further to 17,203 km² by 2081-2100 (Figure 7c, 7d). These projections indicate a shift towards less suitable conditions for Narince cultivar in the future, with certain regions becoming less hospitable.

The decrease in suitable areas poses challenges for the sustainability of Narince (*Vitis vinifera)* cultivar cultivation and could affect the economic viability of grape production in Turkey. The results suggest that adapting agricultural practices and conservation strategies will be crucial to mitigate these impacts. Effective measures might include developing new cultivars resistant to climate stressors, optimizing water use, and enhancing soil management practices.

4. Conclusion

Species distribution modeling utilizes climatic and topographic factors to evaluate the impacts of climate change on species and potential spatial changes. It can also be employed to understand, interpret, and address the consequences of environmental changes on viticulture and biological diversity. Research indicates that climate change affects viticulture suitability and the distribution areas of the Narince cultivar by altering key determinants such as elevation, drought index, precipitation, and soil characteristics, potentially reducing the areas suitable for Narince cultivar cultivation.

In this study, the spatial impact of climate change on the Narince cultivar grapevine in Turkey was modeled using the MaxEnt algorithm. The results indicate that under the medium (SSP 2.45) and high (SSP 5.85) emissions scenarios of the Had-GEM climate model, the distribution areas of the Narince cultivar are projected to shrink between 2041-2060 and 2081-2100. This suggests a reduction in species diversity as one of the negative consequences of climate change.

When evaluating the ecological and economic aspects of the Narince cultivar, which is predicted to experience spatial loss under climate change scenarios, it is evident that negative impacts may arise in terms of biological diversity conservation, climate change mitigation, economic development, agricultural production, human health, and well-being. Currently, solutions to counteract these negative effects are insufficient. However, more research is needed to understand how we can mitigate the adverse effects of climate change on species spatial distributions. Therefore, more detailed studies on the interactions between climate change and species are necessary. Researchers and policymakers can use species distribution models to develop agricultural practices, conservation strategies, and land use plans that support the sustainability of grapevine cultivation. These efforts should address not only environmental impacts but also economic and social outcomes. Future research could facilitate spatial modeling efforts like this study and simplify the implementation of practices for conserving and sustaining biological diversity.

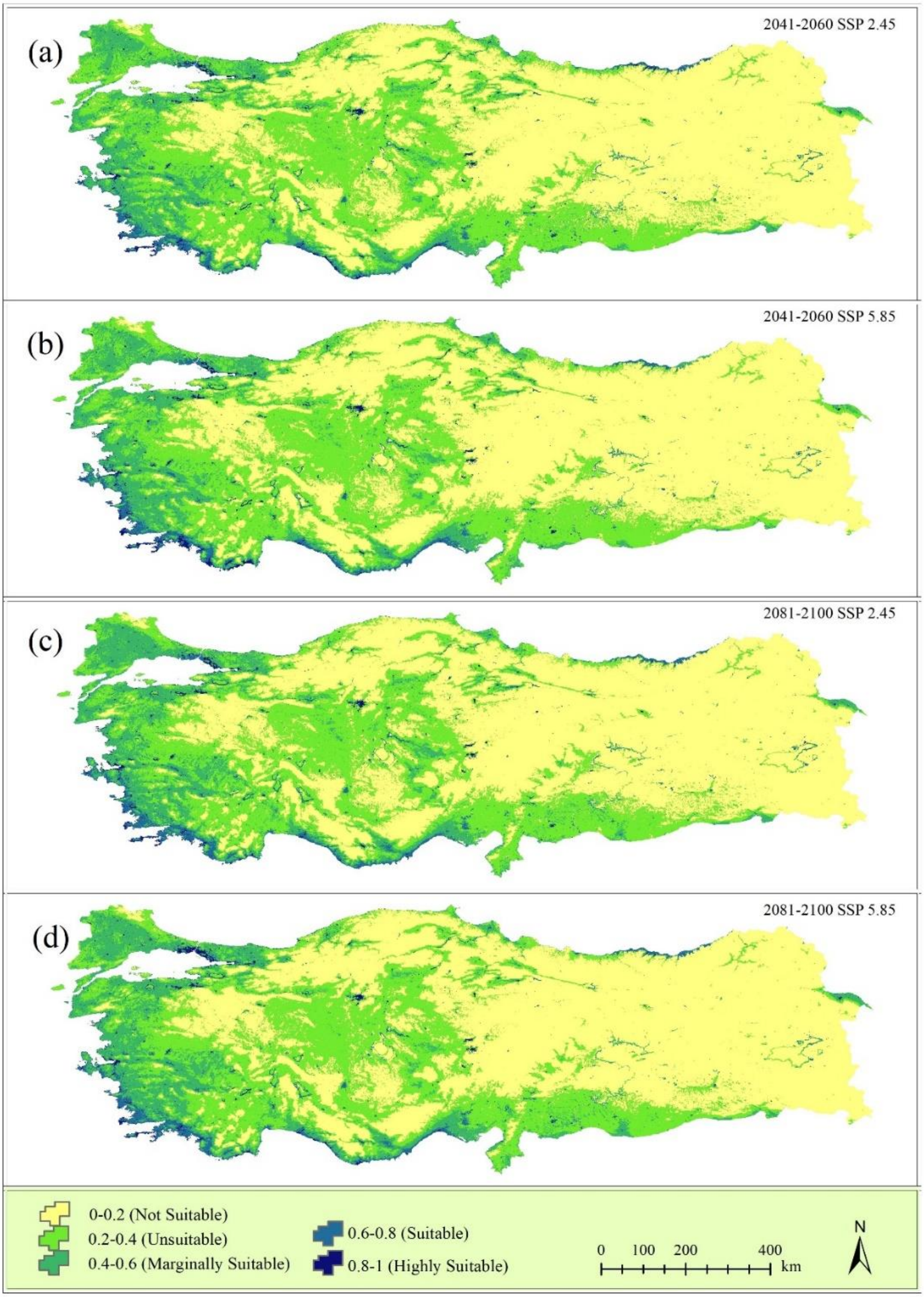

Figure 7: *Vitis vinifera* (Narince cv) Species Projected Spatial Distribution Under SSP 2.45 and SSP 5.85 Scenarios for the Years 2041-2060 and 2081-2100.
Şekil 7. *Vitis vinifera (Narince Çeşidi) türünün 2041-2060 ve 2081-2100 yılları için SSP 2.45 ve SSP 5.85 Senaryoları Altındaki Tahmin Edilen Mekansal Dağılımı.*

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Short-Term Effect of Different Doses Of Sewage Sludge on Soil Physical and Hydraulic Properties with Different Irrigation Regimes in a Silage Maize Field

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Abstract: Soil physical and hydraulic properties may be affected significantly from stabilized sewage sludge, and irrigation regimes may change the magnitude of these effects. Therefore, we examined the effects of different sewage sludge doses (D0: 0 t/ha, D1: 30 t/ha, D2: 60 t/ha, D3: 90 t/ha) and irrigation regimes (S1, S2, S3) on the soil physical and hydraulic properties with two-year study in a silage maize cultivated soil. The experiment was carried out in a completely random factorial design with three replications. In the S1, S2 and S3 irrigation regimes, when the sum of estimated crop evapotranspiration by FAO-Penman-Monteith approach and effective precipitation difference were 25 mm, 50 mm and 75 mm, respectively, irrigations were carried out and the moisture deficit in the soil was completed to field capacity. While moisture regimes did not have significant effects on soil physical and hydraulic properties, sewage sludge doses resulted in significant effects. Compared to D0, 3.1% lower bulk density, 1.9% lower particle density, 14.9% higher wet aggregate stability and 2.6% higher gravimetric field capacity values were determined at the D3 treatment. It was determined that these improvements were due to the high organic matter content of the sewage sludge, which increased the organic matter content in the soil with increasing doses, and it was also supported by significant linear relationships between the organic matter and these parameters. As a conclusion, although the importance of the positive effects of increasing dose can be emphasized, it can be stated that longer-term studies are needed to see more permanent and effective results.

Keywords: Aggregate stability, bulk and particle densities, municipal sewage sludge, water retention capacity

Silajlık Mısır Tarlasında Farklı Sulama Rejimlerinde Farklı Dozlarda Arıtma Çamurunun Toprağın Fiziksel ve Hidrolik Özelliklerine Kısa Süreçte Etkisi

Öz: Stabilize arıtma çamuru, toprağın fiziksel ve hidrolik özelliklerini önemli ölçüde etkileyebilir ve sulama rejimleri bu etkilerin büyüklüğünü değiştirebilir. Bu nedenle, farklı arıtma çamuru dozlarının (D0: 0 t/ha, D1: 30 t/ha, D2: 60 t/ha, D3: 90 t/ha) ve sulama rejimlerinin (S1, S2, S3) toprak fiziksel ve hidrolik özellikleri üzerindeki etkilerini, silajlık mısır yetiştirilen bir toprakta iki yıllık bir çalışma ile incelenmiştir. Deneme, üç tekrarlamalı tamamen rastgele faktöriyel deneme düzeninde yürütülmüştür. S1, S2 ve S3 sulama rejimlerinde, FAO-Penman-Monteith yaklaşımı ile tahmini ürün evapotranspirasyonunun ve etkili yağış farkının toplamı sırasıyla 25 mm, 50 mm ve 75 mm olduğunda, sulamalar yapılmış ve topraktaki nem açığı tarla kapasitesine tamamlanmıştır. Nem rejimlerinin toprak fiziksel ve hidrolik özellikleri üzerinde önemli etkileri olmazken, arıtma çamuru dozları önemli etkilere yol açmıştır. D0 ile karşılaştırıldığında D3 uygulamasında %3.1 daha düşük kütle yoğunluğu, %1.9 daha düşük tane yoğunluğu, %14.9 daha yüksek ıslak agregat stabilitesi ve %2.6 daha yüksek gravimetrik tarla kapasitesi değerleri belirlenmiştir. Bu iyileşmelerin arıtma çamurunun yüksek organik madde içeriğinden kaynaklandığı, bu nedenle de artan dozlarla topraktaki organik madde içeriğinin arttığı belirlenmiş ve organik madde ile bu parametreler arasında önemli doğrusal ilişkiler bulunması da bunu desteklemiştir. Sonuç olarak, artan dozun olumlu etkilerinin önemi vurgulanabilse de, daha kalıcı ve etkili sonuçlar görmek için daha uzun süreli çalışmalara ihtiyaç olduğu söylenebilir.

Anahtar Kelimeler: Agregat stabilitesi, kütle ve tane yoğunluğu, kentsel arıtma çamuru, su tutma kapasitesi

1. Introduction

With increasing urbanization and industrialization, the number of wastewater treatment plants is increasing rapidly, and this causes an increase in sewage sludge wastes (Nahar & Hossen, 2021). Treatment sludge is a by-product formed as a result of the treatment of industrial, domestic and municipal wastewater, and consists of water, organic matter and biosolids (Yuan & Dai, 2016). These wastes, which should be disposed of without harming the environment, are generally used for fertilizer and soil conditioner in agriculture. The use of controlled and appropriate doses of sewage sludge in agricultural areas is one of the simplest disposal methods (Mondal et al. 2015; Çakır & Çimrin, 2018). Municipal sewage sludge with low heavy metal content that has undergone the stabilization process is preferred mostly in agriculture due to its properties as both an economic fertilizer source and soil improvement.

Organic components in sludge increase the stability of soil aggregates, improve water holding capacity and total soil porosity (Ors et al. 2015; Abdallh & Sahin, 2020; Sahin et al. 2020; Badaou & Sahin, 2022; Yerli et al., 2024). Many studies have shown significant improvements in the physical, hydraulic and chemical properties of soils enriched with sewage sludge, such as aggregate stability, water holding capacity, bulk density, particle density, total porosity, aeration capacity, water permeability, cation exchange capacity and exchangeable Ca content (Mondal et al. 2015; Ors et al. 2015; Cherfouh et al. 2018; Norouzian et al. 2018). Mujdeci et al. (2017) also indicated that organic matter improves the available pore distribution among soil aggregates in favor of improving water retention.

Maize is an important agricultural crop that can be effectively grown on almost every continent in the world, except Antarctica, and can adapt to various climatic conditions (Özkan & Bayhan, 2022). The wide adaptability of this plant makes it an option that can successfully integrate into different climates and soil types. Silage maize is an important food source in the livestock sector worldwide. It plays a critical role in nutrition, especially for dairy cattle. This versatility of maize, together with the advantages it provides for the agricultural sector, contributes to the diversification and sustainability of food production. Maize also need to frequent irrigation higher yield (Cakmakci & Sahin, 2021; Yerli et al., 2023). Fast microbial decomposition of organic matter and microbially derived carbon under frequent irrigation iss also known to promote aggregation (Rabbi et al., 2024; Yerli et al., 2024).

Considering findings previous studies, there is a need examining the short-term effects of physical and hydraulic properties of soil irrigated at changed intervals with irrigation regimes under different sewage sludge dose in silage maize field. In this study, it was aimed to improve soil physical and hydraulic properties with irrigation regime management of different doses of stabilized sewage sludge obtained from domestic waste. In Erzurum ecology in Türkiye, moisture regimes were created at various irrigation levels with real-time water consumption approach and available sewage sludge dose was researched.

2. Material and Methods

2.1. Study area and experimental design

The study was carried out in 2021 (May 7 - September 9) and 2022 (May 13 - September 10) at the experimental field of Atatürk University Plant Production Application and Research Center in Erzurum province, Turkey (39.933° N and 41.236° E, 1780 m a.s.l). The average air temperature and total precipitation during the vegetation period were 18.2 °C and 80.1 mm in the first year, and 17.6 °C and 111.2 mm in the second year, respectively. According to the US Soil Taxonomy, the experimental area soil is aridisol (Soil Survey Staff 1992). Prior to the experiment, the texture of the surface layer (0-30 cm) of the experimental field was clay loam, pH and EC values, and organic matter and lime contents were determined as 7.61, 0.163 dS/m, 1.73% and 0.47%, respectively. Irrigation water was applied using groundwater with an average pH value of 7.43 and an EC value of 0.286 dS/m, with a surface drip irrigation system, one lateral to each plant row.

The experiment was carried out with four different sewage sludge doses (D0: 0 t/ha, D1: 30 t/ha, D2: 60 t/ha, D3: 90 t/ha) and three different irrigation regime applications (S1, S2, S3) in a completely random 4x3 factorial design, with doses as the main plots, with 3 replications in a total of 36 plots. Each plot with an area of 25.2 m^2 was arranged as measured of 3.5 m × 7.2 m in 5 rows. DKC 6777 silage maize variety in FAO 700 group was planted with a pneumatic seeder on the soil processed with a vertical rotovator before sowing, with 70 cm row spacing and 15 cm plant distance on the rows.

2.2. Mixing of sewage sludge into soil and other cultural processes

Stabilized sewage sludge was supplied from the wastewater treatment plant located in Erzurum, Türkiye. pH and EC values and organic matter content, Ca content and dry matter ratio of the stabilized sewage sludge before the experiment were determined as 6.72, 6.51 dS/m, 38.6%, 40.96 cmol/kg, and 29.9% respectively. Heavy metal content in sewage sludge was below the limit values specified in the Regulation on the Use of Domestic and Urban Sewage Sludge in Soil in Türkiye (Official Gazette, 2010).

Stabilized sewage sludge was brought to the experimental area in the autumn period (end of September 2020) before the first planting year, spread homogeneously on the surface of the plowed plots and was mixed to a depth of 15 cm with a hoe machine. No additional chemical fertilizer was applied to the plots where sewage sludge was applied in either year. However, considering the results of the fertility analysis in the 0-30 cm soil layer in the D0 plots without sewage sludge, the deficient nitrogen and phosphorus fertilizer amounts were determined, and the deficient amounts were applied to the plots manually according to the doses of 100 kg/ha urea (% 45-46 N) and 150 kg/ha triple super phosphate (% 43-46 P_2O_5). All of the phosphorus and half of the nitrogen were applied immediately after planting. The remaining half of the nitrogen was given when the plants reached 40-50 cm in height (with 4-6 leaves). The selection of current fertilizer doses and the applications were made by considering previous studies on silage maize in this and similar regions (Ors et al., 2015; Cakmakci & Sahin, 2021; Yerli et al., 2023). The first and second hoeing for weed control was done when the plants reached 15-20 cm and 40-50 cm height, respectively.

2.3. Determination of irrigation time and amount

When the total of the difference between estimated crop evapotranspiration (ETc) and effective rainfall (Peff) in S1, S2 and S3 irrigation regimes [$\sum$ (ETc – Peff)] was 25 mm, 50 mm and 75 mm, respectively, irrigations were applied and the moisture deficit according to the field capacity in the soil was completed to the field capacity. In 2021, a total of 310.2 mm, 293.2 mm and 277.9 mm of irrigation water was applied in the S1, S2 and S3, respectively. In 2022, a total of 336.8 mm, 323.9 mm and 297.4 mm of irrigation water was applied. The applied water quantities were calculated according to wetting rates of 0.30 and 0.65, respectively, considering 30 cm soil depth until the 4-6 leaf stage of the plants and then 90 cm soil depth. While the moisture at 30 cm was measured with a field-calibrated moisture meter (TDR, Trime-Pico, IPH/T3, IMKO), and the moisture in the lower layers was measured gravimetrically.

The ETc value was calculated with the following Equation.!

$$ETc = ETo \times kc \quad (1)$$

"kc" values were obtained from the Evapotranspiration Guide for Irrigated Crops in Türkiye (TAGEM, 2017). Reference evapotranspiration (ETo) values were also calculated with the Penman-Monteith (FAO) approach using the CROPWAT program. The climate data required for the calculations were obtained from the Erzurum Airport Meteorological Station near to the experimental area. Rainfall data were also obtained from the pluviometer located in the experimental area since all the precipitation stored in the effective root zone of silage maize, all of it is considered effective precipitation (Peff).

2.4. Determination of soil physical and hydraulic properties

After harvest in the experimental years, disturbed and undisturbed soil samples were taken from 0-30 cm depth from all plots. Particle density was determined by Pycnometer method (Blake & Hartge, 1986a). Bulk density was calculated by dividing the oven dry weights of samples taken with undisturbed soil sampling cylinder by the total sample volume (Blake & Hartge, 1986b). Porosity was calculated using particle and bulk density values (Danielson & Sutherland, 1986). Wet aggregate stability was determined by Yoder type wet sieving device (Kemper & Rosenau, 1986). The amount of water retained at field capacity (- 0.033 MPa) and permanent wilting point (-1.5 MPa) were determined by using ceramic plates in a pressure chamber. (Klute, 1986). Available water content was calculated from the difference between field capacity and permanent wilting point (Cassel & Nielsen, 1986). Soil organic matter content was determined by the Smith-Weldon method (Nelson & Sommers, 1982). Exchangeable Ca was measured by ICP-MS using samples subjected to high pressure wet digestion process (U.S. EPA, 2007).

The initial bulk density, particle density, porosity, wet aggregate stability, field capacity, permanent wilting point and available water content were 1.29 g/cm^3, 2.66, 51.9%, 46.8%, 23.2% of weight, 13.3% of weight, and 38.3 mm, respectively. Exchangeable Ca content was 15.84 cmol/kg, and organic matter content was 1.73%.

2.5. Statistical Analysis

The experimental data were analyzed with General Linear Model approach in the SPSS statistics program. Considering significant effects of sewage sludge doses, irrigation regimes and their interactions, the means at the $p < 0.05$ significance level were classified using the multiple comparison test. In addition, Pearson correlation analyses were applied to determine the binary relationships between some parameters.

3. Results and Discussion

3.1. Soil physical properties

Sewage sludge doses significantly ($p<0.01$) increased bulk and particle densities and wet aggregate stability values in both year and average of years, significant ($p < 0.01$) effect on porosity was determined in the first year only (Table 1). Bulk density decreased with increasing dose, and 3.1% lower bulk density was determined in the D3 treatment compared to the D0 treatment (Table 2). With the increase in the sewage sludge dose, the particle density also decreased and a 1.9% lower value was determined in the D3 dose compared to D0. In porosity, the D3 treatment provided a 0.8% higher value compared to D0 in the first trial year. The aggregate stability value of the D3 treatment was also 14.9% higher compared to D0.

Table 1. Variance analysis results
Çizelge 1. Varyans analizi sonuçları

Parameter	Year	Variance Sources	df	Mean Square	F	P
Bulk density	2021	Dose	3	0.002	18.758	0.000
		Irrigation	2	0.000	0.175	0.841
		Dose × Irrigation	6	7.870E-05	0.708	0.646
		Error	24	0.000E+00		
	2022	Dose	3	0.002	20.444	0.000
		Irrigation	2	0.000	0.576	0.570
		Dose × Irrigation	6	4.907E-05	0.535	0.776
		Error	24	9.167E-05		
	2021-2022	Dose	3	0.002	37.526	0.000
		Irrigation	2	0.000	0.684	0.514
		Dose × Irrigation	6	3.611E-05	0.684	0.664
		Error	24	5.278E-05		
Particle density	2021	Dose	3	0.004	57.538	0.000
		Irrigation	2	0.000	2.423	0.110
		Dose × Irrigation	6	1.417E-04	1.962	0.112
		Error	24	0.000		
	2022	Dose	3	0.004	53.544	0.000
		Irrigation	2	0.000	1.900	0.171
		Dose × Irrigation	6	9.537E-05	1.144	0.368
		Error	24	0.000		
	2021-2022	Dose	3	0.004	56.718	0.000
		Irrigation	2	0.000	1.654	0.212
		Dose × Irrigation	6	1.157E-04	1.603	0.190
		Error	24	0.000		
Porosity	2021	Dose	3	0.953	6.141	0.003
		Irrigation	2	0.230	1.480	0.248
		Dose × Irrigation	6	2.900E-01	1.869	0.128
		Error	24	1.550E-01		
	2022	Dose	3	0.350	1.801	0.174
		Irrigation	2	0.233	1.201	0.318
		Dose × Irrigation	6	1.260E-01	0.650	0.690
		Error	24	1.940E-01		
	2021-2022	Dose	3	0.350	1.801	0.174
		Irrigation	2	0.233	1.201	0.318
		Dose × Irrigation	6	1.260E-01	0.650	0.690
		Error	24	1.940E-01		
Wet aggregate stability	2021	Dose	3	93.330	662.696	0.000
		Irrigation	2	0.054	0.384	0.685
		Dose × Irrigation	6	1.800E-02	0.128	0.992
		Error	24	1.410E-01		
	2022	Dose	3	89.472	496.701	0.000
		Irrigation	2	0.196	1.088	0.353
		Dose × Irrigation	6	6.300E-02	0.348	0.904
		Error	24	1.800E-01		
	2021-2022	Dose	3	91.400	1445.252	0.000
		Irrigation	2	0.067	1.064	0.361
		Dose × Irrigation	6	1.300E-02	0.212	0.969
		Error	24	6.300E-02		
Organic matter	2021	Dose	3	0.074	956.131	0.000
		Irrigation	2	0.006	78.429	0.000
		Dose × Irrigation	6	0.000E+00	3.667	0.010
		Error	24	7.778E-05		
	2022	Dose	3	0.148	321.789	0.000
		Irrigation	2	0.002	4.934	0.016
		Dose × Irrigation	6	0.000E+00	0.331	.914
		Error	24	4.611E-04		
	2021-2022	Dose	3	0.105	665.889	0.000
		Irrigation	2	0.004	23.211	0.000
		Dose × Irrigation	6	0.000E+00	0.871	0.530
		Error	24	1.583E-04		

Table 1. (continued)
Çizelge 1. (devam)

Parameter	Year	Variance Sources	df	Mean Square	F	P
Exchangeable Ca	2021	Dose	3	5.451	36.986	0.000
		Irrigation	2	0.027	0.184	0.833
		Dose × Irrigation	6	0.087	0.592	0.734
		Error	24	0.147		
	2022	Dose	3	1.832	11.902	0.000
		Irrigation	2	0.087	0.565	0.576
		Dose × Irrigation	6	0.053	0.344	0.906
		Error	24	0.154		
	2021-2022	Dose	3	3.324	45.690	0.000
		Irrigation	2	0.052	0.714	0.500
		Dose × Irrigation	6	0.027	0.371	0.890
		Error	24	0.073		
Field capacity	2021	Dose	3	0.697	2.879	0.057
		Irrigation	2	0.008	0.032	0.968
		Dose × Irrigation	6	2.400E-02	0.098	0.996
		Error	24	2.420E-01		
	2022	Dose	3	0.782	6.821	0.002
		Irrigation	2	0.003	0.029	0.971
		Dose × Irrigation	6	3.300E-02	0.291	0.936
		Error	24	1.150E-01		
	2021-2022	Dose	3	0.748	5.894	0.004
		Irrigation	2	0.005	0.042	0.959
		Dose × Irrigation	6	2.900E-02	0.228	0.963
		Error	24	1.270E-01		
Permanent wilting point	2021	Dose	3	0.063	0.453	0.717
		Irrigation	2	0.004	0.026	0.974
		Dose × Irrigation	6	2.000E-03	0.013	1.000
		Error	24	1.380E-01		
	2022	Dose	3	0.041	1.086	0.374
		Irrigation	2	0.008	0.207	0.814
		Dose × Irrigation	6	2.300E-02	0.612	0.718
		Error	24	3.800E-02		
	2021-2022	Dose	3	0.045	0.772	0.521
		Irrigation	2	0.002	0.033	0.968
		Dose × Irrigation	6	9.000E-03	0.159	0.985
		Error	24	5.900E-02		
Available water content	2021	Dose	3	1.055	0.132	0.940
		Irrigation	2	0.385	0.048	0.953
		Dose × Irrigation	6	3.190E-01	0.040	1.000
		Error	24	7.984E+00		
	2022	Dose	3	2.459	1.409	0.265
		Irrigation	2	0.028	0.016	0.984
		Dose × Irrigation	6	8.400E-01	0.481	0.816
		Error	24	1.745E+00		
	2021-2022	Dose	3	1.649	0.578	0.635
		Irrigation	2	0.974	0.341	0.714
		Dose × Irrigation	6	3.056E+00	1.070	0.407
		Error	24	2.855E+00		

*$p < 0.05$

Bulk density decreased with increasing dose, and 3.1% lower bulk density was determined in the D3 treatment compared to the D0 treatment (Table 2). With the increase in the sewage sludge dose, the particle density also decreased and a 1.9% lower value was determined in the D3 dose compared to D0. In porosity, the D3 treatment provided a 0.8% higher value compared to D0 in the first trial year. The aggregate stability value of the D3 treatment was also 14.9% higher compared to D0.

Considering the initial values of particle density as 2.66, bulk density as 1.29 g/cm^3, porosity as 51.9% and wet aggregate stability as 46.8%, at the end of the second trial year, D3 treatment decreased bulk density by 4.7% and particle density by 1.5%, while increasing porosity by 1.9% and wet aggregate stability by 22.2%.

Table 2. Bulk density, particle density, porosity and wet aggregate stability values in 0-30 cm soil layer at different sewage sludge dose and irrigation regimes

Çizelge 2. Farklı arıtma çamuru dozları ve sulama rejimlerinde 0-30 cm toprak tabakasında kütle yoğunluğu, tane yoğunluğu, porozite ve ıslak agrega stabilitesi değerleri

Parameter	Year	Irrigation treatment	D0	D1	D2	D3	Mean
Bulk density (g/cm^3)	2021	S1	1.27±0.01	1.26±0.00	1.25±0.01	1.23±0.01	1.25±0.01
		S2	1.27±0.01	1.26±0.00	1.25±0.00	1.24±0.01	1.26±0.00
		S3	1.26±0.01	1.26±0.01	1.26±0.01	1.23±0.01	1.25±0.00
		Mean	1.27±0.00 A*	1.26±0.00 AB	1.25±0.00 B	1.23±0.00 C	
	2022	S1	1.27±0.01	1.26±0.00	1.24±0.00	1.23±0.00	1.25±0.00
		S2	1.27±0.00	1.26±0.00	1.25±0.01	1.23±0.01	1.25±0.00
		S3	1.26±0.01	1.27±0.00	1.25±0.01	1.24±0.01	1.25±0.00
		Mean	1.27±0.00 A*	1.26±0.00 A	1.25±0.00 B	1.23±0.00 C	
	2021-2022	S1	1.27±0.00	1.26±0.00	1.25±0.00	1.23±0.00	1.25±0.00
		S2	1.27±0.01	1.26±0.00	1.25±0.00	1.23±0.00	1.25±0.00
		S3	1.26±0.00	1.27±0.00	1.25±0.01	1.24±0.01	1.25±0.00
		Mean	1.27±0.00 A*	1.26±0.00 A	1.25±0.00 B	1.23±0.00 C	
Particle density	2021	S1	2.67±0.01	2.66±0.01	2.64±0.00	2.63±0.00	2.65±0.01 A*
		S2	2.67±0.01	2.64±0.00	2.63±0.00	2.62±0.00	2.64±0.01 B
		S3	2.68±0.00	2.64±0.00	2.63±0.00	2.62±0.00	2.64±0.01 B
		Mean	2.67±0.00 A*	2.65±0.00 B	2.63±0.00 C	2.62±0.00 D	
	2022	S1	2.68±0.01	2.66±0.00	2.65±0.00	2.62±0.01	2.65±0.01
		S2	2.67±0.01	2.65±0.00	2.64±0.00	2.62±0.01	2.65±0.01
		S3	2.68±0.00	2.65±0.00	2.63±0.00	2.63±0.01	2.65±0.01
		Mean	2.67±0.00 A*	2.65±0.00 B	2.64±0.00 C	2.62±0.00 D	
	2021-2022	S1	2.67±0.01	2.66±0.01	2.64±0.00	2.62±0.00	2.65±0.01
		S2	2.67±0.00	2.65±0.00	2.64±0.00	2.62±0.00	2.64±0.01
		S3	2.68±0.00	2.64±0.00	2.63±0.00	2.63±0.01	2.64±0.01
		Mean	2.67±0.00 A*	2.65±0.00 B	2.64±0.00 C	2.62±0.00 D	
Porosity (%)	2021	S1	52.4±0.12	52.5±0.24	52.8±0.26	53.2±0.21	52.7±0.13
		S2	52.6±0.24	52.1±0.20	52.3±0.06	52.7±0.32	52.4±0.12
		S3	53.0±0.28	52.1±0.28	52.0±0.17	53.2±0.25	52.6±0.18
		Mean	52.6±0.14 AB	52.3±0.13 B	52.4±0.14 B	53.0±0.15 A*	
	2022	S1	52.7±0.43	52.8±0.11	53.0±0.19	52.9±0.16	52.9±0.11
		S2	52.6±0.22	52.4±0.07	52.6±0.17	53.1±0.29	52.6±0.111
		S3	52.8±0.32	52.1±0.16	52.7±0.29	52.8±0.38	52.6±0.15
		Mean	52.7±0.17	52.4±0.12	52.8±0.13	52.9±0.15	
	2021-2022	S1	52.5±0.17	52.7±0.15	52.9±0.16	53.1±0.11	52.8±0.09
		S2	52.6±0.23	52.3±0.10	52.5±0.05	52.9±0.05	52.5±0.09
		S3	52.9±0.17	52.1±0.16	52.3±0.29	53.0±0.29	52.6±0.14
		Mean	52.7±0.11	52.4±0.11	52.6±0.10	53.0±0.10	
Wet aggregate stability (%)	2021	S1	49.7±0.24	52.0±0.07	54.5±0.29	57.2±0.30	53.4±0.84
		S2	49.7±0.21	51.9±0.14	54.4±0.27	57.0±0.16	53.2±0.83
		S3	49.6±0.27	51.7±0.09	54.5±0.21	57.2±0.21	53.3±0.86
		Mean	49.7±0.12 D	51.9±0.06 C	54.5±0.13 B	57.1±0.12 A*	
	2022	S1	50.0±0.59	52.0±0.08	54.6±0.17	57.0±0.04	53.4±0.80
		S2	49.7±0.37	51.9±0.10	54.4±0.22	57.3±0.21	53.3±0.86
		S3	50.0±0.28	52.2±0.10	54.6±0.09	57.4±0.07	53.5±0.83
		Mean	49.9±0.22 D	52.0±0.07 C	54.5±0.09 B	57.2±0.09 A*	
	2021-2022	S1	49.9±0.28	52.0±0.03	54.5±0.16	57.1±0.15	53.4±0.82
		S2	49.7±0.08	51.9±0.05	54.4±0.13	57.1±0.10	53.3±0.84
		S3	49.8±0.24	52.0±0.06	54.6±0.13	57.3±0.11	53.4±0.84
		Mean	49.8±0.11 D	51.9±0.03 C	54.5±0.07 B	57.2±0.07 A*	

Due to the high organic matter content of the sewage sludge of 38.6%, the organic matter content of the soil increased with the increase in the applied dose, and the highest values were determined in the D3 treatment, and infrequent irrigation regime, which reduced organic matter mineralization (Figure 1). Organic matter in the soil supports microorganism activities and strengthens the bonds between soil mineral materials, thus ensuring that aggregates are more durable and stable (Lu et al., 2021). Therefore, mostly, the presence of organic matter increases aggregate stability and decreases bulk density (Ramezani et al., 2019). Similarly, Asadu et al. (2008), Delibacak et al. (2020) and Aksakal and Cambaztepe (2022) reported that organic matter, bulk density, porosity and aggregate stability in the sewage-amended soil were improved significantly compared to the without sewage soil. Although Sharma (2024) reported that porosity increases as the organic matter content of the soil increases, as parallel to our porosity results, Sort and Alcañiz (1999) reported that although sludge application caused an increase in both fine micro and coarse porosity of the soil, this effect was transient as no significant difference was detected compared to the control one year after application. Simões-Mota et al. (2022) also reported the significant effect on porosity of long-term sewage sludge application. However, Camps-Sagué et al. (2024) indicated that the potential improvement benefits on soil structure of sewage sludge are limited to their use within a certain dose range, and reported no significant differences between sewage sludge application of twenty years and mineral treatment for total porosity were found.

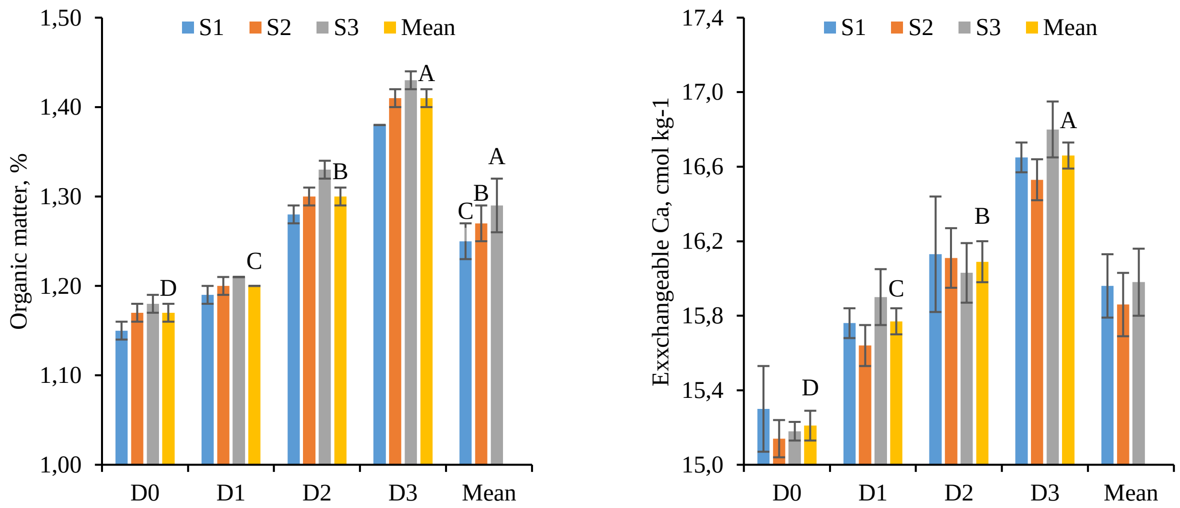

Fig 1. Experimental years averages of organic matter and exchangeable Ca contents in 0-30 cm soil layer under different irrigation regimes with different sewage sludge doses. D0: 0 t/ha, D1: 30 t/ha, D2: 60 t/ha, D3: 90 t/ha. S1, S2, and S3: irrigation when sum of the difference between estimated evapotranspiration and effective precipitation equals to 25 mm, 50 mm and 75 mm, respectively. *: $p < 0.05$

Şekil 1. Farklı sulama rejimleri ve farklı arıtma çamuru dozları altında 0-30 cm toprak katmanındaki organik madde ve değiştirilebilir Ca içeriklerinin deneysel yıl ortalamaları. D0: 0 t/ha, D1: 30 t/ha, D2: 60 t/ha, D3: 90 t/ha. S1, S2 ve S3: tahmini buharlaşma ve etkili yağış arasındaki farkın toplamı sırasıyla 25 mm, 50 mm ve 75 mm'ye eşit olduğunda sulama. *: $p < 0.05$

The binary relationships of particle density, bulk density and wet aggregate stability with the soil organic matter content were significantly ($p < 0.01$) linear (Figure 2). Similarly, Yerli et al. (2024) indicated that while the decrease in bulk density and increases in wet aggregate stability was mostly related to soil organic matter content, organic matter content did not reach a level that affected particle density significantly.

Biswas et al. (2017) indicated that the increase in the organic matter content of the soil leads to a decrease in bulk density. Usman et al. (2012) also stated that as the sewage sludge ratio increases, the bulk density decreases due to increased porosity. Sabtow and Kızıloğlu (2022) reported that the particle density decreases with the increase in dose in soils where organic materials are applied. (Ojeda et al. (2008) reported that aggregate stability generally has a good correlation with soil organic matter content, soil has a higher organic carbon content and greater aggregate stability than the control two years after the surface application of the sludge. Gülser and Candemir (2015) determined significant positive correlations between organic matter and aggregate stability in a soil treated with agricultural wastes. While the many other studies also indicated that the increase in organic matter content significantly increases aggregate stability (Dong et al., 2023; Sun et al., 2023; Halder et al., 2024), many studies also reported a positive relationship between organic matter content and aggregate stability (Sarker et al., 2018; Mbanjwa et al., 2022; Sonsri et al., 2023).

Exchangeable Ca contents in soil in D0 and D1 treatments were lower than the initial value of soil (15.84 cmol/kg) based on probably crop consumption (Figure 1). However, exchangeable Ca contents in soil increased with increasing dose due to high Ca content of sewage sludge (40.96 cmol/kg) in D2 and D3 treatments compared to the initial value of soil. The effect of Ca on aggregate stability is related to the ability of soil colloids to neutralize surface charges. Calcium interacts with negatively charged soil colloids, neutralizes surface charges and increases aggregate stability by allowing soil particles to adhere better to each other. Therefore, it is known that soils with sufficient Ca content generally have higher aggregate stability. Researches show that there is a positive relationship between soil aggregate stability and exchangeable Ca content. As exchangeable Ca content increases, aggregate stability also increases; this relationship is explained by Ca acting as a bridge between clay particles and supporting aggregate formation (Gümüş et al., 2016). These experiment results stated a significant ($p < 0.01$) positive relationship between soil aggregate stability and exchangeable Ca content (Figure 2). Similarly, Bedel et al. (2018) and Yao et al. (2022) also determined a positive relationship between aggregate stability and exchangeable Ca content in their studies.

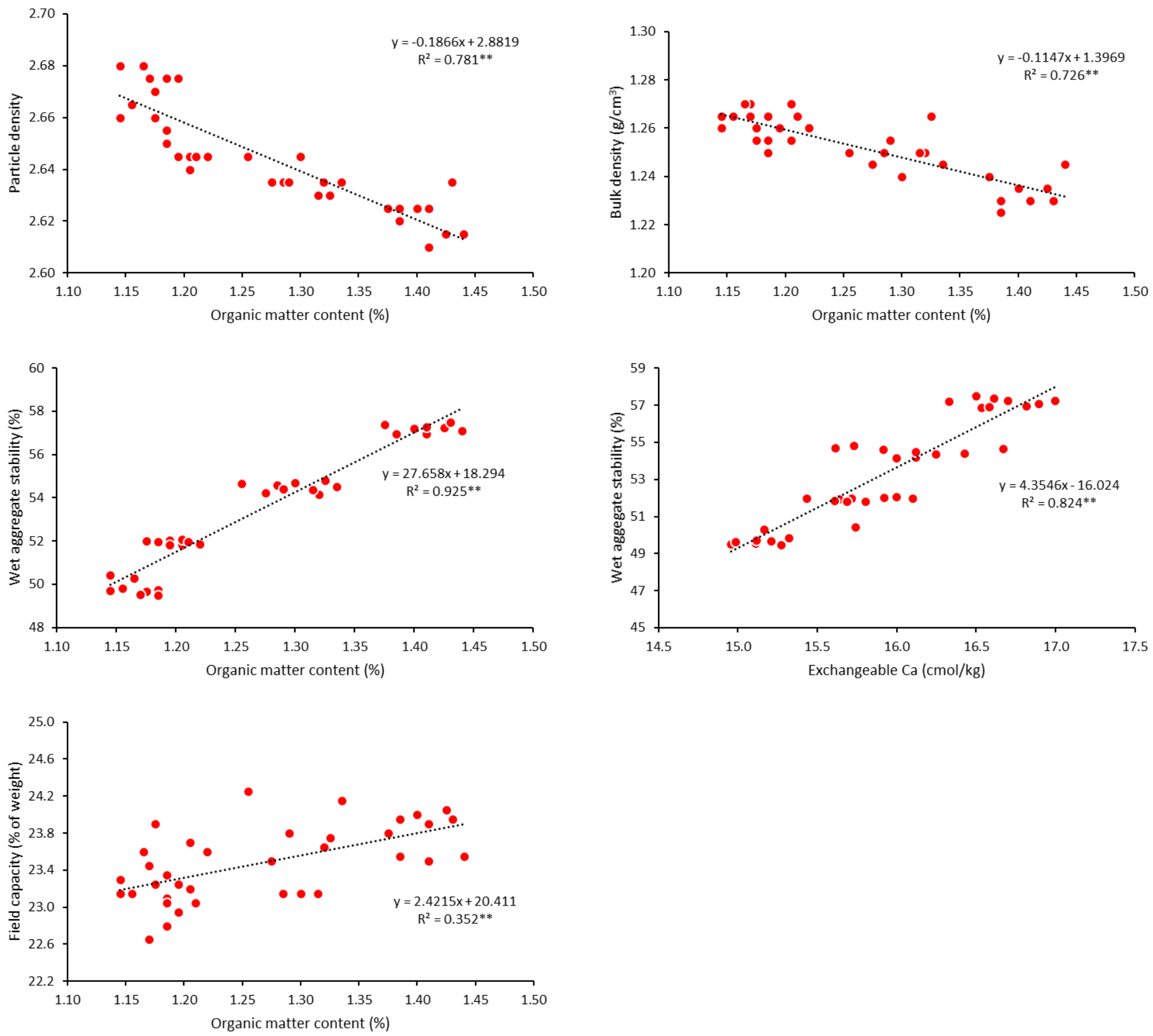

Fig 2. The binary relationships between some soil properties (n=36; **$p < 0.01$)
Şekil 2. Bazı toprak özellikleri arasındaki ikili ilişkiler (n=36; **$p < 0.01$)

3.2. Soil hydraulic properties

The effect of sewage sludge application on field capacity was significant ($p < 0.01$; $p < 0.05$ in the first year) (Table 1). In the two-year average, when compared to D0, gravimetric field capacity values increased by 0.85%, 2.1% and 2.6% in D1, D2 and D3 treatments, respectively (Table 3). Moreover, D3 treatment provided a 2.6% improvement compared to the initial value (23.2% of weight) in the experiment. Sewage sludge, with its high organic matter content, enriches the soil in terms of organic matter and increases its water holding capacity at low tension. Studies show that soils with high organic matter content have higher field capacities. In this study, a significant ($p < 0.01$) positive relationship was found between gravimetric field capacity and organic matter content (Figure 2). Lal (2020) and Ramirez et al. (2023) also stated that the increase in organic matter increases field capacity and that there is a linear relationship between these two parameters. Many other studies have also shown that the increase in organic matter in the soil causes a significant increase in field capacity values (Tunc & Sahin, 2015; Kadıoğlu & Canbolat, 2019; Alaboz & Çakmakcı, 2020; Yerli et al., 2024).

Table 3. Field capacity, permanent wilting point and available water content values in 0-30 cm soil layer at different sewage sludge dose and irrigation regimes

Çizelge 3. Farklı arıtma çamuru dozları ve sulama rejimlerinde 0-30 cm toprak tabakasında tarla kapasitesi, devamlı solma noktası ve kullanılabilir su içeriği değerleri

Parameter	Year	Irrigation treatment	D0	D1	D2	D3	Mean
Field capacity (% of weight)	2021	S1	23.1±0.47	23.5±0.35	23.6±0.32	23.8±0.15	23.5±0.17
		S2	23.4±0.17	23.4±0.26	23.6±0.20	23.8±0.23	23.6±0.11
		S3	23.2±0.35	23.3±0.26	23.7±0.34	23.9±0.09	23.5±0.14
		Mean	23.2±0.18 C	23.4±0.15 AB	23.7±0.15 AB	23.8±0.08 A*	
	2022	S1	23.0±0.09	23.3±0.19	23.6±0.34	23.7±0.21	23.4±0.13
		S2	23.2±0.06	23.3±0.17	23.4±0.22	23.8±0.12	23.4±0.13
		S3	23.1±0.15	23.2±0.09	23.6±0.32	23.7±0.18	23.4±0.12
		Mean	23.1±0.06 C	23.3±0.08 BC	23.6±0.15 AB	23.8±0.09 A*	
	2021-2022	S1	23.0±0.20	23.4±0.26	23.6±0.32	23.8±0.12	23.5±0.13
		S2	23.3±0.09	23.3±0.22	23.5±0.20	23.9±0.18	23.5±0.10
		S3	23.2±0.23	23.3±0.16	23.7±0.29	23.8±0.13	23.5±0.12
		Mean	23.2±0.10 C	23.3±0.11 BC	23.6±0.14 AB	23.8±0.07 A*	
Permanent wilting point (% of weight)	2021	S1	13.2±0.03	13.4±0.20	13.4±0.14	13.5±0.02	13.4±0.06
		S2	13.3±0.20	13.3±0.18	13.4±0.05	13.5±0.26	13.4±0.08
		S3	13.3±0.20	13.3±0.17	13.4±0.28	13.5±0.47	13.4±0.13
		Mean	13.3±0.08	13.3±0.09	13.4±0.09	13.5±0.16	
	2022	S1	13.3±0.03	13.4±0.06	13.2±0.19	13.6±0.09	13.4±0.06
		S2	13.4±0.09	13.4±0.12	13.5±0.07	13.4±0.12	13.4±0.04
		S3	13.4±0.12	13.3±0.09	13.5±0.19	13.5±0.09	13.4±0.06
		Mean	13.4±0.04	13.4±0.05	13.4±0.09	13.5±0.05	
	2021-2022	S1	13.3±0.00	13.4±0.09	13.3±0.16	13.5±0.05	13.4±0.05
		S2	13.3±0.11	13.4±0.13	13.4±0.04	13.5±0.17	13.4±0.06
		S3	13.3±0.16	13.3±0.11	13.5±0.23	13.5±0.24	13.4±0.09
		Mean	13.3±0.06	13.4±0.06	13.4±0.08	13.5±0.09	
Available water content (mm)	2021	S1	37.2±1.99	38.3±2.04	38.1±1.16	38.0±0.50	37.9±0.68
		S2	38.2±1.57	38.1±1.47	38.4±0.95	38.5±2.06	38.3±0.67
		S3	37.6±1.99	38.0±0.62	38.9±2.17	38.3±1.84	38.2±0.77
		Mean	37.7±0.94	38.2±0.75	38.5±0.77	38.3±0.81	
	2022	S1	36.6±0.54	37.5±0.77	38.8±0.65	37.5±0.85	37.6±0.38
		S2	37.4±0.46	37.5±0.65	37.4±0.74	38.4±0.67	37.7±0.30
		S3	36.8±0.85	37.6±0.14	38.0±1.57	37.9±0.40	37.6±0.42
		Mean	36.9±0.34	37.5±0.29	38.1±0.57	37.9±0.36	
	2021-2022	S1	36.9±0.73	37.9±1.40	38.5±0.91	37.8±0.55	37.8±0.44
		S2	37.8±0.82	37.8±1.04	37.9±0.85	38.4±1.32	38.0±0.45
		S3	37.2±1.34	37.8±0.36	38.4±1.75	38.1±1.12	37.9±0.55
		Mean	37.3±0.52	37.9±0.52	38.3±0.63	38.1±0.53	

*$p < 0.05$

Although a statistical change was not observed in the permanent wilting point and available water holding capacity, it was determined that there was a 1.5% improvement in the permanent wilting point and a 2.1% improvement in the available water content compared to D0 at the highest dose with increasing dose in the two-year average. Similarly, Gardner et al. (2010) indicated that although biosolids added to the soil increased gravimetric water retention at field capacity and wilting point, no significant change occurred in gravimetric water retention because of a proportional increase in both field capacity and wilting point values.

Considering the significant increase in field capacity due to organic matter in this study, it was evaluated that the partial increase in the amount of available water was also due to field capacity. Yerli et al. (2024) also stated that there was an increase in field capacity, wilting point and available water content with the increase in the amount of organic matter in the surface soil. Tunc and Sahin (2015) and Dogan Demir and Sahin (2019) stated that increases in the amount of available water have a strong relationship with porosity from soil properties. Considering the views of Ors et al. (2015) that water retention capacity is directly related to pore sizes and that organic material contribute to water retention capacity by improving the pore size distribution, it can be said that the change in pore size distribution may be more important in improving water retention at low tensions, since no significant increases in porosity were detected in this study.

4. Conclusions

This study aim was to examine the effects of four different sewage sludge doses and three irrigation regimes on the soil physical and hydraulic properties. As a result of the study, it was concluded that 90 t/ha dose of stabilized sewage sludge, with non-significant effect of irrigation regimes, can be good practice and contribute to the physical and hydraulic properties of the soil in silage maize cultivation. However, it has been evaluated that the contribution of irrigation at wide intervals to the preservation of organic matter in the soil should also considered, and that this may be important in terms of the effect of doses over 90 t/ha of sewage sludge on soil properties. Therefore, it could be concluded that, further investigation of the long-term effects of sewage sludge with higher doses also on soil physical and hydraulic properties in agricultural areas is necessary to verify the permanence of short-term results. This will be particularly useful in clarifying the effects of increased salinity in soil from sewage sludge on physical and hydraulic soil properties in the long term.

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Developing Leaf Area Prediction Model for Curly Lettuce Grown Under Salinity Stress and Applied with Foliar Salicylic Acid

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Abstract: Accurate and non-destructive methods for measuring leaf area are crucial for understanding the growth and physiological variations of plants under stress conditions. This investigation aimed to develop and assess the effectiveness of various regression models for predicting the leaf area of curly lettuce cultivated under different irrigation water salinities (IWS: 0.30, 4.15, 8.0 dS m^{-1}) and salicylic acid doses (SA: 0, 1, 2 mM). The coefficient of determination (R^2) values for the models ranged from 0.505 to 0.968, with Root Mean Square Error (RMSE) values between 4.59 and 17.79 cm^2 and Mean Absolute Error (MAE) values of 3.44 to 13.05 cm^2. Using only leaf length (LL) and leaf width (LW) can effectively estimate the leaf area of curly lettuce plants (Model 3, R^2: 0.962, RMSE: 7.58 cm^2, MAE: 5.34 cm^2). Incorporating IWS and SA into prediction models enhanced their accuracy and reliability. The best model for estimating the leaf area of curly lettuce was found from Model 13, which integrated all four parameters—SA, IWS, LL, and LW—achieving the highest R^2 (0.968) and the lowest RMSE (4.59 cm^2) and MAE (3.44 cm^2). Finally, using leaf area prediction models that consider stress conditions can enhance crop management by allowing accurate monitoring of plant health and growth in agriculture.

Keywords: Lactuca sativa, leaf dimensions, non-destructive methods, precision agriculture, regression models.

Tuzluluk Stresi Koşullarında Yetiştirilen ve Yapraktan Salisilik Asit Uygulanan Kıvırcık Marul İçin Yaprak Alanı Tahmin Modelinin Geliştirilmesi

Öz: Stres koşulları altında bitkilerin büyümesini ve fizyolojik değişimlerini anlamak için yaprak alanının doğru ve bitkiye zarar vermeyen yöntemlerle ölçülmesi büyük önem taşımaktadır. Bu çalışmada farklı sulama suyu tuzlulukları (IWS: 0.30, 4.15, 8.0 dS m^{-1}) ve salisilik asit dozları (SA: 0, 1, 2 mM) altında yetiştirilen kıvırcık marulun yaprak alanını tahmin etmek için çeşitli regresyon modellerinin geliştirilmesi ve etkinliğinin değerlendirilmesini amaçlanmıştır. Modeller için R^2 değerleri 0.505 ile 0.968 arasında, RMSE değerleri 4.59 ile 17.79 cm^2 ve MAE değerleri 3.44 ile 13.05 cm^2 arasında bulunmuştur. Sadece yaprak uzunluğu (LL) ve yaprak genişliği (LW) kullanılarak kıvırcık marul bitkilerinin yaprak alanı etkili bir şekilde tahmin edilebileceği anlaşılmıştır (Model 3, R^2: 0.962, RMSE: 7.58 cm^2, MAE: 5.34 cm^2). IWS ve SA' nın tahmin modellerine dahil edilmesi elde edilen regresyon eşitliklerinin doğruluk ve güvenilirliklerini artırmıştır. Kıvırcık marulun yaprak alanını tahmin etmek için en iyi model, en yüksek R^2 (0.968) ve en düşük RMSE (4.59 cm^2) ve MAE (3.44 cm^2) değerlerinin elde edildiği dört parametreyi (SA, IWS, LL ve LW) entegre eden Model 13 olduğu belirlenmiştir. Sonuç olarak, stres koşullarını dikkate alan yaprak alanı tahmin modellerinin kullanılması, tarımda bitki sağlığı ve büyümesinin doğru bir şekilde izlenmesine olanak sağlayarak ürün yönetimini iyileştirebilir.

Anahtar Kelimeler: Lactuca sativa, yaprak boyutları, tahribatsız yöntemler, hassas tarım, regresyon modelleri.

1. Introduction

Leaf area (LA) plays a crucial role in studies related to plant growth and physiology, aiding researchers in comprehending the intricate relationships between plants and their surroundings (Rahimikhoob et al., 2023). It offers valuable information on processes such as photosynthesis functions, stomatal behavior, and the distribution of nutrients within leaves (Huang et al., 2022). The size of the leaf is directly connected to a plant's capacity to absorb solar energy, produce energy, and facilitate essential photosynthetic processes crucial for its growth. (Tanaka et al., 2022; Ribeiro et al., 2024). Furthermore, analyzing LA can reveal how plants adapt to various environmental stressors, including water-salt stress, light, and diagnosing nutrient deficiencies (Soheili et al., 2023; Kiremit et al., 2024).

Leaf area can be measured using destructive or non-destructive, direct or indirect methods (Patrício & Rieder, 2018). Destructive methods, which require removing leaves for measurement, can impact plant health and quality even though they yield accurate data (Pandey & Singh, 2011). Non-destructive methods, such as laser scanning and digital imaging, allow for precise LA assessment without damaging the plant throughout its life cycle (Ribeiro et al., 2024; Tunca et al., 2024). Indirect methods, which estimate LA based on dimensions like length and width, are cost-effective and simplify the measurement process. The key difference between direct and indirect methods is that direct methods measure LA outright, while indirect methods rely on observable parameters (Ribeiro et al., 2022).

Regression models for predicting LA offer a practical, non-invasive measurement approach (Cemek et al., 2020; Ribeiro et al., 2022). These models relate measurable parameters (like leaf length and width) to actual LA, reducing the need for invasive techniques and minimizing harm to plants (Pandey & Singh, 2011; Ribeiro et al., 2024). Nevertheless, the performance of these models was significantly affected by plant variety, stress conditions, and data handling (Amorim et al., 2024). Therefore, it is essential to carefully choose and rigorously validate models to guarantee that the forecasts regarding LA are not only trustworthy but also meaningful. This rigorous process is vital for achieving accurate results that can be trusted, as the quality of the models used directly impacts the validity of the predictions made regarding LA. With this perspective, many previous researchers have developed various empirical models to predict LA for different plants, including green pepper (Cemek et al., 2011), bell pepper (Cemek et al., 2020), basil cultivars (Ribeiro et al., 2022), chokeberry (Akyüz & Cemek, 2024), lettuce (Rahimikhoob et al., 2023), sweet potato (Ribeiro et al., 2024).

Lettuce (*Lactuca sativa* L.) is a worldwide famous leafy vegetable, particularly significant in Türkiye for its nutritional benefits and economic contribution to agriculture. (Şalk et al., 2008). Rich in vitamins, minerals, and antioxidants, lettuce is widely consumed fresh and in salads (Şalk et al., 2008). Its cultivation thrives in Türkiye's favorable climate, making it a key component of the country's horticultural sector. Therefore, measuring LA is essential for assessing plant health and growth. Thus, determining the best method for calculating LA is crucial, given lettuce's agricultural and economic relevance.

As far as we know, there is a deficiency of studies evaluating and comparing different methods for forecasting LA in lettuce cultivated under different stress conditions. Therefore, the present work aims to evaluate the effectiveness of different regression models for estimating the LA of curly lettuce through non-destructive techniques and compares predictive models using statistical criteria. Finally, for researchers and agronomists, using prediction models for LA can improve decision-making in agriculture, such as optimizing irrigation and other management practices to improve crop yields.

2. Material and Method

2.1. Experimental site

The pot trial was conducted at the Faculty of Agriculture, Ondokuz Mayıs University, Samsun, Türkiye, from 5 February to 11 May 2020. A plastic sheet was used to cover the top of the research area to protect the experiment against rainfall. During the entire growing season, a data logger recorded daily temperature and relative humidity. The relative humidity ranged from 28.1% to 100%, while the temperature varied between 0.2°C and 29.2°C. The experimental soil used was classified as loam, consisting of 31.0% silt, 23.4% clay, and 45.6% sand. The experimental soil contained 0.78 mg of nitrogen, 69.5 mg of phosphorus, and 148.4 mg of potassium per kg. Additionally, the soil had a saturated electrical conductivity of 0.22 dS m^{-1} and a pH value of 6.81.

Seeds of curly lettuce (Lactuca sativa L., cv. Couster) from Intfa Seed Company were utilized in this research. They were sown in trays and grown in a greenhouse until ready for transplantation. Healthy and uniform lettuce seedlings were chosen and transferred to 4.83 dm^3 circular plastic pots, measuring 22 cm in height, with top and bottom diameters of 18.4 cm and 15 cm, respectively. Each pot was planted with a single seedling. Before planting, the soil was naturally air-dried and sifted through a 4 mm mesh sieve. Each pot was then filled with 4.5 kg of air-dried soil. Base fertilizers consisting of phosphate and potassium were added at rates of 0.58 g and 0.88 g per pot, respectively. Nitrogen fertilizer was applied at 0.35 g per pot, with half added during seedling transplantation and the other half after one month. Diammonium phosphate, potassium sulfate, and urea chemical fertilizers were utilized to provide phosphate, potassium, and nitrogen fertilization, respectively. The fertilization procedure for growing lettuce adhered to the recommendations outlined by (Şalk et al., 2008)

2.2. Experimental design

The research was laid out following a randomized complete block design involving two factors: three doses of salicylic acid (SA) (SA_0: 0, SA_1: 1, and SA_2: 2 mM) and three levels of water salinity (S_1: 0.30, S_2: 4.15, and S_3: 8.0 dS m^{-1}), leading to a total of 9 treatments (3×3) with three replicates for each treatment (totaling 27 pots). Before transplanting, each pot's field capacity was determined by saturating the soil with tap water and covering the tops with a plastic sheet to prevent evaporation. After 48 hours, when drainage ceased, each pot was weighed, and this weight was recorded as the field capacity (Ünlükara et al., 2008; Kiremit and Arslan, 2018). Soil water depletion was monitored by weighing each pot throughout the growth cycles, and irrigation was applied when 30% of the available soil water was used by evapotranspiration during the growing season. Saline water treatments were applied 10 days post-transplanting, along with 15% leaching water during each irrigation to prevent excessive salt buildup in the pots. Two saline irrigation waters (4.15 and 8.0 dS m^{-1}) were prepared by mixing NaCl and $CaCl_2$ in a 1:1 ratio with tap water (0.30 dS m^{-1}). Prior to saline water applications, all pots received equal irrigation using 0.30 dS m^{-1} to ensure seedling adaptability to pot conditions. Foliar solutions of 0, 1, and 2 mM SA were prepared with 0.01% Tween 20 and deionized water, and foliar applications were made using a manual hand sprayer. The 0 mM SA treatment served as a control, consisting only of deionized water. Foliar applications began 12 days post-transplantation and continued every two weeks until harvest.

2.3. Leaf area analysis

The lettuce plants were harvested 76 days after being transplanted from each pot. Subsequently, all lettuce leaves were detached from the stem. The leaf area, width, and length measurements for each treatment were evaluated through image analysis. All lettuce leaves from each plant were photographed and analyzed using Adobe Photoshop CS6 imaging software. The positions of leaf length and leaf width for calculating leaf area are illustrated in Figure 1.

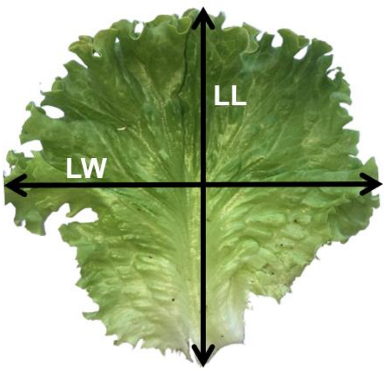

Figure 1. Position for measuring the width (LW) and length (LL) of lettuce leaves.
***Şekil 1**. Marul yapraklarının genişliği (LW) ve uzunluğunu (LL) ölçme konumları.*

2.4. Multi-linear regression analysis

Multiple linear regression analysis was applied to forecast LA using several variables, including leaf length, leaf width, salicylic acid, and irrigation water salinity. Thirteen different models were created with different input parameters to identify the best model for predicting the LA of curly lettuce plants. The model input parameters can be found in Table 1.

Table 1. The input parameters for models.
***Çizelge 1.** Modeller için girdi parametreleri*

Model No	Model input
M1	LL
M2	LW
M3	LL, LW
M4	SA, IWS
M5	SA, LL
M6	SA, LW
M7	SA, LL, LW
M8	IWS, LL
M9	IWS, LW
M10	IWS, LL, LW
M11	SA, IWS, LW
M12	SA, IWS, LL
M13	SA, IWS, LL, LW

#LL: Leaf length; LW: leaf width; SA: Salicylic acid; IWS: Irrigation water salinity.

The regression models were developed using the stepwise regression method principle (Fahrmeir et al., 2022) and analyzed using IBM SPSS 25.0 statistical software. Model variables were considered significant if their significance level was $P \leq 0.05$. Variables with a significance level greater than $P > 0.05$ were not included in the model equation. All variables in the regression models obtained had a significance level of $P \leq 0.05$. The regression model's general equation is as follows:

$$Y = \beta_0 + \sum_{i=1}^{k} \beta_i X_i + \sum_{i=1}^{k-1} \sum_{j=i+1}^{k} \beta_{ij} X_i X_j + \sum_{i=1}^{k} \beta_{ii} X_i^2 + \varepsilon \qquad (1)$$

Where Y depicts the estimated leaf area; X_i and X_j stand for independent variables (LL: leaf length; LW: Leaf width; IWS: Irrigation water salinity, SA: Salicylic acid), and β, β_i, and β_{ii} represent the intercept, linear coefficients, and quadratic coefficients respectively. B_{ij} denotes the interaction coefficients between variables; k is the number of variables examined; and ε represents the error term.

2.6. Statistical evaluation of the developed models

The developed models' accuracy was assessed using three common metrics: the coefficient of determination (R^2), root mean square error (RMSE), and mean absolute error (MAE), as defined in Equations 2-4 by

Willmott and Matsuura (2005). These metrics provide a comprehensive evaluation of the models' effectiveness.

$$R^2 = 1 - \frac{\sum_{i=1}^{n}\left(M_i - P_i\right)^2}{\sum_{i=1}^{n}(M_i - P_{avg})^2} \quad (2)$$

$$RMSE = \sqrt{\frac{\sum_{i=1}^{n}\left(M_i - P_i\right)^2}{n}} \quad (3)$$

$$MAE = \frac{1}{n}\sum_{i=1}^{n}\left|M_i - P_i\right| \quad (4)$$

Where, M_i: measured values of LA; P_i: predicted values of LA; P_{avg}: average of the measured value of LA; n: number of observations; i: th observations of the variables measured and predicted.

3. Results and Discussion

3.1. Description of the sampled data

The average values of leaf parameters (LL, LW, and LA) for the different treatments are depicted in Table 2. The collected data showed that leaf length varied between 7.2 and 10.0 cm for SA_0, 7.9 and 12.5 cm for SA_1, and 8.5-12.5 cm for SA_2 (Table 2). Leaf width ranged from 6.8-12.1 cm for SA_0, 7.3 and 11.8 cm for SA_1, and 7.3-9.6 cm for SA_2 (Table 2). Moreover, the actual LA of curly lettuce plants varied from 31.9 to 75.6 cm^2 (SA_0), 37.5 and 92.0 cm^2 (SA_1), and 40.2 and 72.6 cm^2 (SA_2) (Table 2). The leaf width and length of the lettuce decreased linearly as salinity stress increased. The smallest values for these characteristics were noted at 8.0 dS m^{-1} under all foliar application conditions. However, lettuce plants treated with 1 and 2 mM SA via foliar application exhibited greater leaf length values compared to those treated with 0 mM SA (Table 2).

Table 2. Some statistical values of lettuce grown under different water salinity and foliar applied salicylic acid.
***Çizelge 2**. Farklı tuzlu su ve yapraktan salisilik asit uygulanan koşullarda yetiştirilen marul bitkisinin bazı istatistiksel değerleri.*

Treatments	Leaf length (LL, cm)			Leaf width (LW, cm)			Leaf length (LA, cm^2)		
	Mean ± SD	Min	Max	Mean ± SD	Min	Max	Mean ± SD	Min	Max
$SA_0 \times S_1$	10.0 ± 2.5	5.6	16.0	12.1 ± 1.7	8.9	16.1	75.6 ± 31.1	38.5	159.9
$SA_0 \times S_2$	8.1 ± 1.9	5.2	12.9	10.8 ± 1.3	6.0	13.3	52.5 ± 15.1	24.6	91.2
$SA_0 \times S_3$	7.2 ± 0.7	5.5	8.4	6.8 ± 1.0	5.0	8.5	31.9 ± 6.8	16.0	44.8
$SA_1 \times S_1$	12.5 ± 1.5	9.8	15.4	11.8 ± 1.7	8.6	15.2	92.0 ± 22.2	56.8	139.5
$SA_1 \times S_2$	10.5 ± 1.2	8.9	13.8	8.7 ± 1.8	5.4	13.3	56.6 ± 15.5	36.6	111.0
$SA_1 \times S_3$	7.9 ± 0.7	6.1	9.7	7.3 ± 0.9	5.3	8.9	37.5 ± 5.2	25.2	49.3
$SA_2 \times S_1$	12.5 ± 1.3	9.8	15.0	9.6 ± 1.9	6.4	13.1	72.6 ± 19.9	45.7	110.9
$SA_2 \times S_2$	9.3 ± 1.1	7.2	11.5	7.7 ± 1.8	3.9	10.7	45.8 ± 12.6	18.9	67.6
$SA_2 \times S_3$	8.5 ± 1.0	6.4	10.8	7.3 ± 1.8	3.4	10.2	40.2 ± 12.6	17.1	59.5

#SA_0, SA_1, and SA_2 denote 0, 1, and 2 mM SA doses, respectively. S_1, S_2, and S_3 indicate 0.30, 4.15, and 8.0 dS m^{-1} saline waters, respectively.

Table 3. Developed regression equations for the prediction of lettuce leaf area by using various input parameters.
***Çizelge 3.** Farklı girdi parametreleri kullanılarak marul yaprağı alanının tahmini için geliştirilen regresyon denklemleri.*

Model No	Regression equation
M1	$LA = -34.23 + 9.40\times LL$
M2	$LA = -23.50 + 8.69\times LW$
M3	$LA = 10.36 + 0.85\times LW - 1.68\times LL + 0.66\times LW\times LL - 0.12\times LW^2 + 0.01\times LW\times LL^2$
M4	$LA = 87.74 - 10.11\times IWS - 2.8\times SA^2 + 0.44\times IWS^2 + 0.49\times SA^2\times IWS$
M5	$LA = 20.33 + 9.59\times SA - 2.47\times LL - 1.84\times SA\times LL + 0.7\times LL^2$
M6	$LA = 64.64 - 13.61\times SA - 12.54\times LW + 5.74\times SA\times LW - 5.7\times SA^2 + 1.08\times LW^2 - 0.21\times SA\times LW^2$
M7	$LA = -11.92 + 4.68\times LW + 0.3\times LL + 0.81\times SA\times LL + 0.19\times LW\times LL - 0.18\times LW^2 - 0.08\times SA\times LL^2 + 0.02\times LW\times LL^2$
M8	$LA = 80.7 - 2.59\times IWS - 10.78\times LL + 0.89\times LL^2 + 0.01\times IWS\times LL^2$
M9	$LA = 31.64 - 2.96\times IWS - 0.69\times LW + 0.23\times IWS^2 + 0.45\times LW^2 - 0.01\times IWS\times LW^2$
M10	$LA = 27.54 - 1.04\times IWS - 1.3\times LW - 2.72\times LL + 0.85\times LW\times LL - 0.02\times IWS^2 - 0.09\times LW^2 + 0.01\times IWS^2\times LW$
M11	$LA = 27.1 + 20.55\times SA + 2.23\times IWS - 6.14\times LW - 4.03\times SA\times IWS + 3.48\times SA\times LW - 10.59\times SA^2 + 0.83\times LW^2 + 0.44\times SA^2\times IWS - 0.2\times SA\times LW^2 - 0.03\times IWS\times LW^2 + 0.06\times SA^2\times IWS^2 + 0.02\times SA\times IWS\times LW^2$
M12	$LA = 60.98 + 13.52\times SA - 2.54\times IWS - 7.36\times LL - 3.07\times SA \times IWS - 2.36\times SA\times LL - 0.3\times SA^2 + 0.86\times LL^2 + 0.27\times SA\times IWS\times LL + 0.19\times SA\times IWS^2 + 0.01\times SA^2\times LL^2$
M13	$LA = 4.37 - 0.09\times IWS + 1.93\times LW - 2.51\times LL + 1.03\times SA\times LW + 0.01\times SA\times LL + 0.29\times LW\times LL + 0.05\times IWS^2 + 0.35\times LL^2 - 0.01\times IWS\times LW\times LL - 0.09\times SA\times LL^2$

LA: Leaf area, LL: leaf length; LW: Leaf width; IWS: Irrigation water salinity, SA: Salicylic acid.

The maximum leaf width was observed in the $SA_0 \times S_1$ treatment. In particular, applying 1 and 2 mM SA to the leaves improved the leaf length of lettuce plants under S_1 conditions in comparison to 0 mM SA. In terms of LA values, the $SA_1 \times S_1$ treatment showed a higher LA value than the other treatments. As shown in Table 2, the foliar application of SA had a positive impact on the LA of lettuce plants under salt-stress conditions. Exogenous salicylic acid significantly reduced lipid damage and maintained membrane integrity, thereby preventing oxidative damage from salt stress (Peng et al., 2021). Additionally, Nigam et al. (2022) concluded that foliar spray of SA enhanced spinach LA by increasing photosynthesis and improving water and nutrient uptake. Ghassemi-Golezani and Farhadi (2022) reported that endogenous salicylic acid reduced the translocation of toxic ions (Na^+ and Cl^-) to the shoots, enhanced the uptake of essential cations, and improved the LA of pennyroyal plants under salinity stress. Our results align with Kusvuran and Yilmaz (2023) and Yavuz et al. (2023), who reported that exogenous SA increased the LA of lettuce plants under saline conditions.

Table 4. Statistical evaluation of the developed models.
Çizelge 4. *Regresyon modellerinin istatistiksel değerlendirmesi*

Model No	R^2	RMSE (cm^2)	MAE (cm^2)
M1	0.728	13.18	9.70
M2	0.719	13.40	10.29
M3	0.962	7.58	5.34
M4	0.505	17.79	13.05
M5	0.837	10.22	8.10
M6	0.853	9.71	7.07
M7	0.965	5.58	4.09
M8	0.794	11.47	8.90
M9	0.787	11.70	8.23
M10	0.966	4.95	3.72
M11	0.844	10.94	8.36
M12	0.866	9.29	7.25
M13	0.968	4.59	3.44

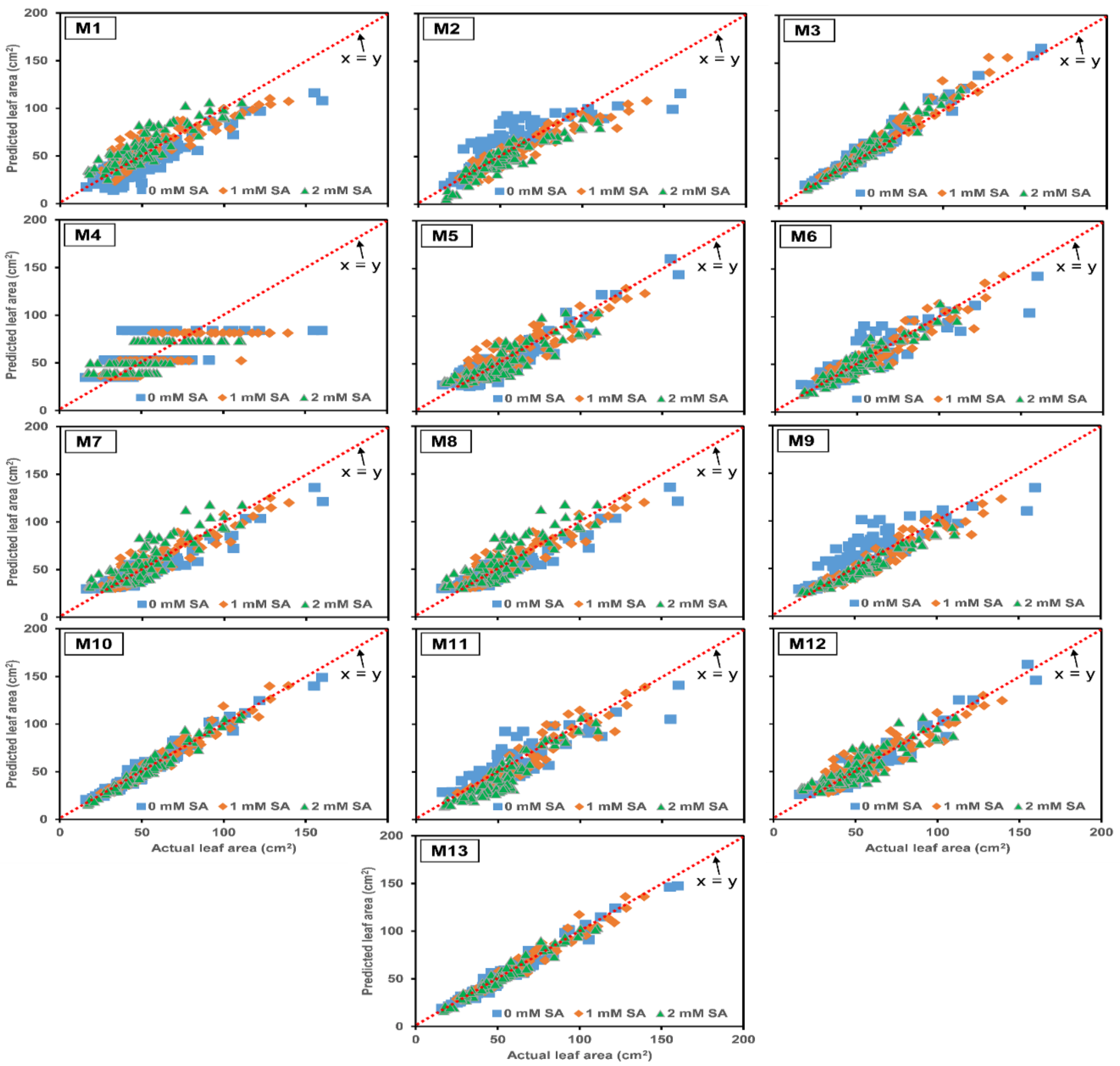

Figure 2. Relation between actual leaf area and predicted leaf area with different regression models.
Şekil 2. *Gerçek yaprak alanı ile tahmin edilen yaprak alanı arasındaki ilişki.*

3.2. Comparison of developed regression models

In the present investigation, 13 models were developed to forecast the LA of curly lettuce under varying salinity stress and SA doses. The regression equations are detailed in Table 3, with determination coefficients (R^2) ranging from 0.505 to 0.968 (Table 4). The RMSE values varied between 4.59 and 17.79 cm^2, while the MAE ranged from 3.44 to 13.05 cm^2. The scatter plots for the actual LA and predicted LA for each model are depicted in Fig. 2. Notably, Models 1 and 2, which used only one leaf dimension (LL or LW), explained 72% of the total variation in LA (Table 4). However, combining both dimensions in Model 3 significantly enhanced estimation accuracy, achieving a high R^2 of 0.962 and low RMSE (7.58 cm^2) and MAE (5.34 cm^2). Model 4 showed the poorest accuracy in predicting the LA, with the highest RMSE (17.79 cm^2) and MAE (13.05 cm^2). This suggests that creating a regression equation using SA and IWS inputs is not appropriate for accurately predicting the LA of curly lettuce. When IWS or SA parameters were included either individually or together in LL and LW models, the predictability of the regression models significantly increased. This suggests that incorporating IWS or SA in LA regression models is crucial for achieving highly accurate models. When comparing Model 7 and Model 10, they both exhibit high R^2 values (0.965 and 0.966) that are very close. However, Model 7 has RMSE and MAE values of 5.58 and 4.09 cm^2, respectively, while Model 10 has values of 4.95 and 3.72 cm^2, respectively. These results suggest that incorporating IWS along with LL and LW parameters significantly enhanced the accuracy of the model's predictive capability. However, in Model 11 and Model 12, incorporating SA and IWS with only LL or LW parameters significantly reduced the predictive capability of the models, suggesting that the use of LL and LW parameters enhanced the accuracy of the developed models. Among all the models, Model 13 exhibited the highest accuracy in predicting the LA of curly lettuce, with R^2, RMSE, and MAE values of 0.968, 4.59 cm^2, and 3.44 cm^2, respectively. By including IWS, SA, LL, and LW parameters in the development of prediction models, the accuracy of the regression equation was improved. Therefore, incorporating stress conditions into LL and LW prediction models will lead to more accurate predictions of the LA for curly lettuce cultivation. This additional information provides a more detailed insight into the predictive capabilities of the models when stress conditions are considered. Considering all of these factors, the use of Model 13 resulted in the highest accuracy in estimating LA, improving prediction accuracy by reducing RMSE by 7.84% and 21.57% compared to models M10 and M7, respectively.

Leaf area is a vital variable in studies of plant growth and development, affecting light absorption, photosynthesis, and the efficiency of water loss. It directly influences how plants respond to fertilizers and irrigation methods (Soheili et al., 2023). For instance, in lettuce, a larger LA enhances light capture and photosynthesis, improving growth, yield, and nutritional quality. Understanding LA is essential for optimizing productivity and maintaining the quality of lettuce crops. Numerous studies have focused on estimating LA by measuring leaf dimensions, typically using the combination of LL and LW as parameters in LA models. For instance, Peksen (2007) proposed $LA = 0.919 + 6.82 \times LL \times LW$ equation for faba pean leaf estimation. Cemek et al. (2011) utilized leaf measurements (LL and LW) to create regression models for estimating the LA of green pepper. They suggested that incorporating these measurements significantly enhanced the predictive accuracy of the models. Kandiannan et al. (2009) introduced a model to estimate the LA of ginger as $LA = 0.0146 + 0.6621 * LW$, with an R^2 value of 0.997. This model is considered a dependable method for non-destructively estimating the LA of ginger plants. Ribeiro et al. (2020) proposed the equation $LA = 0.6740 * LW$, which effectively estimates the LA of E. pauferrense using a linear model without intercept.

As mentioned above, using leaf length and width parameters effectively predicts LA across different plants. Our findings show that incorporating stress-related variables, such as IWS and SA, enhances the model's ability to account for variations in LA due to these stressors. This results in more accurate predictions, as the model is better suited to address the complexities of real-world scenarios where stress conditions are prevalent. Consequently, the model closely aligns with the actual observed LA in plants under diverse environmental conditions. For instance, integrating water deficit rates and irrigation water salinity parameters with LL and LW models enhanced predictability for bell pepper (Cemek et al., 2020) and chokeberry (Akyüz and Cemek, 2024) compared to using LL and LW alone. Kiremit (2024) suggested that melatonin doses and soil salinity parameters can effectively predict the LA of sweet corn seedlings. Finally, it can be confidently recommended that Model 13 is highly suitable for providing efficient and precise predictions regarding the area of curly lettuce leaves. This model effectively eliminates the necessity for expensive and potentially cumbersome methods, making it a practical choice for those seeking to enhance their prediction accuracy without incurring high costs.

4. Conclusion

The study presents a model for estimating LA in curly lettuce based on leaf width and length, enabling non-invasive and straightforward predictions without requiring specialized staff or costly equipment. In this context, Model 3 can accurately predict LA using only leaf width and length as inputs. Moreover, Model 13 is suitable for predicting LA in curly lettuce under varying irrigation water salinity and salicylic acid levels. By incorporating these factors into the prediction models, the accuracy and reliability of the models are improved, making them valuable tools for understanding and managing plant growth in different conditions. The regression equations developed can help researchers in future studies on the growth, physiology, and propagation of curly lettuce, providing a precise and non-destructive method.

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Seed and Pollen Transmission of Tomato spotted wilt orthotospovirus (TSWV) in Pepper

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Abstract: Orthotospovirus tomatomaculae also known by Tomato spotted wilt orthotospovirus (TSWV) is a harmful pathogen that significantly reduces crop production and quality. TSWV can infect approximately 1,000 plant species in 84 different plant families, such as tomato, pepper, lettuce, tobacco, and various types of wild plants, belonging to the Solanaceae family. TSWV is a member of the *Orthotospovirus* genus and is an isometric-enveloped particle. TSWV can be transmitted mechanically, seed-borne, and by nine species of thrips belonging to the genera Thrips, Scirtothrips,Frankliniella.In this study, it was determined that TSWV could be transmitted by seeds, but no transmission by pollen was detected. RT-PCR and visual assessment data demonstrate the seed transmission ability of TSWV in peppers. TSWV was detected in infected pepper seeds (seed and seed coat). Additionally, TSWV was detected in different organs of the pepper plant. TSWV was detected in the endocarp, mesocarp, exocarp, pedicel, and placenta of pepper fruit and sepal, petal, pistil, and stamen of pepper flower. After the germination of infected pepper seeds, TSWV was detected with specific primers by RT-PCR in the cotyledons and true leaves, but no symptoms were observed. These data provided information about the localization of TSWV in pepper plants and provided further evidence of TSWV seed transmission ability.

Keywords: Pepper plant, tomato spotted wilt orthotospovirus, pollen transmission, seed transmission, localization of virus.

Biberde Domates Lekeli Solgunluk Virüsünün Tohum ve Polen ile Taşınması

Öz: Orthotospovirus tomatomaculae olarakta bilinen Domates lekeli solgunluk virüsü (TSWV), ürün verimini ve kalitesini önemli ölçüde azaltan zararlı bir patojendir. TSWV, domates, biber, marul, tütün ve Solanaceae familyasına ait çeşitli yabani bitkiler gibi 84 farklı bitki familyasındaki yaklaşık 1.000 bitki türünü enfekte edebilir. TSWV, Orthotospovirüs cinsinin bir üyesi olup izometrik zarflı bir partiküldür. TSWV, mekanik olarak, tohum yoluyla ve Thrips, Scirtothrips ve Frankliniella cinslerine ait dokuz trips türü tarafından bulaşabilir. Bu çalışmada, TSWV'nin tohumlarla taşınabildiği, ancak polen yoluyla taşınma kabiliyetinin olmadığı belirlenmiştir. RT-PCR ve görsel değerlendirme verileri, TSWV'nin biberlerde tohumla taşınma kabiliyetini göstermektedir. Enfekteli biberlerden toplanan tohumlarda (tohum ve tohum kabuğu) TSWV tespit edilmiştir. Ek olarak, enfekteli, biber meyvesinin endokarp, mezokarp, ekzokarp, pedicel ve plasentasında ve biber çiçeğinin çanak yaprağı, taç yaprağı, pistili ve erkek organında TSWV saptanmıştır. Enfekteli biber tohumlarının çimlenmesinden sonra, TSWV kotiledonlarda ve gerçek yapraklarda spesifik primerler kullanılarak RT-PCR ile tespit edilmiş, ancak hiçbir simptom gözlemlenmemiştir. Bu veriler, TSWV'nin biber bitkisinde ki lokalizasyonu hakkında bilgi sağlamış ve TSWV tohum ile taşınabildiğine dair daha fazla kanıt sağlamıştır.

Anahtar kelimeler: Biber bitkisi, domates lekeli solgunluk virüsü, polen taşınması, tohum taşınması, virüs lokalizasyonu.

1. Introduction

Orthotospovirus tomatomaculae (TSWV) is a plant virus belonging to the *Bunyaviridae* family. It is characterized by isometric enveloped particles and possesses a tripartite genome consisting of three RNA segments: 8.9 kb L-RNA, 5.4 kb M-RNA, and 2.9 kb S-RNA (de Haan et al., 1990). Among these, one RNA is negative-sense while the other two are positive-sense (Usta et al., 2023). TSWV is notorious for its detrimental impact on nearly 1000 plant species across 84 families, including important crops like tomato, pepper, squash, and others within the Solanaceae family (Gordillo et al., 2008). Transmission of TSWV primarily occurs through mechanical transmission and via thrips vectors. Nine species of thrips from the *Thrips* sp., *Scirtothrips* sp., and *Frankliniella* sp., genera can transmit the virus in a circulative and propagative manner. Notably, *Frankliniella occidentalis* is particularly effective in transmitting TSWV (Coutts & Jones, 2005).

Symptoms of TSWV infection in plants are distinct, including leaf bronzing, black spots on leaves and stems, fruit wilting, and deformations (Güldür et al., 1995). Fruits exhibiting these symptoms are unsuitable for fresh consumption and lose economic value, particularly affecting industrial tomato production (Turhan & Korkmaz, 2006).

Apart from mechanical and vector transmission, several studies have also demonstrated seed transmission of Tomato spotted wilt virus (TSWV). The initial study on *Senecio cruentu* reported a 96% efficiency in TSWV seed transmission (Jones, 1944). Another study indicated a 1% transmission rate of TSWV through tomato seeds (Crowley, 1957). Furthermore, RT-PCR and electron microscope analyses confirmed the transmission of TSWV through pepper seeds in a different study (Wang et al., 2022). Additionally, Groundnut bud necrosis orthotospovirus (GBNV) and Soybean vein necrosis virus (SYNV), both belonging to the *Tospoviridae*, are seed-transmitted in soybean and peanut, respectively (Groves et al., 2016; Sastry, 2013). Despite all this evidence, some studies have reported that plants infected with TSWV cannot transmit the virus to new-generation plants (Antignus et al., 1997). The seed transmission of plant viruses depends on whether the virus can enter the plant and reach its reproductive organs, infect these organs, and whether the infected reproductive organs can proliferate and survive. Therefore, the most determining factor in seed transmission is the virus's ability to accumulate in the host reproductive organs (Cobos et al., 2019).

In this study, TSWVAntRB isolate was infected with pepper plants through mechanical inoculation. Visual and RT-PCR analyses confirmed the seed transmission of TSWV, with the virus detected in both the pepper seed and seed coat. However, TSWV was not detected in the second-generation pepper plants obtained through pollination from TSWV-infected pepper plants. Total nucleic acid isolation was performed from various parts of TSWV-infected pepper plants to determine the localization of TSWV within the pepper plants.

2. Materials and Methods

2.1. Growing conditions and plant material

The pepper cultivar Maxibell was used to investigate the seed and pollen transmissibility of the Tomato spotted wilt virus (TSWV). Maxibell seeds were generously provided by Assoc. Prof. Ümit Özyılmaz from Aydın Adnan Menderes University. (Aydın, Turkey). All plants were maintained in a growth chamber at Aydın Adnan Menderes University (Aydın, Turkey), under controlled conditions of 22±5 °C temperature, 60% relative humidity, an 8/16-hour photoperiod, and watered every 3 days with tap water. Healthy plants and virus-infected plants were segregated and placed on separate shelves within the same room.

2.2. Virus inoculation

The TSWV used in this study is a common strain (TSWVAntRB) which is shown blackspot containing light and dark-green areas in tomato. Inoculum was prepared by grinding infected tissues at the rate of 1:6 (wt/vol) tissue to buffer ratio in freshly prepared ice-cold 0.05 M potassium phosphate buffer, pH 7.0, containing 0.1% mercapto-ethanol with a chilled mortar and pestle. Debris was removed by squeezing the extract through a cotton bud. The inoculum was kept on ice until the inoculation was completed. Inoculum was applied by rubbing both surfaces of the leaf with a brush on pepper plants at four leaf stages (8 to 10 days after planting). After inoculation, the plants were sprayed with distilled water and kept in two growth chambers having the same environmental conditions (Oğuz et al., 2009).

2.3. Sample Collection

Symptomatic leaves of pepper plants were collected at 45 days post-inoculation (dpi). During the generative stage, flowers were collected from each of the TSWV-inoculated plants. Flower samples were collected during anthesis and dissected into sepals, petals, pistils, and stamens using sterilized scalpels. To prevent contamination, scalpels were sterilized with 70% ethanol and flamed to remove excess ethanol before each dissection. Total nucleic acid (TNA) was extracted from each flower part for virus detection.

Young pepper fruits were carefully separated into exocarp, mesocarp, endocarp, placenta, pedicel, and seeds using sterilized scalpels. TNA extraction was performed on each part to detect the virus. Seed coats were collected from germinating seeds, while cotyledons and leaves were collected upon the emergence of the first true leaf.

2.4. Cross-pollination

Cross-pollination between healthy and infected pepper plants was performed to investigate the vertical transmission of TSWV. Pepper plants produce perfect flowers containing both pistils and stamens within the same flower. Female flower sources were selected from healthy pepper plants that had not yet bloomed. Before pollination, all flower parts except the sepals and pistils were removed to prevent self-pollination. These female flowers were then pollinated with pollen collected from infected pepper plants.

2.5. Total nucleic acid (TNA) extraction and RT-PCR detection

Total nucleic acid samples were extracted from TSWV-infected parts of pepper plants to detect the presence of TSWV (Svanella-Dumas et al., 2000). PCR analysis was conducted using 1-Step Hot Start Master Mix® (Thermo Fisher, Massachusetts, USA). The PCR mixture included 12.5 µl of Hot Start master mix, 0.5 µl of verso enzyme, and 1.25 µl of RT-enhancer for amplification. Inoculum plant infected with TSWV was used as a positive control. The following PCR-specific primers of TSWV (Mumford et al., 1994) (F: 5'-AATTGCCTTGCAACCAATTC-3' and R: 5'-ATCAGTCGAAATGGTCGGCA-3') was used to detect a 276 base pair fragment of the TSWV L-RNA segment. The amplification conditions for the PCR reaction were as follows 50°C 15 min, 95°C 35 min for 1 cycle 94°C for 1 min, 55°C for 1min and 72°C 1 min for 35 cycles: 5 min for 72°C and each 25µl sample mixture (Fidan & Sari, 2019). Data analysis was performed using electrophoresis.

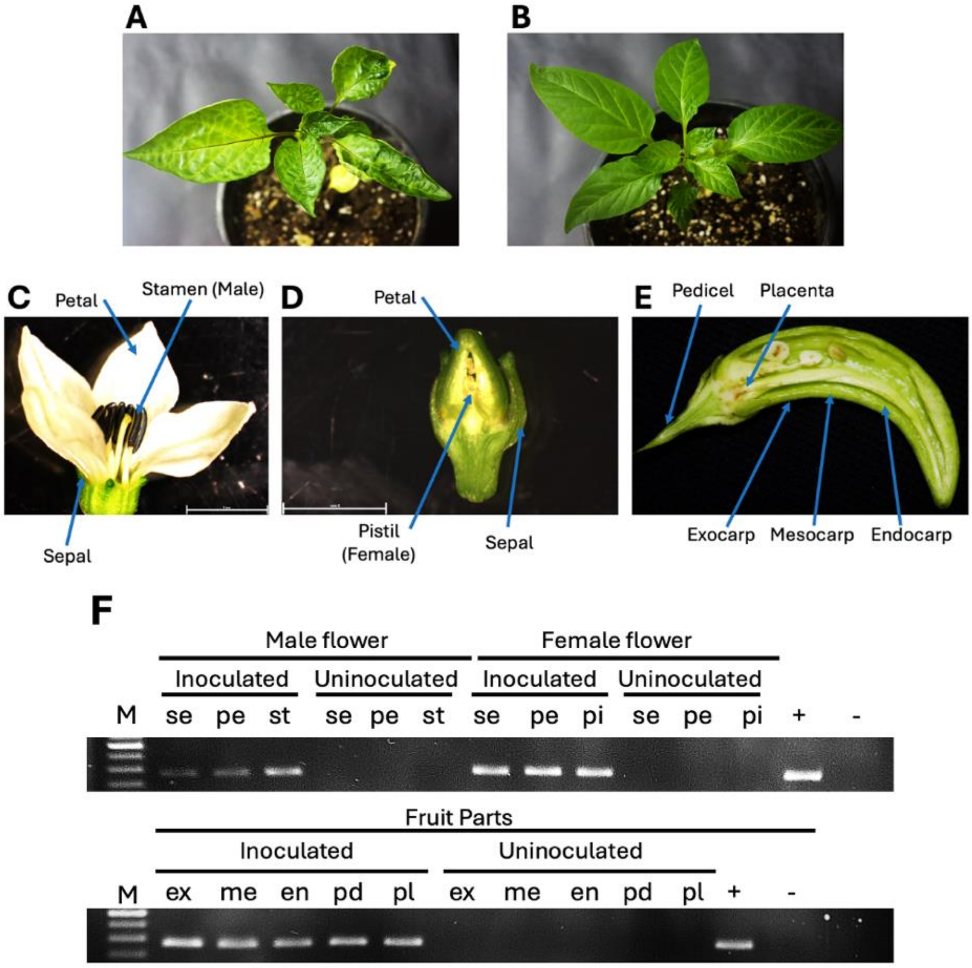

Figure 1. Symptom development, sample collection, and detection of Tomato spotted wilt virus (TSWV) in infected pepper flowers and fruits. **A,** Symptoms of a pepper leaf induced by TSWV. **B,** Healthy pepper leaf. The symptom photos were taken at 45 days post-inoculation. **C,** Pepper male flower parts. **D,** Pepper female flower parts. **E,** Pepper fruit parts. **F,** RT-PCR detection of pepper flower and fruit parts using TSWV-specific primer. M= marker; + = PCR positive control; **-** = PCR negative control; se = sepal; pe = petal; pi = pistil; st = stamen; ex = exocarp; me = mesocarp; en = endocarp; pd = pedicel; and pl = placenta.

Şekil 1. *Enfekteli biber çiçek ve meyvelerinde Domates lekeli solgunluk virüsü'nün (TSWV) simptom gelişimi, örnek toplanması ve tespiti.* ***A,*** *TSWV tarafından oluşturulan biber yaprağındaki simptomlar.* ***B,*** *Sağlıklı biber yaprağı. Simptom fotoğrafları inokulasyondan 45 gün sonra çekildi.* ***C,*** *Biber erkek çiçek organları.* ***D,*** *Biber dişi çiçek organları.* ***E,*** *Biber meyvelerinin parçaları.* ***F,*** *TSWV'ye özgü primer kullanılarak biber çiçeği ve meyve parçalarının RT-PCR tespiti. M= işaretleyici; + = PCR pozitif kontrol;* ***-*** *= PCR negatif kontrol; se = çanak yaprak; pe = taç yaprak; pi = pistil; st = stamen; ex = ekzokarp; me = mezokarp; en = endokarp; pd = pedicel; ve pl = plasenta.*

3. Results

3.1.TSWV moves to and accumulates in systemic leaves, flowers, and fruit parts

Yellowing and leaf curling symptoms were observed on pepper plants at 45 days post-inoculation (dpi) with TSWV compared with healthy peppers (Figure. 1A and B). The presence of TSWV infection was confirmed using RT-PCR with virus-specific primers. Female and male flowers, as well as immature fruit samples, were collected and carefully dissected into various parts for virus detection (Figure. 1C to E). Positive RT-PCR signals indicating TSWV presence were detected in all tested flower parts and fruit tissues (Figure. 1G). These results showed that TSWV could move to all tested flower parts and fruit tissues of pepper plants.

3.2. TSWV exists in the progenies of infected plants

Pepper plants infected with TSWV produced fewer mature seeds compared to healthy plants. Mature pepper fruits and seeds were collected at 90 days post-inoculation (dpi). Following germination, various tissues were sampled for total nucleic acid (TNA) extraction and RT-PCR analysis. TSWV was detected in seed, seed coat,cotyledon, and leaf tissues with no observable symptoms on virus-infected cotyledons(Figure.2).

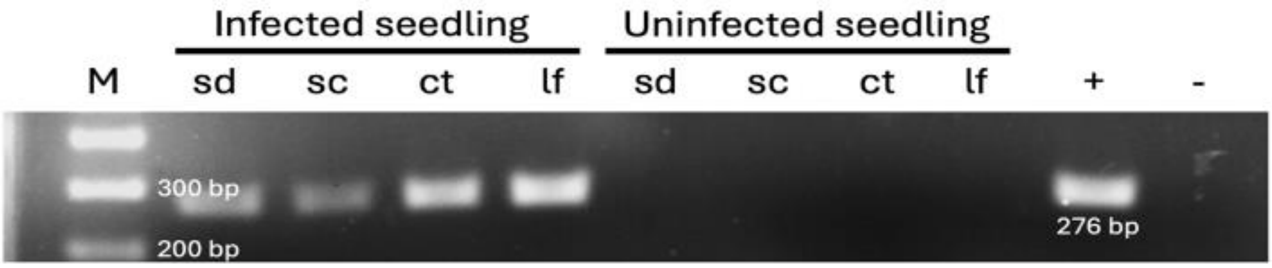

Figure 2. PCR results of whole seed (sd), seed coat (sc), cotyledon (ct), and leaf (lf) using TSWV-specific primers. Lane of DNA marker (M) is labeled. + = PCR positive control, – = PCR negative control.

Şekil 2. *TSWV'ye özgü primerler kullanılarak tüm tohum (sd), tohum kabuğu (sc), kotiledon (ct) ve yaprağın (lf) PCR sonuçları. DNA marker (M) etiketlenmiştir. + = PCR pozitif kontrol, – = PCR negatif kontrol.*

Table 1. Detection of Tomato spotted wilt virus in progenies derived from virus-infected pepper

Çizelge 1. *Virüsle enfekte olmuş biberlerden elde edilen ilk nesillerde Domates lekeli solgunluk virüsünün tespiti*

Virus	Germination rate[a]	Infection rate[b]
TSWV	117/150 (78%)	68/117 (58.12%)
Uninoculated plant	142/150 (94.67%)	0/142 (0%)

[a]Germination rate = (number of germinated seeds/numbers of tested seeds) × 100%.
[b]Infection rate = (number of seeds tested positive with PCR/number of germinated seeds) × 100%.

A total of 150 seeds derived from TSWV-infected plants were germinated and tested for virus presence, alongside 150 seeds from uninoculated plants used as a control group. Seeds from TSWV-infected plants exhibited a germination rate of 78% and an infection rate of 58.12%. In contrast, seeds from uninoculated plants showed a germination rate of 94.67%, and no virus was detected in their progeny (Table 1).

3.3. TSWV is not pollen-transmissible

To investigate the potential transmission of TSWV through pollination, pollen from virus-infected plants was collected and used for cross-pollination of female flowers on healthy plants (Figure. 3A and B). Systemic leaves and mature fruits resulting from cross-pollination were collected simultaneously. No symptoms were observed on the pepper plants six weeks after fruit collection. Analysis revealed the absence of TSWV in all tested parts of the cross-pollinated fruits and systemic leaves. Furthermore, PCR testing conducted on systemic leaves of the pollinated plants and on a subsequent self-pollinated fruit, which grew adjacent to the cross-pollinated fruit on the same stalk, yielded negative results (Figure. 3C).

4. Discussion

This study aims to determine the seed and pollen-mediated transmission ability of TSWV and to determine the localization of TSWV in pepper plants. Thus, we aimed to create a step forward by obtaining more information on TSWV transmission and localization and filling the gaps for future management of TSWV.

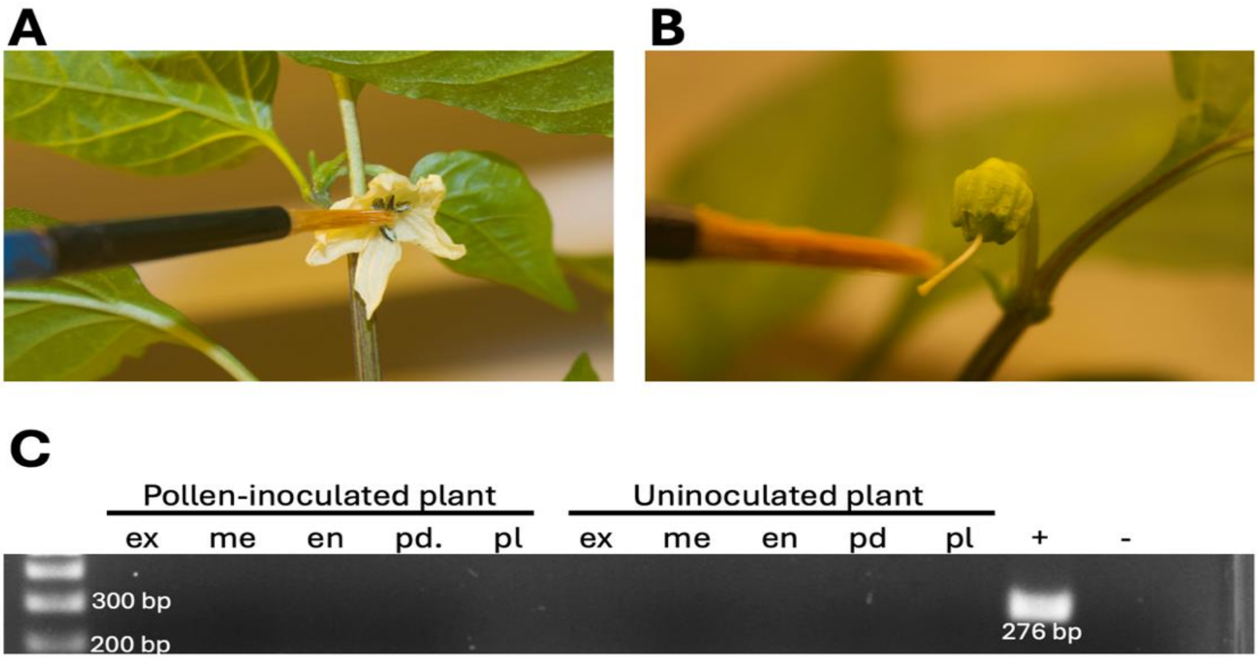

Figure. 3. Cross-pollination of TSWV-infected pepper and healthy plant and PCR detection of Tomato spotted wilt virus in pepper fruit after cross-pollination. **A,** Collection of pollen from the TSWV-infected pepper male reproductive organ (stamen). **B,** Cross-pollination of collected pollen by applying it to the female reproductive organs (pistil) of healthy pepper plants. **C,** RT-PCR detection of TSWV in inoculated pepper plants after cross-pollination using specific primers. M = marker; + = PCR positive control; – = PCR negative control; ex = exocarp; me = mesocarp; endo = endocarp; pd = pedicel; and pl = placenta.

Şekil 3. *TSWV ile enfekteli biber ve sağlıklı biberin çapraz tozlaşmasından sonra biber meyvesinde Domates lekeli solgunluk virüsünün PCR ile tespiti.* ***A,*** *TSWV ile enfekte biberin erkek üreme organından (stamen) polen toplanması.* ***B,*** *Toplanan polenin sağlıklı biber bitkilerinin dişi üreme organlarına (pistil) uygulanması yoluyla çapraz tozlaşması.* ***C,*** *Spesifik primerler kullanılarak çapraz tozlaşmadan sonra aşılanmış biber bitkilerinde TSWV'nin RT-PCR ile tespiti. M = marker; + = PCR pozitif kontrol; – = PCR negatif kontrol; ex = ekzokarp; me = mezokarp; endo = endokarp; pd = pedisel; ve pl = plasenta.*

Seed transmission occurs in 20% of plant viruses, and thus can rapidly emerge in new generations of plants (Sandra et al., 2020; Schaad, 1988). Apart from mechanic and vector transmission, seed transmission of TSWV is a gap that is not fully clarified. In this study, we provide further evidence for seed transmission of TSWV. TSWV was detected by RT-PCR in pepper seeds obtained from TSWV-infected plants (Figure 2.). TSWV was detected not only in the seed but also in the seed coat, cotyledon, and leaves. Another study showed that pepper seeds carry TSWV by RT-PCR and electron microscopy (Wang et al., 2022). In addition, studies are showing that TSWV is also carried in *Senecio cruentu* and tomato seeds (Crowley, 1957; Jones, 1944). Although seed transmission of Tomato spotted wilt virus (TSWV) has not been detected in peanuts, the virus has been detected in the testa of immature seeds and freshly harvested mature seeds. However, serological detection of the virus in testa was successful only in dried seeds. When freshly harvested mature seeds containing the infective virus in the testa were tested using growth assays, they did not transmit TSWV (Prasada Rao et al., 2009; Reddy et al., 1983). The data obtained may indicate that seed transmission may be affected by environmental conditions, genetics, and virulence, but may not be sufficient to determine the effectiveness of its spread in other pepper varieties or different plants. Although seed-mediated transmission is seen in this pepper variety, the route of seed transmission from male and female individuals needs to be further investigated.

Most studies have shown that seed-mediated transmission of plant viruses generally occurs through the embryo (Ellis et al., 2020; Johansen et al., 1994). Plant viruses enter the seed embryo either directly by entering the embryo of the seed or indirectly through pollen or egg cells (Suruthi et al., 2018). In this study, we proved that TSWV can be transmitted directly (by entering the seed embryo), but not indirectly (by pollen). Cross-pollination was carried out from infected plants to healthy plants, but no transmission was observed (Figure.3.). Replication and spread of TSWV in plants is markedly restricted by its unique seed structure, which lacks the plasmodesmata required for effective virus movement (Carrington et al., 1996; Crowley, 1957). Therefore, the localization, accumulation, and presence of the virus in the seed and embryo may vary in different pepper varieties. TSWV was detected in cotyledons developing from infected seeds. It may occur because of factors such as low immunity in young seedlings and cotyledons, the plant's need for rapid growth, and its inability to meet the necessary nutritional needs (Zou et al., 2018). TSWV-infected pepper seeds often cannot be detected because the disease does not show any symptoms during germination (Kothandaraman et al., 2016; Suruthi et al., 2018). TSWV has been found in pepper seeds and can be transmitted symptomless to future generations without being detected by visual inspection.

In this study, the localization of TSWV in different organs of pepper was also determined. TSWV was detected in the sepal, petal, stamen, pistil (flower) and exocarp, mesocarp, endocarp, pedicel, and placenta (fruit) (Figure. 1.). The presence of TSWV in all tested parts of both flower and fruit proves that it spreads rapidly and strongly within the plant.

This study provides evidence of seed-borne transmission of Tomato spotted wilt virus (TSWV) in pepper (*Capsicum annuum* L.) plants. Additionally, TSWV was detected in the seed, seed coat, and young seedlings of the second generation of peppers, indicating a potential new mode of seed transmission in this plant. However, further investigation is needed to understand the key factors involved in this process. The timing of virus infection in crops could play a crucial role in seed transmission under field and greenhouse conditions. Our cross-pollination analyses demonstrated that TSWV is not pollen transmissible. Furthermore, this study mapped the localization of TSWV in various organs of the pepper plant, including the sepal, petal, stamen, pistil (flower), and exocarp, mesocarp, endocarp, pedicel, and placenta (fruit). Preventing seed transmission is equally critical as controlling thrips and mechanical transmission of TSWV for the seed industry, both in production and marketing.

This report represents the first documentation that TSWV is not pollen transmissible and provides insights into its localization within different parts of the pepper plant.

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Effect of Foliar Application of Boron on Yield and Yield Components of Linseed Cultivars (*Linum usitatissimum* L.)

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Abstract: Boron is a crucial micronutrient for the growth and development of plants. Foliar application of boron positively impacts the growth, yield, and yield components of oilseed crops. The objective of this study was to determine the effect of boron (B) on some yield and quality traits of linseed cultivars. The field experiments of this study were carried out in spring season of 2022 and 2023 under ecological conditions of Tekirdag. The experiment was carried out as a split plot design based on RCBD with three replications, in which cultivars constituted the main plot with three varieties (Karakız, Sarı Dane and Beyaz Gelin), and boron constituted the sub-plot with four doses (0, 100, 200 and 300 mg L^{-1}) in both years. Boron doses were applied by foliar spraying at 45 DAS. In the study plant height, branch number, number of capsules per plants, number of seeds per capsule, 1000 seed weight, seed yield, oil content and oil yield were investigated. Analysis of variance showed that boron application significantly affected all the examined traits except for 1000 seed weight and oil content. The results showed that the highest seed yield was obtained from 200 and 300 mg L^{-1} boron application (91.45 and 93.85 kg da^{-1}, respectively). However, the differences in oil content were not statistically significant in terms of cultivar and boron application. In conclusion, foliar application of appropriate doses of boron contributes to increase the yield of linseed, especially in medium and low input farming systems.

Key words: Boron, Linseed, Micronutrients, Quality, Yield

Yapraktan Bor Uygulamasının Keten (*Linum usitatissimum* L.) Çeşitlerinin Verim ve Verim Unsurları Üzerine Etkisi

Öz: Bor, bitki büyümesi ve gelişimi için önemli mikro besin elementlerinden birisidir. Borun yapraktan uygulanması yağlı tohumlarda tohum verimini ve verim unsurlarını olumlu yönde etkilemektedir. Bu çalışmada, farklı dozlarda bor uygulamasının keten çeşitlerinin verim ve bazı kalite özelliklerine etkilerinin belirlenmesi amaçlanmıştır. Çalışma, Tekirdağ ekolojik koşullarında 2022 ve 2023 yıllarında yazlık olarak iki yıl süreyle yürütülmüştür. Deneme Tesadüf Bloklarında Bölünmüş Parseller Deneme Desenine göre çeşitler ana parselde (Karakız, Sarı Dane ve Beyaz Gelin), bor dozları ise alt parselde (0, 100, 200 ve 300 mg L^{-1}) olacak şekilde 3 tekrarlamalı olarak kurulmuştur. Bor dozları bitkilere ekimden 45 gün sonra yapraktan püskürtme yoluyla uygulanmıştır. Çalışmada bitki boyu, dal sayısı, bitki başına kapsül sayısı, kapsüldeki tohum sayısı, 1000 tane ağırlığı, tohum verimi, yağ oranı ve yağ verimi gibi özellikler incelenmiştir. Varyans analizi sonucuna göre bor uygulaması 1000 tane ağırlığı ve yağ oranı hariç incelenen tüm özellikleri önemli ölçüde etkilemiştir. Araştırma sonuçlarına göre en yüksek tohum verimi 200 ve 300 mg L^{-1} bor uygulamasından elde edilmiştir (sırasıyla 91,45 ve 93,85 kg da^{-1}). Ancak yağ oranı arasındaki farklılıklar çeşit ve bor uygulaması bakımından istatistiki olarak önemli bulunmamıştır. Sonuç olarak uygun dozlarda borun yapraktan uygulanması, özellikle orta ve düşük girdili tarım sistemlerinde ketenin tohum veriminin artmasına katkıda bulunabilir.

Anahtar kelimeler: Bor, Kalite, Keten, Mikro Besin, Verim

1. Introduction

Although there are many oilseed crops in the world, the plants that are widely used in the vegetable oil industry today are soybean, rapeseed, cottonseed, peanut, sunflower, sesame, safflower, poppy, linseed, hemp, castor oil, jojoba, corn, olive, palm and coconut. All of these oilseed crops except jojoba, palm and coconut can be cultivated in Turkiye due to the ecological conditions they require (Vollmann & Rajcan, 2010; Onat et al., 2017). Linseed (*Linum usitatissimum* L.) is the only economically important crop species in the *Linaceae* family, which comprises 13 genera and approximately 300 species. The term *usitatissimum*, meaning "most useful," is used because of the linseed's long-standing significance as an agricultural crop (Chand & Fahim, 2008; Koçak & Bayraktar, 2011). It is grown either for its fiber (fiber flax) or for its oil (oilseed flax) (Hall et al., 2016). Tall, heavily branched varieties with strong fibers are cultivated for fiber production, while shorter, fewer branching varieties are grown for oil production (Yıldırım & Arslan, 2013).

Linseed contains 35-45% and oil 20-25% protein (Gill, 1987). Its oil is a rich source of omega-3 (ω-3, alpha-linolenic acid) which is important in human nutrition. Omega-3 contains approximately 50-55% of fatty acids (Bloedon & Szapary, 2004; Bayrak et al., 2010; Culpan & Gürsoy, 2023). Studies and researches continue for the development and popularization of linseed cultivation. Among these studies, micronutrient applications play an important role in increasing seed yield and quality. Boron is a crucial micronutrient for the growth and development of plants (Warington, 1923; Güneş et al., 2017). It participates in the process of pollination, fertilization and plant fruit setting. In addition, boron enhances the formation of adenosine triphosphate (ATP) and serves as a compound that accelerates the movement of sugars into the active areas during growth throughout the reproduction stages of the plant (Bolanos et al., 1996; Al-Juheishy, 2020).

It is known that when the boron level in the soil is 1 ppm, boron is sufficient, but when it exceeds 5 ppm, it has a toxic effect on the soil (Sahin, 2014). Many researchers have reported that in boron-deficient soils, boron application through soil and foliar spraying positively affects seed yield and yield components of plants (Katar et al., 2014; Rawashdeh & Sala, 2014; Mekki, 2015; Kurşun et al, 2016; Megha et al., 2023). These positive effects are especially evident in soils deficient in boron (< 1 ppm).

Boron content of 409 soil samples representing the sunflower cultivation areas of Thrace region showed a distribution between 0.14 mg kg^{-1} and 2.42 mg kg^{-1}. In 304 of the samples, boron content was 1 mg kg^{-1} and below. In 16% of the sunflower cultivation areas in Thrace region, boron level is insufficient and below 0.5 mg kg^{-1}, and boron fertilization in these areas in the amount and form recommended according to the results of soil analysis is necessary for both yield increase and crop quality (Kurşun et al., 2016). In general, 100-200 g da^{-1} boron is recommended for plants with low boron requirements such as cereals, while this rate can increase to 400 g for plants with high boron requirements such as sugar beet, rapeseed and sunflower. In foliar boron fertilization, the generally accepted rate is 250-300 mg L^{-1} (Güneş et al., 2017).

The aim of this study was to determine the effects of boron (B) on some yield and quality characteristics of linseed cultivars under Tekirdağ ecological conditions.

2. Materials and Methods

The research was conducted at the Area of Research and Experiment, Field Crops Department, Faculty of Agriculture, Tekirdag Namik Kemal University, during 2022 and 2023. The climatic and soil characteristics of the experimental area are given in Table 1 and Table 2. The climate characteristics of the research field are sufficient and suitable for linseed (Table 1), but when the soil properties are examined, it is seen that it is insufficient in terms of boron. (Table 2).

In the study, Karakız, Sarı Dane and Beyaz Gelin cultivars obtained from the Trakya Agricultural Research Institute (Edirne) were used as material. These cultivars are oilseed flax types, with seed yield values ranging from 80 to 100 kg da^{-1} and oil content between 30% and 35% (Anonymous, 2020).

The experiment was arranged in a split plot with randomized complete block design in three replications, in which cultivars constituted the main plot with three varieties (Karakız, Sarı Dane and Beyaz Gelin), and boron constituted the sub-plot with four doses (0, 100, 200 and 300 mg L^{-1}) in both years. Boron is obtained from water-soluble 8% w/w Boron Ethanolamine (Boron-8, Gubretas). Boron doses were applied by foliar spraying 45 days after sowing (at 45 DAS) (Alam et al., 2020; Singh et al., 2020).

Table 1. The climatic conditions of the experimental area and long-term averages
Çizelge 1. Deneme alanının iklim verileri ve uzun yıllar ortalamaları

Month	Mean temperature (°C)			Total rainfall (mm)			Mean humidity (%)		
	2022	2023	LTA	2022	2023	LTA	2022	2023	LTA
March	5.2	9.3	7.7	9.5	48.4	49.8	71.5	81.2	80.6
April	12.7	12.0	11.9	70.6	93.5	42.8	74.3	82.3	78.2
May	16.9	15.8	16.7	15.7	27.6	39.1	75.2	80.3	77.1
June	22.5	21.2	21.3	32.5	16.3	39.2	74.5	77.5	73.9
July	24.3	25.7	23.9	1.5	15.2	26.5	68.8	71.2	70.4
Mean/Total	16.3	16.8	16.3	129.8	201.0	197.4	72.8	78.5	76.0

* Tekirdag Meteorological Station Data, LTA: long-term averages (1972-2022)

Table 2. The soil characteristics of the experimental area
Çizelge 2. *Deneme alanının toprak özellikleri*

Variable	Unit	2022	2023	Assessment
pH	-	7.17	7.22	medium
Salt	%	0.02	0.02	none
Organic matter	%	1.06	1.15	low
Phosphorus	kg da^{-1}	6.4	8.7	low
Potassium	kg da^{-1}	96.6	102.5	high
Boron (B)	ppm	0.49	0.55	low
Texture	-	Clay	Clay	-
Depth	cm	0-20	0-20	-

Nitrogen and phosphorous were applied at the rate of 80 kg ha^{-1} N (urea) and 60 kg ha^{-1} P (Diammonium phosphate), respectively. All the phosphorus and half of the nitrogen were applied before sowing the seeds, while the remaining nitrogen was applied during the stem elongation stage (about one month after sowing). Ten plants per plot were randomly selected to determine yield and yield components. In the study plant height (cm), branch number (pcs), number of capsules per plants (pcs), number of seeds per capsule (pcs), 1000 seed weight (g), seed yield (kg da^{-1}), oil content (%) and oil yield (kg da^{-1}) were investigated. The linseed plants were harvested by hand on August 2, 2022, and July 17, 2023. For the seeds from each plot, the oil content (as a percentage) was measured using a Nuclear Magnetic Resonance (NMR) device. The oil yield was calculated by multiplying the seed yield by the oil content of the seeds (Abou Chehade et al., 2022).

The analysis of variance was carried out over two years for the values of all traits. The data obtained were analyzed according to randomized complete block design. The means were compared by using the Least Significant Difference (LSD) test at the 0.05 and 0.01 levels as described by Steel and Torrie (1980) with MSTAT-C statistical software (MSTAT, 1989).

3. Results and Discussion

The variance analysis results of this study, which was conducted to determine the effects of boron applications on the seed yield, yield components, oil content, and oil yield of linseed under field condition, are given in Table 3.

Cultivars had a significant effect on 1000 seed weight at the 1% level, and on branch number, seed yield, and oil yield at the 5% level. In terms of applied doses, all the examined traits, except for 1000 seed weight and oil content, showed statistically significant differences at the 1% level. Cultivar × Boron doses interactions had significant effects ($p<0.05$) on branch number and 1000 seed weight (Table 3).

In terms of plant height, the lowest plant height was observed in the control plots where boron was not applied (38.08 cm). In the cultivar × boron interaction, plant height values varied between 36.07-45.12 cm, but this difference was not found statistically significant ($p>0.05$). Zahoor et al. (2011) reported that plant height significantly increased when boron levels increased in sunflower. Alam et al. (2020) showed that treating linseed with a foliar application of $ZnSO_4$ at 0.5% combined with Borax at 0.3% at 45 days after sowing resulted in significantly higher plant heights (53.75 cm and 55.14 cm) compared to the control (42.18 cm and 43.53 cm). Beyzi et al. (2019) found that the highest plant height (58.18 cm) in fenugreek was obtained from 800 mg L^{-1} boron application and reported that plant height increased with boron applications compared to control applications. Our results confirm the findings of earlier researchers. In addition, plant height, which is much more important in fiber flax varieties and desired to be longer, is a character that directly affects seed yield to a certain extent in oilseed flax varieties, provided that side branches are formed (Culbertson, 1954; Çöl Keskin et al., 2020).

In boron application, the branch number increased significantly with increasing boron doses and the highest branch number was obtained from 300 mg L^{-1} boron application with 8.65 pcs (Table 4). In the cultivar × boron interaction, the highest branch number was obtained from Sarı Dane cultivar with 300 mg L^{-1} boron application and Beyaz Gelin cultivar with 200 and 300 mg L^{-1} (8.92, 8.39 and 8.78 pcs, respectively).

Table 3. Analysis of variance for some agronomical and technological traits of linseed
Çizelge 3. *Ketenin bazı agronomik ve teknolojik özelliklerine ilişkin varyans analizi*

SV	DF	Plant height	Branch number	Number of capsules per plants	Number of seeds per capsule	1000 seed weight	Seed yield	Oil content	Oil yield
Cultivar (C)	2	0.7934^{ns}	7.7444*	1.4571^{ns}	2.7942^{ns}	28.9777**	7.3128*	4.2125^{ns}	7.8346*
Error$_1$	4								
Boron (B)	3	11.8957**	28.7479**	15.7711**	8.5118**	1.8097^{ns}	18.1866**	0.6244^{ns}	16.1485**
C x B	6	0.4093^{ns}	2.8136*	0.4371^{ns}	0.4340^{ns}	2.8908*	0.6726^{ns}	0.2710^{ns}	0.5273^{ns}
Error	18								
CV (%)		5.46	4.09	17.32	5.71	2.26	5.56	1.15	6.05

SV: source of variation, DF: degree of freedom, ns: not significant, *, ** significant at 0.05 and 0.01 probability levels, respectively

A high number of branches in linseed is a desirable character as it contributes to the increase in the number of capsules per plant, especially in oilseed flax varieties (Endes, 2010). Karaaslan and Tonçer (2001) determined the number of branches per plant as 4.3-6.6 as a result of a study conducted in Diyarbakır ecological conditions using 11 different linseed varieties as winter crops. Kurt et al. (2015) reported that the branch number per plant of 30 linseed lines varied between 4.4-7.4 pcs in Samsun ecological conditions. Al-Doori (2021) determined the branch number in linseed as 9.06-11.54 pcs and reported that the branch number increased with increasing boron levels. Our results fell within the ranges reported by these researchers.

The number of capsules per plants in linseed is low in frequent sowing and high in infrequent sowing (Uzun, 1992; Saeidi, 2002). In boron application, the number of capsules per plants increased significantly with increasing boron doses and the lowest number of capsules per plants was observed in the control plots where boron was not applied (11.20 pcs). In the cultivar × boron interaction, number of capsules per plants values varied between 10.00-23.40 pcs, but this difference was not found statistically significant ($p>0.05$). Studies have shown that number of capsules per plants in linseed varies between 16.4 and 22.2 pcs (Gür, 1998), 14.1 and 29.0 pcs (Akçalıcan et al., 2003), 25.6 and 47.1 pcs (Kurt et al., 2015), 14.07 and 19.07 pcs (Arslan & Culpan, 2021). Al-Doori (2021) reported that increasing foliar application of boron significantly enhanced number of capsules per plant. Beyzi et al. (2019) found that greatest number of pods per plant (8.63 pods plant^{-1}) in fenugreek was obtained from 800 mg L^{-1} boron application and reported that plant height increased with boron applications compared to control applications. In previous studies, it was reported that the number of pods per plant increased with boron applications in chickpea (Ceyhan et al., 2007), common bean (Harmankaya et al. 2008), peanut (El-Kader & Mona, 2013) and sesame (Khuong et al., 2022) plants compared to control treatments. The present findings were similar with the results of previous researchers.

The number of seeds per capsule was significantly affected by boron application and highest number of seeds per capsule was obtained from 300 mg L^{-1} boron application (8.49 pcs). Kurt et al. (2015) and Yıldırım & Arslan (2013) reported that the number of seeds per capsule in linseed varied between 6.40-8.10 and 7.98-9.03 pcs, respectively. Alam et al. (2020) emphasize that the significantly higher number of seeds per capsule (6.00 and 8.57 pcs) was observed with the foliar application of $ZnSO_4$ at 0.5% and Borax at 0.3% at 45 days after sowing (DAS), compared to the control.

Table 4. Some agronomical and technological traits of linseed and LSD groups
Çizelge 4. *Ketenin bazı agronomik ve teknolojik özelliklerine ait ortalama değerler ve önemlilik grupları (EKÖF)*

		Plant height (cm)	Branch number (pcs)	Number of capsules per plants (pcs)	Number of seeds per capsule (pcs)	1000 seed weight (g)	Seed yield (kg da^{-1})	Oil content (%)	Oil yield (kg da^{-1})
Cultivar (C)									
Karakız		43.23	7.68 **b**	15.32	7.78	7.21 **a**	105.19 **a**	35.96	37.80 **a**
Sarı Dane		41.75	7.91 **ab**	19.32	7.38	6.57 **b**	71.72 **ab**	36.02	25.83 **ab**
Beyaz Gelin		40.12	8.35 **a**	16.38	8.58	6.18 **b**	84.36 **b**	35.46	29.90 **b**
Boron Doses (B)									
0		38.08 **b**	7.47 **c**	11.20 **b**	7.42 **b**	6.57	78.75 **b**	35.66	28.09 **b**
100		41.47 **a**	7.51 **c**	19.07 **a**	7.93 **ab**	6.63	84.30 **b**	35.84	30.19 **b**
200		43.35 **a**	8.29 **b**	18.32 **a**	7.82 **b**	6.69	91.45 **a**	35.84	32.78 **a**
300		43.88 **a**	8.65 **a**	19.44 **a**	8.49 **a**	6.72	93.85 **a**	35.91	33.65 **a**
C x B									
Karakız	0	39.72	6.94 **d**	10.00	7.13	7.08 **b**	95.50	35.89	34.31
	100	43.53	7.30 **d**	17.60	7.80	7.21 **ab**	100.76	35.95	36.17
	200	45.12	8.23 **bc**	16.66	7.73	7.43 **a**	111.42	36.10	40.18
	300	44.54	8.25 **bc**	17.00	8.47	7.12 **b**	113.09	35.90	40.56
Sarı Dane	0	38.46	7.29 **d**	12.20	6.80	6.56 **cd**	65.99	35.72	23.57
	100	41.45	7.19 **d**	20.73	7.33	6.59 **cd**	70.67	36.09	25.48
	200	42.30	8.24 **bc**	20.97	7.40	6.40 **de**	73.94	36.04	26.65
	300	44.76	8.92 **a**	23.40	8.00	6.75 **c**	76.26	36.22	27.62
Beyaz Gelin	0	36.07	8.19 **c**	11.40	8.33	6.07 **f**	74.76	35.36	26.40
	100	39.42	8.05 **c**	18.87	8.67	6.10 **f**	81.47	35.47	28.91
	200	42.62	8.39 **abc**	17.33	8.33	6.25 **ef**	89.00	35.40	31.52
	300	42.35	8.78 **ab**	17.93	9.00	6.29 **ef**	92.22	35.62	32.76
Mean		**41.69**	**7.98**	**17.01**	**7.91**	**6.65**	**87.09**	**35.81**	**31.17**

Means followed by the same letter within columns are not significantly different at the level of $p < 0.05$

Cultivars had significant effects on 1000 seed weight ($p<0.01$) and its highest was obtained from Karakız cultivar (7.21 g). In the cultivar × boron interaction, the highest 1000 seed weight was obtained from Karakız cultivar with 100 and 200 mg L^{-1} boron application (7.21 and 7.43 g, respectively). In oilseeds, 1000 seed weight refers to the fullness and size of the seeds. Therefore, it is very important in terms of both seed yield and oil content (Vollman & Rajcan, 2009; Tunçtürk & Tunçtürk, 2021). Studies have shown that 1000 seed weight in linseed varies between 7.10-7.60 g (Rokade et al., 2015), 7.87-9.30 g (Maurya el al. 2017) and 5.04-5.77 g (Arslan & Culpan, 2021). Alam et al. (2020) found that the significantly higher 1000 seed weight (7.59 g and 7.64 g) was observed with the foliar application of $ZnSO_4$ at 0.5% and Borax at 0.3% at 45 days after sowing, compared to the control. Saeed et al. (2015) emphasized that foliar boron application increased 1000-achene weight (42.74 g) in sunflower and the lowest value (38.63 g) was obtained in the control treatment. Gunnes et al. (2003) and Khan et al. (2006) found that boron application significantly increased 1000 grain weight in wheat compared to control.

Cultivars had significant effects on seed yield ($p<0.05$) and Karakız cultivar had the highest value with 105.19 kg da^{-1} (Table 4). The seed yield was significantly affected by boron application and the highest seed yield was obtained from 200 and 300 mg L^{-1} boron application (91.45 and 93.85 kg da^{-1}, respectively). In the cultivar × boron interaction, seed yield values varied between 65.99-113.09 kg da^{-1}, but this difference was not found statistically significant ($p>0.05$). Seed yield in linseed is closely related to its yield components. Yield components are affected by both genetic and environmental conditions. The seed yield is positively correlated with the number of capsules per plant, number of seeds per capsule and 1000 seed weight (Çopur et al., 2006; Ibrar et al., 2016). In oilseed flax, seed yield was reported by Tunçtürk (2007) 99.7-149.0 kg da^{-1}, Tanman (2009) 118.6-175.6 kg da^{-1}, Endes (2010) 65.30-124.10 kg da^{-1}, Katar et al. (2023) 109.0-212.0 kg da^{-1}. Alam et al. (2020) found that foliar application of 0.5% $ZnSO_4$ with 0.3% Borax 45 days after sowing significantly increased seed yield in both years compared to the control (129.9 and 133.5 kg da^{-1}). Bungla et al. (2021) reported that boron application significantly increased seed yield in linseed and that the positive interaction between boron and sulfur significantly and synergistically affected seed yield of linseed. The high doses of boron significantly increased seed yield in safflower and the highest seed yield were obtained in 500 g da^{-1} boron application (Katar et al., 2014). The results confirm the findings of earlier researchers.

Cultivars and boron applications did not affect the oil content statistically significantly ($p>0.05$). Although the highest oil content was obtained from Sarı Dane cultivar of 300 mg L^{-1} boron application (36.22%), this value was not found to be statistically significant. Yıldırım & Arslan (2013) and Arslan & Culpan (2021) reported that the oil content in linseed varied between 29.83-34.08% and 33.76-36.15%, respectively. In addition, some researchers found that different boron doses relatively increased the oil content in oilseeds, but this was not statistically significant (Nasef et al., 2006; Katar et al., 2014). These results are consistent with our findings. Differences in oil content among the cultivars examined can be attributed to the genetic and genomic diversity present in their overall genetic make-up (Ibrar et al., 2016).

Cultivars had significant effects on oil yield ($p<0.05$) and Karakız cultivar had the highest value with 37.80 kg da^{-1} (Table 4). The seed yield was significantly affected by boron application and the highest seed yield was obtained from 200 and 300 mg L^{-1} boron application (32.78 and 33.65 kg da^{-1}, respectively). In the cultivar × boron interaction, seed yield values varied between 23.57-40.56 kg da^{-1}, but this difference was not found statistically significant ($p>0.05$). Oil yield is calculated based on seed yield and oil content (Cvejić et al., 2023). The increase in seed yield or oil content directly affects oil yield. Boron applications positively affected seed yield (Rawashdeh & Sala, 2014; Beyzi et al., 2019; Bungla et al., 2021; Khuong et al., 2022) and therefore oil yield was also positively affected. The oil yield values we obtained as a result of the research were within the limits specified by Endes (2010) 21.9-39.6 kg da^{-1}, Lemessa and Zerihun (2022) 42.20-69.59 kg da^{-1}, Katar et al. (2023) 37.0-77.0 kg da^{-1}.

4. Conclusion

Application of boron at a concentration of 200 and 300 mg L^{-1} at 45 DAS helped to increase yield and quality parameters of plant height, branch number, number of capsules per plants, number of seeds per capsule, seed yield and oil yield of linseed. The positive effect of boron applications on yield components led to an increase in seed yield. In particular, seed yield increased by 16.12% and 19.17% at 200 and 300 mg L^{-1} doses, respectively, compared to the control. Therefore, the increase in seed yield directly affected the oil yield. In conclusion, foliar application of appropriate doses of boron contributes to increase the yield of linseed, especially in medium and low input farming systems.

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Identification of Beneficial Bacteria in Rosemary Rhizospheres and Determination of Plant Growth Promoting (PGP) Potential

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Abstract: Plant growth-promoting rhizobacteria (PGPR), which colonize the rhizosphere, are eco-friendly and beneficial bacteria that directly or indirectly promote plant growth. In this study, 13 isolates from the rhizosphere of *Rosmarinus officinalis* L. (Rosemary) were identified using MALDI-TOF-MS to assess morphology, biochemistry, and plant growth-promoting traits and to evaluate their antagonistic effects against *Fusarium oxysporum*. Among all isolates, 9 isolates fixed nitrogen, 8 isolates dissolved inorganic phosphate, 8 isolates produced siderophores, 7 isolates produced IAA (Indole-3-Acetic Acid), and 6 isolates produced HCN. Isolate BBR-6 showed the highest antifungal activity against *Fusarium oxysporum*, with an inhibition rate of 61.54 %. The isolate BBR-10 (19.40 %) showed the weakest effect against *F. oxysporum*. Although research on PGPRs has increased recently, research on rosemary is still limited. This study aimed to identify the local bacterial community in the rhizosphere of rosemary and assess its growth-promoting properties and antifungal activity against disease-causing *F. oxysporum*, potentially acting as a microbial fertilizer and biocontrol agentents.

Key words: Rosemary, *Fusarium oxysporum*, MALDI TOF MS, microbial fertilizer, PGPR

Biberiye Rizosferindeki Yararlı Rizobakterilerin Tanımlanması ve Bitki Gelişimini Teşvik Edici Özelliklerinin Belirlenmesi

Öz: Bitki büyümesini teşvik eden rizobakteriler (PGPR), rizosferde kolonize olan, doğrudan ya da dolaylı olarak bitki büyümesini destekleyen çevre dostu faydalı bakterilerdir. Bu çalışmada, *Rosmarinus officinalis* L. (Biberiye) rizosferinden on üç izolat MALDI-TOF-MS methodu ile tanımlanarak morfolojik, biyokimyasal, bitki büyümesini teşvik edici özellikleri ile *Fusarium oxysporum*'a karşı antagonistik özellikleri değerlendirildi. Tüm izolatlar arasında 9 izolatın azot fikse ettiği, 8 izolatın norganik fosfatı çözdüğü, 8 izolatın siderofor ürettiği, 7 izolatın IAA (indole-3-acetic acid) ürettiği ve 6 izolatın HCN ürettiği belirlendi. BBR-6 izolatı, *Fusarium oxysporum*'a karşı % 61.54'lük bir inhibisyon oranıyla en yüksek antifungal aktiviteyi gösterdi. BBR-10 izolatı ise % 19.40 ile *F. oxysporum*'a karşı en zayıf etkiyi gösterdi. PGPR'ler üzerindeki araştırmalar son zamanlarda artsa da biberiye üzerine yapılan araştırmalar hala sınırlıdır. Bu çalışma, biberiye rizosferindeki yerel bakteri topluluğunu tanımlamayı, bitki büyümesini teşvik edici özelliklerini, mikrobiyal gübre ve biyokontrol ajan potansiyeli ile biberiye bitkisinde kök hastalığa neden olan *F. oxysporum*'a karşı antifungal aktivitesini değerlendirmeyi amaçlamaktadır.

Anahtar Kelimeler: Biberiye, *Fusarium oxysporum,* MALDI TOF MS, mikrobiyal gübre, PGPR

1. Introduction

Rosmarinus officinalis L. (Rosemary), belonging to the Lamiaceae family, is an important medicinal and aromatic plant that can grow up to 3 meters tall. *Rosmarinus officinalis* L. which grows naturally in many Mediterranean countries, has been used for various medical and culinary purposes since ancient times. The essential oils in rosemary contain different chemical compounds such as alcohol, hydrocarbons, phenols, aldehydes, esters, and ketones. It has been indicated that the essential oils obtained from rosemary flowers and leaves are widely used to treat conditions such as asthma, cataracts, rashes, headaches, indigestion, and renal colic (Hammer & Junghanns, 2020). Chemical fertilizers are harmful to soil and ecosystems, particularly human health. Excessive use of chemical fertilizers causes significant soil pollution and decreases crop yield. Therefore, using eco-friendly natural fertilizers instead of chemical ones in growing rosemary is crucial for sustainable farming. PGPR are bacteria colonizing the plant's rhizosphere and directly or indirectly supporting plant growth. PGPRs utilize soil nutrients for plant growth, produce many regulators, protect plants from phytopathogens, improve soil structure, and reduce harmful compounds like pesticides. Therefore, rhizobacteria are crucial microorganisms for soil fertility and sustainable crop production (Rochlani et al., 2022). Although research on PGPRs has increased recently, it is still limited. Recent studies suggest that developing microbial formulations with local isolates showing activity in different ecosystems and plant species should be increased. Therefore, developing microbial fertilizers that protect the environment and comply with sustainable agriculture can significantly reduce chemical fertilizer use. This study aimed to isolate, identify, and determine the plant growth-promoting properties of rhizobacteria associated with *Rosmarinus officinalis* used in many fields, especially in medicine and pharmacy.

2. Material and Method

2.1. Sample collection

Rhizospheric soil samples were collected in May 2023 from *R. officinalis* in the Medicinal Plants Garden of the Department of Field Crops of Ankara University Faculty of Agriculture (39°57'44.2"N, 32°51'36.7"E).

2.2. Isolation of rhizobacteria

Rhizospheric bacteria were isolated from 1 g of dried soil samples by serial dilution method. Each rhizospheric soil sample was diluted from 10^{-1} to 10^{-6}. These dilutions were spread on nutrient agar (NA) solidified Petri dishes and incubated at 28 °C. After incubation, different colonies were selected and planted in Petri dishes containing NA medium until a pure colony was obtained.

2.3. Identification of bacterial Isolates

Biochemical and morphological characterization of isolates

Physiological, biochemical tests and Gram staining of the bacterial isolates were examined using methods described by Palleroni et al. (1984).

2.3.1. Identification of isolates

MALDI-TOF MS was used for bacterial identification by analyzing unique molecular fingerprints with the MALDI Biotyper System (Sivri & Öksüz, 2019).

2.4. Plant growth promoting (PGP) properties

2.3.1. Determination of nitrogen fixation ability

Determination of the nitrogen fixation abilities of the isolates was done according to the protocol specified by Wilson and Knight (1952) and Park et al. (2005). The nitrogen fixation activity was assigned three time intervals (+++: development after 6 hours, ++: development after 12 hours, and +: development after 24 hours).

2.3.2. Evaluation of siderophore production

Chrome Azurol S (CAS) agar media was used to identify the production of siderophores. (Schwyn & Neilands, 1987). The siderophore activity was assigned three time intervals (+++: color change after 12 h, ++: color change after 24 h, +: color change after 36 h).

2.3.3. Determination of HCN-producing isolates

The HCN production assay was carried out according to Bakker and Schippers (1987). The HCN activity was assigned three time intervals (+++: color change after 6 h, ++: color change after 12 h, +: color change after 24 h).

2.3.4. Assessment of indole-3-acetic acid (IAA) production

The Sarwar and Kremer (1995) protocol was used to assess the isolates' capacity to produce IAA. The IAA activity was assigned three time intervals (+++: color change after 1 h, ++: color change after 6 h, and +: color change after 12 h).

2.3.5. Determination of inorganic phosphate dissolving capacity

The inorganic phosphate dissolving capacities of the isolates were determined according to the protocol described by Mehta and Nautiyal (2001).

2.5. Assessment of antifungal activity

The antifungal activity of the isolates was tested against *Fusarium oxysporum* using the dual culture method (Landa et. al., 1997). The mycelial growth diameter of each phytopathogen was measured according to the method described by Royse and Ries (1978) to determine the percentage of fungal inhibition (FI). The experiments were conducted in triplicates.

$$FI (\%) = (B-M) / B \times 100$$

Where B is the average diameter of mycelial development without bacterial isolate.

M is the mean diameter of mycelial growth in the presence of the bacterial isolate.

2.6. Data analysis

Data for antifungal activity and IAA production were analyzed using JMP Pro 17.0 statistical software, based on three replicates. Dependant variables with normal distribution were shown as mean ± standard deviation (SD).

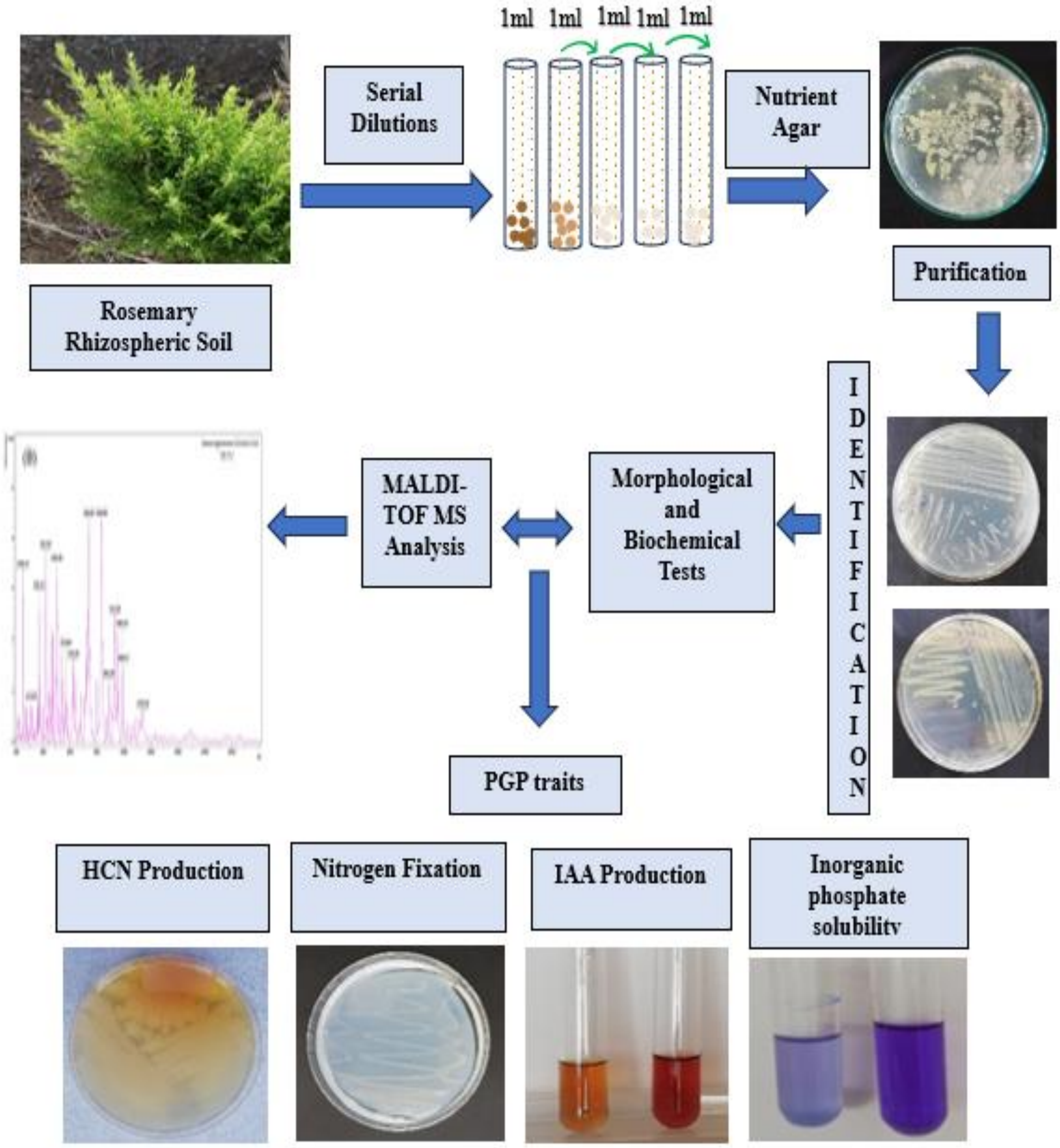

Figure 1. Flow chart for identification of isolates in Rosemary rhizospheric soil samples
Şekil 1. *Biberiye rizosferik toprak örneklerinde izolatların tanımlanmasına yönelik akış şeması*

3. Results and Discussion

3.1. Identification of isolates

In this study, 13 isolates were identified from Rosemary's rhizospheric soil samples using the MALDI-TOF MS method; these include 6 Bacillus (BBR-1, BBR-2, BBR-3, BBR-6, BBR-9, BBR-13), 3 Pseudomonas (BBR-7, BBR-8, BBR-12), 2 Lactobacillus (BBR-5, BBR-11), 1 Pantoea (BBR-4) and 1 *Pseudarthrobacter* (BBR-10). Some morphological and biochemical characteristics of rhizobacterial isolates are given in Table 1.

MALDI-TOF MS is an extremely useful tool for identifying bacteria at the genus, species, and strain levels. In previous studies, many researchers used the MALDI-TOF MS method to identify bacteria (Nazir et al., 2020; Tamura, 2023). Recently, this method has gained popularity due to its high accuracy and rapid results. Martínez-Hidalgo et al. (2021) identified endophytic bacteria from canola roots using the MALDI TOF MS method. Similarly, Öksel et al. (2022) used the MALDI-TOF MS method to identify bacteria in wheat rhizosphere. The findings of this study revealed that *Bacillus* (46%) *Pseudomonas* (23%) and were the most common bacterial genera in the Rosemary rhizosphere.

Table 1. Morphological and biochemical traits of rhizobacterial isolates
Çizelge 1. *Rizobakteriyel izolatların morfolojik ve biyokimyasal özellikleri*

Isolates No	MALDI-TOFMS results	Gram Stain Test	Motility	Colony color	Biochemical	Characteristics	
					Catalase	Oxidase	KOH %3
BBR-1	*Bacillus thuringiensis*	+*	+	whitish	+	-	-
BBR-2	*Bacillus pumilus*	+	+	cream	+	**	-
BBR-3	*Bacillus megaterium*	+	+	white	+	-	-
BBR-4	*Pantoea agglomerans*	-	+	light yellow	+	-	+
BBR-5	*Lactobacillus paracasei*	+	-	white	-	-	-
BBR-6	*Bacillus mojavensis*	+	+	white	+	+	-
BBR-7	*Pseudomonas libanensis*	-	+	light yellow	+	+	+
BBR-8	*Pseudomonas chlororaphis*	-	+	white	+	+	+
BBR-9	*Bacillus cereus*	+	+	white	+	-	-
BBR-10	*Pseudarthrobacter scleromae*	+	-	white	+	-	-
BBR-11	*Lactobacillus oligofermentans*	+	-	white	-	-	-
BBR-12	*Pseudomonas fluorescens*	-	+	white	+	+	+
BBR-13	*Bacillus simplex*	+	+	cream	+	-	-

Note: * +, positive; –, negative ** Not detected

3.2. Plant growth-promoting properties of isolates

In the current study, 13 isolates were screened for siderophores, phosphate solubility, nitrogen fixation, IAA, and HCN production abilities. Among all isolates, 69% (BBR-2, BBR-3, BBR-4, BBR-7, BBR-8, BBR-9, BBR-10, BBR-12, BBR-13) fixed nitrogen, 46% (BBR-1, BBR-5, BBR-6, BBR-9, BBR-11, BBR-12) produced HCN, 61% (BBR-2, BBR-3, BBR-4, BBR-6, BBR-7, BBR-8, BBR-12, BBR-13) dissolved inorganic phosphate, 61% (BBR-2, BBR-4, BBR-6, BBR-8, BBR-9, BBR-10, BBR-12, BBR-13) produced siderophores, and 54% (BBR-1, BBR-4, BBR-6, BBR-7, BBR-9, BBR-10, BBR-12) produced IAA. Siderophores are low-molecular-weight, iron-chelating organic substances produced by many rhizospheric bacteria. The majority of Pseudomonas and Bacillus strains in the rhizosphere can produce siderophores (Joseph et al., 2007). Rudakova et al. (2023) reported that *Bacillus pumilus* strain 3-19, which has plant growth-promoting properties, produces siderophores. In the present study, *B. pumilus* BBR-2, *B. mojavensis* BBR-6, and *B. cereus* BBR-9 produced siderophore. Many studies are showing that *Pseudomonas* sp. produces siderophores (Saranraj et al., 2023; Clericuzio et al., 2024). Subramanium & Sundaram (2020) reported that *P. fluorescens* PSF02 isolated from agricultural soils also produced siderophores. In the current study, *P. fluorescens* BBR-12 produced siderophore. Wei et al. (2023) reported that rhizospheric *Pseudomonas chlororaphis* IRHB3 produced siderophore. Interestingly, In the current study, *P. chlororaphis* BBR-8 produced siderophore. Previous studies have shown that *Pantoeae agglomerans* also produced siderophores (Hynes et al., 2008; Shariati et al., 2017). Similarly, In the present study, *P. agglomerans* BBR-4 produced siderophore. Nitrogen is a vital element used in many structural and functional processes in the cell. There are a lot of studies that prove bacteria in the roots of plants like rosemary (Stamenov et al. 2021) and oregano (Loera-Muro et al. 2021) can fix nitrogen. *Bacillus* spp., about which a great deal of research has been conducted, are among the most abundant bacteria in soil. According to the available literature, *B. pumilus* (Agake et al. 2021), and *B. subtilis* (Sharma et al. 2021) strains were identified to exhibit nitrogen fixation. In the present study, *B. pumilus* BBR-2 *and B. megaterium* BBR-3 fixed nitrogen. Singh et al., (2023) determined that *P. koreensis* CY4 isolated from sugarcane rhizosphere fixed nitrogen. Shi et al., (2023) reported that *Pseudomonas* sp. in the rhizosphere of *A. mongolicus*, a Chinese medicinal plant, fixed nitrogen. Similarly, In the current study, *P. libanensis* BBR-7 *and P. chlororaphis* BBR-8 fixed nitrogen. According to Sharma et al. (2021), 80% of bacteria isolated from the rhizosphere can produce IAA. According to spectrophotometric analysis, the highest IAA production was obtained in *B. mojavensis* BBR-6 with 18.37 $\mu g/mL^{-1}$ in line with the present study. This was followed by *B. cereus* BBR-9 with 17.89 $\mu g/mL^{-1}$ and *P. agglomerans* BBR-4 with 17.66 $\mu g/mL^{-1}$, respectively. Stamenov et al. (2021) reported that *Pseudomonas* sp. P42 strain in rosemary rhizosphere produced IAA. Similarly, in this study, *P. fluorescens* BBR-12, and *P. libanensis* BBR-7 produced IAA. In a recent study, Patel et al. (2024) reported that rhizospheric *P. chlororaphis* did not produce IAA. Likewise, in this study, *P. chlororaphis* BBR-8 did not produce IAA. Khatami et al. (2023) reported that rhizospheric *Bacillus* sp. synthesized high amounts of IAA. Similarly, In the current study, *B. thuringiensis* BBR-1, *B. mojavensis* BBR-6, and *B. cereus* BBR-9

Table 2. Plant growth promoting traits of the rhizobacterial isolates
Çizelge 2. *Rizobakteriyel izolatların bitki gelişimini teşvik edici özellikleri*

Isolates	PGP traits					OD at 530 nm Mean ± SD
	Inorganic phosphate solubization	Nitrogen fixation	Siderophore production	HCN production	IAA production	
Bacillus thuringiensis BBR-1	-	-	-	+++*	+	8.42 ± 0.07^{d}
Bacillus pumilus BBR-2	+	++**	+	-	-	$2.33\pm0.22^{\text{hı}}$
Bacillus megaterium BBR-3	+	+++	-	-	-	3.81 ± 0.37^{f}
Pantoea agglomerans BBR-4	+	+++	+++***	-	+++****	17.66 ± 0.67^{a}
Lactobacillus paracasei BBR-5	-	-	-	+	-	$3.06\pm0.19f^{gh}$
Bacillus mojavensis BBR-6	+	-	+	+	+++	18.37 ± 0.08^{a}
Pseudomonas libanensis BBR-7	+	++	-	-	+	6.56 ± 0.29^{e}
Pseudomonas chlororaphis BBR-8	+	++	+	-	-	$2.18\pm0.05^{\text{ı}}$
Bacillus cereus BBR-9	-	+	+	+	+++	17.89 ± 0.09^{a}
Pseudarthrobacter scleromae BBR-10	-	+	+	-	++	11.55 ± 0.33^{b}
Lactobacillus oligofermentans BBR-11	-	-	-	+	-	3.27 ± 0.26^{fg}
Pseudomonas fluorescens BBR-12	+	++	+++	++	++	10.51 ± 0.12^{c}
Bacillus simplex BBR-13	++	+	+	-	-	$2.79\pm0.14^{\text{ghı}}$

* The color changes for HCN activity are as follows: +++: after 6 hours, ++: after 12 hours, +: after 24 hours.**Nitrogen fixation activity (+++: development after 6 h, ++: development after 12 h, +: development after 24 h).*** The color changes for siderophore activity are as follows: +++: color change after 12h, ++: color change after 24h., +: color change after 36h.**** The color changes for IAA activity are as follows: +++: colour change after 1h, ++: colour change after 6h, and +: colour change after 12h. For OD at 530 nm: $p<0,001$; statistically significant level. a-ı: The difference between the means shown by different letters in the same column is statistically significant. (Mean ± SD: Mean±Standard Deviation)

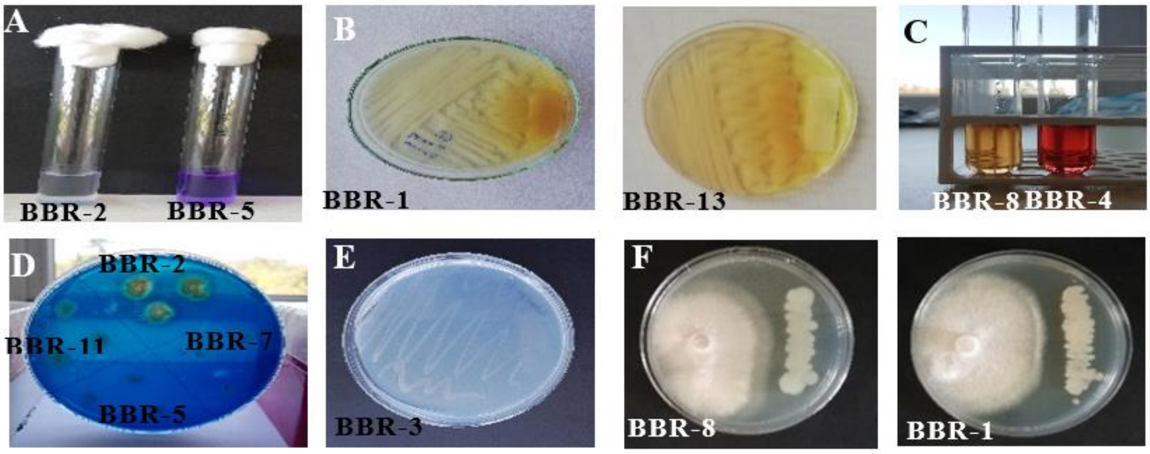

Figure 2. PGPR and antifungal activity test images of rhizobacterial isolates (A: Inorganic phosphate solubization B: HCN production C: IAA production D: Siderophore production E: Nitrogen fixation F: Antifungal test of isolates against *F. oxysporum*)
Şekil 2. *Rizobakteriyel izolatların PGPR ve antifungal aktivite testi görüntüleri (A: İnorganik fosfat çözünümü B: HCN üretimi C: IAA üretimi D: Siderophore üretimi E: Azot fiksasyonu F: İzolatların F. oxysporum'a karşı antifungal testi)*

produced IAA. According to Anderson and Kim (2018), HCN produced by *Pseudomonas* and *Bacillus* strains is an important factor that protects the plant against phytopathogens. Ahmad et al. (2008) determined that among rhizospheric bacteria, 50% of *Bacillus* isolates and 80% of *Pseudomonas* isolates were positive for HCN production. Singh et al. (2019) determined that *B. thuringiensis* SF 23, *P. aeruginosa* SF 44, *B. subtilis* SF 48, and *B. subtilis* SF 90 isolate produced HCN. Stamenov et al. (2021) determined that *Pseudomonas* and *Bacillus* obtained from rosemary rhizosphere produced HCN. Kumar et al. (2021) reported that *Bacillus pumilus* strain JPVS11 in the rice rhizosphere produced HCN. Halimursyadah et al. (2023) reported that *P. fluorescens* produced HCN among the 37 isolates from the patchouli rhizosphere. Similarly, In the current study, *P. fluorescens* BBR-12 also produced HCN. Bacteria like *Pseudomonas* and *Bacillus* in the rhizosphere employ various mechanisms to dissolve phosphate and release it into the soil. According to Rawat et al. (2021), the most prevalent inorganic phosphate-solubilizing bacteria in the rhizosphere are *Bacillus*, *Enterobacter*, and *Pseudomonas*. Similarly, in the current study, *B. pumilus* BBR-2, *B. megaterium* BBR-3, *B. mojavensis* BBR-6, *B. simplex* BBR-13 dissolved inorganic phosphate. Sharma et al. (2021) reported that *Pseudomonas libanensis* HB4N3 strain has high inorganic phosphate solubilization ability. Similarly, In the current study, *P. libanensis* BBR-7 dissolved inorganic phosphate. Amri et al. (2023) determined that *Pseudomonas fluorescens* dissolved inorganic phosphate at high density (618.57 µg mL^{-1}). In the current study, *P. fluorescens* BBR-12 dissolved inorganic phosphate. Our findings are consistent with other studies. Table 2 and Figure 2 presents the plant growth-promoting properties of the isolates.

3.3. Antifungal activity

In the current study, the Antifungal activity of isolates obtained from *R. officinalis* rhizosphere was tested against *Fusarium oxysporum* and the inhibition percentages varied between 19.40% and 61.54%. Among the isolates, isolate BBR-6 demonstrated the strongest antagonism against *F. oxysporum* with a high percentage inhibition value (61.54%), followed by isolate BBR-9 (55.35%). Isolate BBR-10 (19.40%) showed the weakest effect against the pathogen (Table 3). *F. oxysporum* is a major soil-borne plant pathogen that causes economically significant diseases in agricultural production worldwide. *R. officinalis* wilt caused by *F. oxysporum* causes yield losses. *Bacillus* spp. is regarded as a successful bacteria capable of synthesizing a diverse range of useful compounds. The production of antifungal metabolites by PGPRs such as *Bacillus* is a well-documented biocontrol agent against phytopathogens (Chowhan, et al., 2023). In the current study, *B. mojavensis* BBR-6 showed a maximum inhibition rate of 61.54%. Similar results were obtained by Diabankana et al. (2021) and Abdelkefi et al. (2024). It has been shown in many studies that *B. mojavensis* and *B. cereus* produce fungal wall-degrading enzymes (Ramírez et al., 2022; Chowhan, et al., 2023). In this study, we can suggest that the high antifungal activity of *B. mojavensis* BBR-6 and *B. cereus* BBR-9 against *F. oxysporum* can be attributed to these enzymes. Bautista et al. (2010) reported that *Bacillus megaterium* B14 inhibited the mycelium development of *F. oxysporum* by 40%. In the current study, *B. megaterium* BBR-3 showed a high inhibition rate of 30.35% against *F. oxysporum*. Numerous studies have shown that *Pseudomonas* sp., which is commonly found in soil and rhizosphere, prevents the growth of plant diseases by secreting various compounds (Wang et al., 2020). Rathore et al. (2020) determined that *P. fluorescens* Pf-5 showed 82.51% growth inhibition against *Fusarium* sp. In the current study, *P. fluorescens* BBR-12 showed a high inhibition rate of 54.04% against *F. oxysporum*. In a recent study, Yang et al. (2024) determined that *P. libanensis* P3P4 showed 78.17 % growth inhibition against *F. oxysporum*. In the current study, *P. libanensis* BBR-7 showed a high inhibition rate of 39.52% against *F. oxysporum* (Table 3).

Table 3. Antifungal activity test results of rhizobacterial isolates against *F. oxysporum*
***Çizelge 3.** Rizobakteriyel izolatların F. oxysporum'a karşı antifungal aktivite test sonuçları*

Isolates	Colony diameter of *F. oxysporum* (mm) Mean ± SD	Inhibition percentage (%) of *F. oxysporum*
Bacillus thuringiensis BBR-1	51.7±0.80^{g}	45.59
Bacillus pumilus BBR-2	62.1±1.05^{cd}	33.21
Bacillus megaterium BBR-3	64.5±0.75^{bc}	30.35
Pantoea agglomerans BBR-4	47.2±1.41^{h}	50.95
Lactobacillus paracasei BBR-5	67.6±3.22^{b}	26.66
Bacillus mojavensis BBR-6	38.3±1.05^{ı}	61.54
Pseudomonas libanensis BBR-7	56.8±1.70^{ef}	39.52
Pseudomonas chlororaphis BBR-8	53.9±0.52^{fg}	42.97
Bacillus cereus BBR-9	43.5±0.60^{h}	55.35
Pseudarthrobacter scleromae BBR-10	73.7±0.87^{a}	19.40
Lactobacillus oligofermentans BBR-11	66.4±0.91^{b}	28.09
Pseudomonas fluorescens BBR-12	44.6±0.26^{h}	54.04
Bacillus simplex BBR-13	60.2±1.60^{de}	35.47

*For antifungal activity: p<0,001; statistically significant level. a-ı: The difference between the means shown by different letters in the same column is statistically significant. (Mean ± SD: Mean±Standard Deviation)

4. Conclusion

To our knowledge, the current study is the first in Turkey to isolate PGPR from *R. officinalis* rhizosphere. Over the last two decades, multiple studies have indicated that PGPR strains in many plant rhizospheres aid plant growth and development. PGPR plays roles in producing phytohormones, increasing nutrient availability, and protecting the plant against many pathogens. Research is scarce on determining the ecology of PGPR. There is a need to screen strategies for selecting the best local rhizobacterial strains for use as environmentally friendly biofertilizers to prevent the long-term use of fungicides that cause environmental and ecological problems. Rhizobacteria isolated from *R. officinalis* exhibit significant plant growth-promoting properties and antifungal activities. These isolates can serve as effective microbial fertilizers, offering an environmentally friendly alternative to chemical fertilizers and contributing to sustainable agriculture. Therefore, further research on PGPR is necessary to help create more effective local rhizobacterial strains that can function in several agroecological environments.

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Fertilization and Compost Effects on Nutrient Content and Growth in Cut Tulip Cultivation

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Abstract: This study has two different objectives. The first one is to compost the grape marc obtained from a food processing factory and different materials together to obtain a new organic material. The second was to use this organic material as an alternative growing medium to cultivate cut tulips and determine the effects of different fertilization rates on plant growth, development, and leaf nutrient uptake. Compost was added to the growing medium at 20% and 40%, and chemical fertilization was applied at different electrical conductivity (EC) levels (0.75, 1.10, and 1.45 dS m^{-1}). According to the results, the highest flower stem length (33.94 cm) and perianth length (42.71 mm) were obtained in the medium with 20% compost, while the longest vase life (8.94 days) was obtained with 40% compost and 1.45 dS m^{-1} EC level. The highest branch weight (28.29 g) was measured with 20% compost and 1.45 dS m^{-1} EC. In the cultivar 'Jan van Nes', 20% compost gave more favorable results than the other treatments. Increasing the EC level limited plant growth but increased the macro- and micronutrient content in the leaves. These results suggest that 20% compost can be used as an effective organic material in cut tulip production.

Keywords: Cut flower, Soilless agriculture, waste management, *Tulipa gesneriana*.

Gübreleme ve Kompostun Kesme Lale Yetiştiriciliğinde Besin İçeriği ve Büyüme Üzerine Etkileri

Öz: Bu çalışmanın iki farklı amacı vardır. Birincisi, bir gıda işleme fabrikasından elde edilen üzüm posası ve farklı materyallerin bir araya getirilerek kompostlaştırılması ve yeni bir organik materyal elde edilmesidir. İkincisi ise bu organik materyalin kesme lale yetiştiriciliğinde alternatif bir yetiştirme ortamı olarak kullanımını ve farklı gübreleme oranları ile bitkilerde büyüme, gelişim ve yapraktaki besin alımı üzerine etkilerin belirlenmesidir. Kompost yetiştirme ortamına %20 ve %40 oranlarında eklenmiş ve farklı elektriksel iletkenlik (EC) seviyelerinde (0.75, 1.10 ve 1.45 dS m^{-1}) kimyasal gübreleme uygulanmıştır. Sonuçlara göre, en yüksek çiçek sapı uzunluğu (33.94 cm) ve periant uzunluğu (42.71 mm) %20 kompost içeren ortamda elde edilirken, en uzun vazo ömrü (8.94 gün) %40 kompost ve 1.45 dS m^{-1} EC seviyesinde elde edilmiştir. En yüksek dal ağırlığı (28.29 g) %20 kompost ve 1.45 dS m^{-1} EC seviyesinde ölçülmüştür. 'Jan van Nes' çeşidinde, %20 kompost diğer uygulamalara göre daha olumlu sonuçlar vermiştir. Artan EC seviyeleri bitki büyümesini sınırlamış ancak yapraklardaki makro ve mikro besin içeriklerini artırmıştır. Bu bulgular, %20 kompostun kesme lale yetiştiriciliğinde etkili bir organik materyal olarak kullanılabileceğini göstermektedir.

Anahtar Kelimeler: Kesme çiçek, topraksız tarım, atık yönetimi, *Tulipa gesneriana*.

1. Introduction

Tulipa gesneriana, commonly known as the tulip, is a bulbous plant across the Mediterranean, Central and East Asia, Europe, and North Africa and is extensively cultivated globally (Hu et al., 2023). Tulipa is divided into two subgenera, Eriostemones and Leiostemones, and these subgenera are further subdivided into eight sections and contain approximately 55 species (Lim and Van, 2006). *Tulipa gesneriana* is the most widely cultivated species (Benschop et al., 2010). Today, tulips are commonly used in landscaping and as cut flowers (Su et al., 2022).

Tulips have recently been intensively produced in a soilless culture. In soil cultivation, soil-borne disease factors cause yield and quality loss. For this reason, substrate culture is preferred as the most usable method and has the highest applicability. In substrate culture, organic media such as peat, coconut peat, sawdust, tree bark, paddy husk, peanut shell, inorganic media such as sand, gravel, volcanic tuff, perlite, vermiculite, expanded clay, rock wool or synthetic media such as polyurethane foam are used (Eken & Şirin, 2018). The sustainability of organic and inorganic media has been decreasing, and new growing media are being sought to replace them. While determining these media, sustainability is at the forefront, and it is also aimed to have effects that can increase the quality and yield of the plant.

With rapid urbanization and global population growth, significant amounts of organic matter are wasted (Bong et al., 2017; Thomson et al., 2022). The primary organic wastes are food waste from residential, commercial, and manufacturing sectors and food processing factories (Melikoglu et al., 2013). These wastes are disposed of in ways incompatible with traditional organic waste management, such as landfilling, burial, or incineration. However, these organic wastes can be transformed by the biological decomposition of organic matter under aerobic conditions, and this process is known as 'composting' (Thomson et al., 2022). The compost obtained at the end of the composting process is used for soil improvement and as a growing medium. It also has commercial and agricultural value as an approach to dealing with organic waste (Hubbe et al., 2010; Meyer-Kohlstock et al., 2013; Walker et al., 2006). Compost is an increasingly popular product used as a substrate in soilless agriculture, offering numerous advantages through environmental and plant health approaches (Avilés et al., 2011).

The uncontrolled use of chemical fertilizers to stimulate plant growth, which started with the "Green Revolution," has led to severe environmental problems (Ning et al., 2017; Nikolaou et al., 2023; Tennakoon et al., 2019). These problems may lead to irreparable consequences if serious measures are not taken. In this context, the most critical solution to the problem is to use natural resources more rationally, reuse some waste, and incorporate it into plant production environments, thus ensuring sustainability. This study was planned, conducted, and finalized for this purpose. In this study, compost obtained from grape pomace as a substrate was tested at different ratios in the growing medium of ‘Jan van Nes’ cut tulip variety to determine its potential to be used as a new growing medium in addition to commercial growing media and its effectiveness in reducing the use of chemical fertilizers.

2. Materials and Methods

2.1. Plant and compost materials

The study used the 'Jan van Nes' variety of tulips (*Tulipa gesneriana*) as plant material. Tulip bulbs of the variety were provided from Asya Lale (Konya, Turkey). The species and variety, widespread in bulb and cut flower production abroad and becoming widespread in our country, were selected. The bulbs of the 'Jan van Nes' variety have a circumference of 11 cm, a flowering time of 35-40 days, a number of flowers per branch, a plant height of 50-55 cm, a flower color of yellow, and vase life of 6-8 days. The Triumph tulip group includes the 'Jan van Nes' tulip variety. The characteristics of Triumph tulip varieties are as follows: no catkins, medium-length stems, mid-season flowering, and Single Early and Single Late hybrids.

In the research, 8 tonnes of grape pomace, 1 tonne of barnyard manure, 8 straw bales of 12 kg, 40 kg of lime and 4 kg of urea obtained from Tokat Dimes Food Processing Factory were used for composting. For nutrient solution, 2 nutrient tanks with a capacity of 1000 L were preferred. The study was carried out in a 450 m² glass greenhouse with roof ventilation, without heating and cooling.

2.2. Methods

2.2.1. Preparation of the compost used in the study

The materials (barnyard manure, straw, lime, and urea) for compost to be used as a growth medium for tulip cultivation were mixed and produced under aerobic conditions according to the sequential heap composting method (Windrow Method) (Durmuş & Kızılkaya, 2018). The compost matured after 135 days, and 1 kg samples were taken after Ankara Soil, Fertilizer, and Water Resources Central Research Institute analyzed maturation. The physical, chemical and biological properties of the compost content were reported in the study conducted by Alkaç and Güneş (2022). At the end of the study, pH Richards (1954) and EC Maas (1986) analyses of tulip growing media were performed on samples taken from the crates after the bulbs were removed.

2.2.2. Pre-treatment of tulip bulbs and planting

The tulips were harvested from the soil in mid-June 2021 (when the leaves were dry). The cleaned bulbs were classified depending on the size of the calibration machine. The bulbs with completed female flower formation (G Stage) were taken to cold storage. The bulbs were stored at 20, 15, and 12 °C for 14 days at intervals of 20, 15, and 12 °C, respectively. The relevant company held the bulbs at 10 °C until November 18, when the bulbs were supplied. Bulbs delivered to us on November 18 were kept at seven °C for 14 days between November 18 and December 1 and at 5 °C for 70 days between December 1 and February 8. At the end of storage, the bulbs were planted in crates with a depth of 20 cm, a length of 60 cm and a width of 40 cm. 10x9=90 tulip bulbs were planted in each crate with 10 bulbs in size and 9 bulbs in the width of the crate, covered with fine sand with a thickness of 1.5-2 cm, and the crates were kept in cold storage at 10 °C and 75% moisture content. When the bulbs reached about 10 cm shoot length in the cold storage, they were moved to the greenhouse.

Fertilization: Fertilization was adapted according to solution formulations prepared according to Hoagland and Arnon (1950) and Alkaç and Güneş (2022). Tulips received only 1/3 of the specified amount of nutrient solution. The EC level of the first nutrient tank was adjusted to EC 1.1 ds m^{-1}, and the EC level of the second nutrient tank was adjusted to 1.45 ds m^{-1}. As a control, only water was given to the plants. Irrigation and fertilization were done with a drip irrigation system. The EC value of tap water applied as control was 0.75 ds m^{-1}. The tanks were checked every seven days, and the nutrient solutions' EC levels and pH values (5.6) were kept at the desired levels (Dole, 2005; Özzambak & Zeybekoğlu, 2004).

Greenhouse climatic conditions: The prepared crates were placed on the floor in the greenhouse. The climatic conditions inside the greenhouse were recorded with a data logger (Hobo, Datalogger). During the experiment, the average temperature in the greenhouse was 17 °C± 2 °C, the highest temperature was 33±2 °C and the lowest temperature was 5±1 °C, while the relative humidity in the greenhouse was 52±2%. In the greenhouse, 55% of the shading was made with a green net in the study area. Plants were irrigated with two rows of drip irrigation pipes (2 L h^{-1} flow rate) placed in the crates. The amount of irrigation was determined as 1 L of water per case, and the duration was calculated and given at regular intervals every two days. In the study, fertilization was started when tulip shoots reached 10 cm in length. Harvesting of tulips was carried out at the nodes where leaf formation started above the soil level before the petals turned color and the tips opened (Van Doorn, 1998).

Observations and measurements on tulips: In tulips, bud emergence time (days) and rate (%), full flowering time (days), flower stalk length (cm), flower stalk thickness (mm), branch weight (g), flower bud diameter (mm), flower bud length (mm) and vase life (days) characteristics were examined. Vase life was determined by cutting the stems of the flowers brought to the laboratory at a length of 30 cm and placing them in glass bottles containing 500 mL pure water in 3 replicates with three plants in each replicate. Vase life was determined when the flowers were placed in the vase until the leaves started turning yellow, the petals wilt and fall, and the flower stem bent (Karunaratne et al., 1997). The conditions of the vase life chamber were temperature 21±2 °C, humidity 52±5% (Hobo Data Logger U12-012, Onset, United States of America), light 482 lux, and day length 11 hours light and 13 hours dark.

2.2.3. Leaf analysis

Macro and microelement analysis: Leaf samples taken during the bud stage of tulip plants were dried in an oven at 80 °C and prepared for analysis. According to the method reported by Miller (1998), the readings of macro and micronutrients such as phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), iron (Fe), manganese (Mn), zinc (Zn) and copper (Cu) dissolved in hydrochloric acid were determined by ICP-AES device. Nitrogen (N) was determined by the standard Kjeldahl fresh digestion method. According to the methods reported by Bowman et al. (1988) and Jones (2001), the ammonium (NH_4) formed from the breakdown of nitrogen in the plant sample by fresh combustion using Kjeldal flasks with sulfuric acid in the presence of catalysts was measured by distillation method.

2.2.4. Experimental design and statistical analysis

The study was established in a factorial arrangement in the randomized complete block design (RCBD) and each treatment in the study had 3 replicates and 90 bulbs per replicate were used in the study. The results obtained were evaluated by analysis of variance (ANOVA) in SPSS (IBM SPSS Statistics for Windows, Version 26.0. Armonk, NY: IBM Corp.) statistical programme. Duncan multiple comparison test was applied to determine the significance of the differences between treatments

3. Results

3.1. Effect of compost rates and fertilization with different EC levels on some quality characteristics of 'Jan van Nes' tulip cultivar

When we evaluated the effects of compost ratios (0%, 20%, 40%) and EC levels (0.75, 1.10, 1.45 dS m^1) on the quality traits of the tulip variety 'Jan van Nes', it was found that the differences in flower stem length, petal width and vase life were very significant, while flower stem thickness and branch weight were significant. No significant difference was observed for petal length. However, when the interaction of factors was considered, in some cases not only the effect of compost ratio or EC level was significant, but also the effect of these two factors together. For example, 20% compost and 1.10 EC resulted in the lowest stem length (27.84 cm), while 0% compost and the same EC resulted in a significant increase in stem length (33.81 cm). This shows that not only the compost ratio or the EC level, but also the interaction of these two factors played an important role. Similarly, the differences between 40% compost and 0.75 EC and 40% compost and 1.45 EC were too large to be explained by the independent effects of each factor alone. For example, 40% compost and 0.75 EC resulted in the widest petal width (17.73 mm), while 40% compost and 1.45 EC resulted in the narrowest petal width (14.91 mm). Similar interactions between branch weight and vase life were also observed. 20% compost and 1.45 EC produced the highest branch weight (28.29 g), while 0.75 EC produced a lower branch weight (23.69 g) at the same compost rate. In terms of vase life, 40% compost and 1.45 EC produced the longest vase life (8.94 days), while 20% compost and 1.10 EC produced the shortest vase life (7.36 days) (Table 1).

Table 1. Effect of fertilization treatments with different compost rates and EC levels on some quality characteristics of 'The Jan van Nes tulip cultivar.

Çizelge 1. *Farklı kompost oranları ve EC seviyeleri ile gübreleme uygulamalarının 'The Jan van Nes lale çeşidinin bazı kalite özellikleri üzerine etkisi.*

Compost (%) x Fertilization (EC)	FSL (cm)	FST (mm)	PW (mm)	PL (mm)	BW (g)	VL (day)
0 x 0.75	31.62±0.72 ab	8.70±0.10 a	17.19±0.28 a	41.64±0.51	28.03±0.91 a	7.74±0.14 b
0 x 1.10	27.84±0.70 c	8.31±0.11 ab	15.70±0.27 cd	39.13±0.57	25.28±0.54 ab	7.42±0.18 b
0 x 1.45	30.74±1.35 abc	8.70±0.11 a	17.03±0.35 ab	41.23±0.83	27.24±0.89 ab	7.08±0.14 b
20 x 0.75	33.47±0.47 a	8.11±0.11 b	15.66±0.32 cd	42.32±0.49	25.76±0.82 ab	8.71±0.20 a
20 x 1.10	33.81±1.36 a	8.75±0.27 a	15.78±0.38 cd	41.17±1.16	26.22±0.96 ab	7.36±0.25 b
20 x 1.45	31.18±1.00 ab	8.80±0.08 a	16.84±0.30 abc	42.25±0.72	28.29±0.94 a	7.46±0.22 b
40 x 0.75	29.00±0.00 bc	8.89±0.10 a	17.73±0.86 a	42.02±0.34	23.69±0.58 b	7.75±0.11 b
40 x 1.10	31.67±1.26 ab	8.37±0.35 ab	15.96±0.51 bcd	40.82±1.03	23.95±1.24 b	6.38±0.37 c
40 x 1.45	32.01±0.64 ab	8.34±0.10 ab	14.91±0.37 d	41.32±0.46	27.11±1.81 ab	8.94±0.26 a
Significance	0.001	0.038	0.001	0.122	0.040	0.001

FSL: Flower Stem Length, **FST:** Flower Stem Thickness, **PW:** Petal Width, **PL:** Petal Length, **BW:** Branch Weight, **VL**: Vase Life, **±:** Standart Error.

3.2. The effect of different compost rates and fertilization treatments with different EC levels on some phenological observations of 'Jan van Nes' tulip cultivar

It was observed that compost levels (0%, 20%, 40%) and EC levels (0.75, 1.10, 1.45 dS m^{-1}) had significant effects on the phenological characteristics of the tulip variety 'Jan van Nes'. Significant differences were found between bud emergence time and rate and full flowering time. However, it was observed that interactions also played an important role in these traits. For example, 0% compost and 0.75 EC resulted in the earliest bud emergence (47.17 days) and the highest emergence rate (63.70%), while 40% compost and 1.10 EC resulted in the latest bud emergence (57.83 days). This difference can be explained not only by the effect of EC level or compost ratio, but also by the effect of the combination of these factors. In addition, interactions are also important for flowering time. For example, 20% compost and 0.75 EC resulted in the longest flowering time (59.79 days), while 20% compost and 1.10 EC resulted in the shortest flowering time (57.04 days), indicating that not only the effects of both factors but also their interactions play a significant role (Table 2).

Table 2. Effect of fertilization treatments with different compost rates and EC levels on phenological observations in the 'Jan van Nes' tulip cultivar.

Çizelge 2. *Farklı kompost oranları ve EC seviyeleri ile gübreleme uygulamalarının 'Jan van Nes' lale çeşidinde fenolojik gözlemler üzerine etkisi.*

Compost (%) x Fertilization (EC)	BET (day)	BER (%)	FFT (day)
0 x 0.75	47.17±2.48 d	63.70±2.84 a	59.58±0.29 a
0 x 1.10	52.17±2.39 bcd	53.06±1.42 bc	59.33±0.38 a
0 x 1.45	51.29±1.79 cd	51.44±1.00 bcd	57.58±0.34 bc
20 x 0.75	57.83±0.39 a	50.33±1.64 bcd	59.79±0.46 a
20 x 1.10	56.04±0.67 abc	49.08±1.30 cd	57.04±0.41 c
20 x 1.45	51.17±1.68 cd	54.54±1.70 b	58.58±0.36 ab
40 x 0.75	56.00±0.00 bc	47.33±0.21 d	59.00±0.00 a
40 x 1.10	57.13±0.33 ab	46.53±0.34 d	57.50±0.33 bc
40 x 1.45	56.83±0.17 bcd	49.91±0.99 bcd	58.58±0.51 ab
Significance	0.001	0.001	0.001

BET: Bud Emergence Time, **BER:** Bud Emergence Rate, **FFT:** Full Flowering Time, **±:** Standart Error.

3.3. EC values of tulip growing media

At the end of the vegetation period of 'Jan van Nes' tulip cultivar, the EC levels of the samples taken from growing media with different compost rates (0%, 20%, and 40%), different EC levels (0.75, 1.10 and 1.45 dS m^{-1}) treatments were examined and the EC level was determined as 'No Salt' (0-4000=No Salt) in all treatments. The highest measured EC level (400 µS cm^{-1}) was obtained from the growing medium at a 20% compost rate, and the EC level was 1.10. The lowest EC level (160 µS cm^{-1}) was measured in the growing medium with no compost (0%) and an EC level of 0.75 dS m^{-1}.

3.4. pH values of tulip growing media

At the end of the vegetation period of 'Jan van Nes' tulip variety, when the pH levels of the samples taken from growing media with different compost ratios and different EC levels treatments were examined, pH levels were determined as 'neutral' (6.5<neutral 7.5) in all treatments in growing media without compost, with 20% and 40% compost. The highest pH level (7.60) was measured in the medium with 40% compost treatment and an EC level of 1.10. The lowest pH level (6.23) was measured in the growing medium without compost (0%) and with an EC level of 0.75.

3.5. Macro and micronutrient contents of tulip leaf samples

It was observed that the combination of different compost applications and EC levels significantly affected the macro and micro nutrient content in the leaves of 'Jan van Nes' tulip. It was found that the nutrient content was influenced not only by the independent effects of compost ratio and EC level but also by the interactions between these factors. The media provided the highest value in terms of nitrogen content (2.22%) with 0% compost and an EC value of 1.45 dS m^{-1}. On the other hand, the media with 40% compost and 0.75 dS m^{-1} EC had the lowest nitrogen content (1.52%). This difference can be explained by the effect of compost ratio or EC level and the interaction of these two factors. In particular, nitrogen content decreased as the compost ratio increased and EC level decreased, while media with higher EC levels provided higher nitrogen content. Phosphorus content was also affected by the interactions in a similar way. While the highest phosphorus content (0.52%) was obtained with 20% compost and 1.45 dS m^{-1} EC, 40% compost and 0.75 dS m^{-1} EC decreased the phosphorus content to the lowest level (0.32%). This result shows that phosphorus content remains low, especially for environments with high compost rates and low EC levels. In terms of potassium content, the highest value (1.78%) was obtained with 40% compost and 1.45 dS m^{-1} EC. On the other hand, the lowest potassium content (1.16%) was obtained with 0% compost and 0.75 dS m^{-1} EC. Here, it is clearly seen that interactions play an important role in potassium content. Iron content reached the highest value (148.17 ppm) in 20% compost and 1.45 dS m^{-1} EC level, while 40% compost and 0.75 dS m^{-1} EC level decreased the iron content to the lowest level (72.92 ppm). This interaction provides important information on how compost rate and EC level can affect iron uptake. In terms of copper content, 20% compost and 1.45 dS m^{-1} EC yielded the highest copper content (7.45 ppm), while 40% compost and 0.75 dS m^{-1} EC yielded the lowest copper content (3.53 ppm). This result shows that copper can change with the effect of compost rate and EC level. Zinc content reached the highest value (48.70 ppm) with 40% compost and 1.45 dS m^{-1} EC level, while 20% compost and 0.75 dS m^{-1} EC level decreased the zinc content to the lowest level (14.76 ppm). This difference also reflects the interactive effect of compost ratio and EC level. Manganese content reached the highest level (55.24 ppm) with 40% compost and 1.45 dS m^{-1} EC, while 0% compost and 0.75 dS m^{-1} EC showed the lowest manganese content (19.16 ppm). This shows that manganese uptake is particularly associated with high compost and high EC environments. Calcium content reached the highest value (1.61%) with 40% compost and 1.45 dS m-1 EC, while 0% compost and 0.75 dS m-1 EC provided the lowest calcium content (0.94%). This interaction shows that calcium uptake also changes with the effect of compost and EC level. Magnesium content reached the highest level with 20% compost and 1.45 dS m^{-1} EC, while 0% compost and 0.75 dS m^{-1} EC had the lowest magnesium content. This interaction also shows that magnesium uptake can be associated with high compost and EC levels (Table 3).

Table 4 presents the correlation matrix for the effects of different compost ratios and various EC levels on the quality parameters of tulips. The highest correlation was found between flower stem length and perianth length (r=0.698), while the lowest correlation was observed between the full bloom duration and perianth width (r=-0.00).

Table 3. The effect of different compost ratios and EC level fertilization applications on the macro and micro-nutrient contents in leaves of the 'Jan van Nes' tulip variety.
Çizelge 3. *Farklı kompost oranlarının ve EC düzeyinde gübreleme uygulamalarının 'Jan van Nes' lale çeşidinin yapraklarındaki makro ve mikro besin elementi içerikleri üzerine etkisi.*

Compost (%) x Fertilization (EC)	N (%)	P (%)	K (%)	Ca (%)	Mg (%)	Fe (ppm)	Cu (ppm)	Zn (ppm)	Mn (ppm)
0 x 0.75	1.57±0.02c	0.33±0.00d	1.16±0.01e	0.94±0.01e	0.24±0.01b	76.92±2.18d	4.12±0.15c	13.19±0.32f	19.16±0.64f
0 x 1.10	1.89±0.02b	0.43±0.00c	1.43±0.01c	1.43±0.01c	0.33±0.00b	125.92±2.31c	5.99±0.15b	28.48±0.69d	34.98±0.94d
0 x 1.45	2.22±0.03a	0.49±0.00b	1.67±0.03b	1.59±0.02a	0.45±0.01b	134.25±2.29ab	7.34±0.05a	40.98±0.47b	45.30±1.10b
20 x 0.75	1.54±0.02c	0.33±0.01d	1.21±0.02de	0.99±0.02e	0.24±0.01b	74.58±1.28d	3.83±0.13cd	14.76±0.27e	22.53±0.40e
20 x 1.10	1.86±0.02b	0.44±0.01c	1.45±0.01c	1.44±0.02bc	0.36±0.00b	126.58±2.27c	6.02±0.14b	32.12±0.50c	42.16±0.65c
20 x 1.45	2.17±0.01a	0.52±0.01a	1.74±0.02a	1.59±0.02a	0.83±0.36a	148.17±5.01a	7.45±0.05a	47.50±0.92a	54.91±0.52a
40 x 0.75	1.52±0.02c	0.32±0.00d	1.23±0.02d	1.04±0.01d	0.25±0.01b	72.92±1.17d	3.53±0.05d	15.59±0.21e	22.80±0.32e
40 x 1.10	1.87±0.02b	0.44±0.00c	1.40±0.02c	1.49±0.02b	0.36±0.01b	134.42±1.42ab	5.76±0.34b	32.68±0.23c	44.41±0.39b
40 x 1.45	2.19±0.02a	0.51±0.00ab	1.78±0.01a	1.61±0.02a	0.49±0.01ab	143.17±2.00a	7.42±0.03a	48.70±0.84a	55.24±0.43a
Significance	0.001	0.001	0.001	0.001	0.024	0.001	0.001	0.001	0.001

±: Standart Error.

Table 4. Correlation coefficients between the examined characteristics in the 'Jan van Nes' tulip variety.
Çizelge 4. *'Jan van Nes' lale çeşidinde incelenen özellikler arasındaki korelasyon katsayıları.*

Variables	FSL	FST	PW	PL	BW	VL	BET	BER	FFT
FSL		0.206*	0.095	0.698**	0.497**	0.115	0.113	0.154	-0.081
FST	0.206*		0.590**	0.513**	0.449**	-0.043	-0.124	0.212*	-0.080
PW	0.095	0.590**		0.451**	0.288**	-0.085	-0.202*	0.223*	-0.001
PL	0.698**	0.513**	0.451**		0.546**	0.066	0.015	0.166	-0.088
BW	0.497**	0.449**	0.288**	0.546**		0.148	-0.210*	0.397**	0.075
VL	0.115	-0.043	-0.085	0.066	0.148		0.133	0.150	0.429**
BET	0.113	-0.124	-0.202*	0.015	-0.210*	0.133		-0.490**	-0.108
BER	0.154	0.212*	0.223*	0.166	0.397**	0.150	-0.490**		0.437**
FFT	-0.081	-0.080	-0.001	-0.088	0.075	0.429**	-0.108	0.437**	

FSL: Flower Stem Length, **FST:** Flower Stem Thickness, **PW:** Petal Width, **PL:** Petal Length, **BW:** Branch Weight, **VL**: Vase Life, **BET:** Bud Emergence Time, **BER:** Bud Emergence Rate, **FFT:** Full Flowering Time, ****:** $p < 0.01$. ***:** $p < 0.05$.

4. Discussion

4.1. Effects of compost applications on the growth and flowering quality of tulip plants

It was observed that the addition of 20% compost to the growing medium positively affected tulip flower stem length but had a negative effect on bud emergence time. Statistically significant differences were found in bud emergence time and rates in the non-compost medium (0%). These findings are consistent with the study conducted by Khomami et al. (2019), who applied compost at 0%, 15%, 30%, 45%, 60%, and 100% v/v rates in *Dieffenbachia amoena* and reported that plant growth was increasingly limited as compost application rates increased. Similarly, Cristiano et al. (2018) investigated the effects of peat and sewage sludge-based compost (0%, 30%, and 60%) on *Lantana montevidensis* and found that higher compost concentrations (30 and 60%) negatively affected plant diameter, shoot development, leaf growth, and both fresh and dry mass.

In contrast, Baran et al. (2001) found that the optimum growth of *Hypostases phyllostagya* occurred when grape pomace compost was mixed with peat and perlite at a ratio of 50%. Zulfiqar et al. (2019) reported that 10% compost had a positive effect on the growth of *Dracaena deremensis*, while Papafotiou et al. (2017) showed that 25% compost application had beneficial effects on the growth of *Ficus benjamina*. Despite these positive findings in other plants, our results suggest that increased compost application in tulip cultivation has a negative effect on plant growth. This result may be due to the excessive nutrients provided by the compost, which may be detrimental to tulips, given the relatively low fertilizer requirements of tulips. These findings highlight the importance of optimizing compost application rates for specific plant species, as nutrient requirements and growth responses may vary significantly in different plant species.

4.2. The effects of fertilizer applications on the growth and flowering quality of tulip plants

Farmers commonly use chemical fertilizers to enhance soil fertility, but it is well-documented that excessive use of these fertilizers can degrade soil quality and harm the environment (Ali & Çığ, 2018). In this study, different fertilization applications were tested at various electrical conductivity (EC) levels (EC 0.75, EC 1.10, and EC 1.45) in an effort to reduce fertilizer usage while still maintaining soil fertility. Previous studies have shown that chemical fertilizers can positively affect plant growth. For instance, Boboc et al. (2021) reported that the application of 20-20-20 compound fertilizer enhanced plant growth in *Passiflora caerulea*. Similarly, Vâșcă-Zamfir et al. (2019) found that fertilization increased the number of leaves and branches in *Murraya exotica L.*, and Baltazar-Bernal and Jaen-Contreras (2020) indicated that fertilization promoted vegetative growth in *Heliconia cv. Tropics*.

In this study, the effect of fertilization treatments based on EC levels on tulip plants was examined. The results revealed significant differences between fertilization levels and EC values. Typically, plant growth and flower quality improve with higher fertilization doses. However, in this study, the highest growth was observed at EC 0.75 dS m^{-1}. This finding contrasts with general trends reported in the literature, where higher EC levels are usually associated with better growth. High EC and pH levels, however, were found to have negative effects, which is notable since high EC can lead to salt stress, and low pH levels can cause root rot. The ideal pH for tulip cultivation is between 6 and 7, suggesting that careful pH management is crucial for optimal growth. Additionally, the study found that EC levels increased in direct proportion to the fertilization doses applied, emphasizing the importance of closely monitoring EC levels when applying fertilizers. While artificial fertilizers are sometimes used to increase soil EC, careful balancing is essential to prevent adverse effects. In this study, the highest EC level (1.45 dS m^{-1}) resulted in negative outcomes, highlighting the potential risks of excessive fertilization.

In conclusion, this study provides important insights into the relationship between fertilization and EC levels in tulip cultivation. The findings challenge general assumptions found in the literature, particularly regarding the beneficial effects of higher EC levels, and underscore the need for further research to determine the most effective fertilization strategies for tulips.

5. Conclusion

In the study, grape waste, commonly cultivated in the region and composted over approximately 4.5 months, was utilized as a new organic material in tulip cultivation. Different ratios of this compost (20% and 40%) were applied. In the 'Jan van Nes' tulip variety, particularly favorable results in terms of critical quality criteria such as flower stem length (33.94 cm), perianth length (42.71 mm), and branch weight (28.29 g) were observed in plants with 20% compost application. Another quality criterion, vase life (8.94 days), yielded the best results in plants with 40% compost content and a fertilization level of 1.45 dS m^{-1}. Generally, the application of 20% compost showed better growth and flowering characteristics than the control and 40% compost ratio. Considering the significant input of imported peat in tulip cultivation in soilless agriculture, this study revealed that compost below 20% did not negatively affect plant development, allowing for the utilization of organic waste in the cultivation environment. Another application of the study was different levels of fertilization practices. The application with an EC level of 0.75 dS m^{-1} generally yielded the best results. Adverse effects on plant development were observed when the upper limit of fertilization with an EC level of 1.45 dS m^{-1} was reached. Considering the low fertilizer needs of tulips, it was observed that compost with a high EC, when combined, directly affected plant development.

When all treatments in the study were evaluated together, it was determined that different compost ratios and different EC levels positively affected the macro and micronutrient content in tulip leaves. When tulip cultivation is considered in general, the use of compost below 20% can be recommended. Due to the low fertilizer requirement in tulip cultivation, it is predicted that it will be beneficial to use compost and reduce fertilizer costs with low compost rates.

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Web-Based Environmental Monitoring System Design and Dust Concentration Modeling for Dairy Barns

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Abstract: This study focuses on designing and evaluating a web-based environmental conditions monitoring system for dairy barns, targeting dust concentration and temperature/humidity levels. A prototype device integrating Arduino technology with GSM module was developed to enable real-time data transmission to a web-based platform. The system was tested in a small-scale dairy barn, where it effectively monitored dust, temperature, and humidity while calculating the Temperature Humidity Index (THI). Statistical analysis using multiple regression (MR) and artificial neural networks (ANN) revealed that temperature and humidity significantly influence dust concentrations, with both models achieving strong predictive performance (MR: R^2 = 0.7, ANN: R^2 = 0.71). These results indicate that both methods can accurately predict dust levels based on environmental conditions. This low-cost solution not only improves air quality monitoring but also offers a viable tool for controlling heat stress in dairy barns, enhancing animal welfare and productivity. The system's affordability makes it accessible for small family farms and adaptable for broader agricultural applications.

Keywords: Air quality, Arduino, dairy cattle, dust concentration, temperature humidity index.

Süt Sığırı Ahırları İçin Web Tabanlı Çevre İzleme Sistemi Tasarımı ve Toz Konsantrasyon Modellemesi

Öz: Bu çalışmada, süt ahırları için toz konsantrasyonu ve sıcaklık/nem seviyelerini ölçmeyi hedefleyen web tabanlı bir çevre koşulları izleme sistemi tasarımı ve değerlendirilmesi amaçlanmıştır. GSM modülüyle Arduino teknolojisini entegre eden bir prototip cihaz, web tabanlı bir platforma gerçek zamanlı veri iletimi sağlamak için geliştirilmiştir. Sistem, Sıcaklık Nem İndeksi'ni (SNI) hesaplayarak tozu, sıcaklığı ve nemi etkili bir şekilde izlediği küçük ölçekli bir süt sığırı ahırında test edilmiştir. Çoklu regresyon (ÇR) ve yapay sinir ağları (YSA) kullanılarak yapılan istatistiksel analiz, sıcaklık ve nemin toz konsantrasyonlarını önemli ölçüde etkilediğini ve her iki modelin de güçlü tahmin performansına ulaştığını ortaya koymuştur (YSA: R^2 = 0,7, YSA: R^2 = 0,71). Bu sonuçlar, her iki yöntemin de çevre koşullarına bağlı olarak toz seviyelerini doğru bir şekilde tahmin edebileceğini göstermektedir. Bu düşük maliyetli çözüm yalnızca hava kalitesi izlemeyi iyileştirmekle kalmayıp aynı zamanda süt ahırlarında ısı stresini kontrol etmek, hayvan refahını ve üretkenliğini artırmak için uygulanabilir bir araç sunmaktadır. Sistemin uygun fiyatlı olması, onu küçük aile çiftlikleri için erişilebilir ve daha geniş tarımsal uygulamalar için uyarlanabilir hale getirmektedir.

Anahtar kelimeler: Hava kalitesi, Arduino, süt sığırı, toz konsantrasyonu, sıcaklık nem indeksi.

1. Introduction

Sustainable production in livestock operations can be achieved where human-animal-environmental factors are considered. Environmental conditions affecting animals are of great importance in the quantity and quality of the product. Hence, parameters such as temperature, humidity, lighting and pollutant gas emissions are the most important environmental conditions. In a sustainable livestock enterprise, as well as fulfilling the environmental demands of the animals housed in barns, the negative effects of the animals on the environment during the production process must be taken into account.

Dairy cattle farming is the branch of livestock operations where the highest productivity loss due to inadequate indoor environmental conditions occur. The majority of dairy cattle operations Türkiye are small family enterprises with less than 50 dairy animals (Uzundumlu, 2012). Almost all of these operations have ventilation-related problems. Natural and/or mechanical ventilation systems are either not used or inadequate.

The environmental problems encountered in barns, especially in dairy cattle operations, are not limited to temperature and humidity, but indoor air quality also has significant effects on both the health and welfare of the animals kept and the health of the workers working in the operations. It is known that there are approximately 250,000 livestock operations and approximately 1 million workers employed in these enterprises in the United States alone. Since livestock barns are structures where concentrated pollutants are present and many animals are housed, the health risks they may pose are much higher than other structures (Tan & Zhang, 2004).

One of the most important pollutants in the indoor air of animal barns is particulate matter (PM). PM found in shelter air is almost entirely of biological and organic origin and consists of a mixture of feed, animal dander, feathers, urine, manure and many microorganisms (Harry, 1978; Carpenter, 1986). PM concentrations vary depending on animal species, barn type, feeding methods, animal density, how effectively cleaning is done, seasonal conditions and even time of day (Carpenter, 1986; Takai et al., 1997). It is known that bronchial diseases and inflammatory complaints occur as a result of breathing PM contaminated by endotoxins (Bakutis et al., 2004). Epidemiological studies conducted on farm animals have shown that long-term exposure to endotoxins causes chronic bronchitis and lung failure (Hartung, 1999).

Although the relationship between indoor dust concentration and ammonia/odor concentration has not been mathematically demonstrated (Hartung 1986, Williams 1989, Maghirang et al. 1991, Gay et al. 2003), there are studies showing that when any of these are controlled, the concentrations of the others are significantly reduced (Ullman et al. 2004). For example, it has been observed that odor-related problems are largely eliminated as a result of effective ventilation and removal of dust inside the shelter (Hartung, 1986, Burnet, 1969, Donham et al., 1986, Bottcher et al., 2000).

There are no specific standards regarding the upper permissible limits of dust emissions in shelters. However, OSHA (American Occupational Safety and Health Administration) recommends 15 mg m^{-3} as the total allowable dust concentration (Kirkhorn & Garry, 2000). Takai et al. (1997) stated the upper limit value of respirable dust concentration in barns as 0.38 mg m^{-3}.

There is no economical environmental monitoring and control system that can be used in small family operations Türkiye. A system that can be used especially for monitoring dust concentrations along with temperature and humidity is needed. The aim of this study is to design an on-site dust concentration and temperature/humidity monitoring device with the latest low-cost, easy-to-use and web-based data management system that can be easily accessed over the internet. The developed device can also be used in other agricultural structures.

2. Materials and Methods

2.1. Prototype and software development

Arduino microprocessor card was employed in the prototype. This board has a cost effective and high-capacity microprocessor that can easily integrate different sensors. It was chosen due to its ease of programming, ability to use MS Office applications and other technical features. This card, which can accommodate multiple sensors, be integrated with a touch screen, and provides its own software platform free of charge, provides a dynamic technology development opportunity.

The microprocessor card, the temperature/humidity and dust sensor integrated on it are low-cost technologies, and the data received from the sensors are stored and processed in the same environment and the results are made meaningful with the software developed. The GSM circuit has also been integrated into the system, allowing the data obtained to be transmitted to cloud data storage.

After procuring the necessary electronic material, the prototype system was designed by taking into account the dimensions and technical specifications of the material. The goal here was to bring together the microprocessor, sensors, GSM module and touch control screen on a main body of the smallest possible size. It was thought that if dusty air was pushed into the sensor with a mini air pump, dust would accumulate inside the pump and cause the pump to malfunction in a short time. Therefore, the active surface of the sensor was mounted on the top of the device in a way that it would be in constant contact with the air outside. A rechargeable battery unit was created by connecting two Li-ion 18650 batteries in series. It was used with a TP4056 battery charging module and a 5 V 1 amp DC power supply. The dimensions of the device are given in Figure 1.

The optical sensor integrated into the system is used to measure dust concentration (Sharp Corporation, Japan). There is an infrared transmitter and phototransistor on the sensor. It works by measuring the reflection of the emitted infrared ray from dust particles. The dust sensor, which has a very low current consumption (20mA max.) and can operate with a supply voltage of up to 7 V, has a sensitivity of 0.5 V $1mg^{-1}$ m^{-3} and provides analog voltage output proportional to the dust density. The Sharp GP2Y1010AU0F dust sensor generates an analog

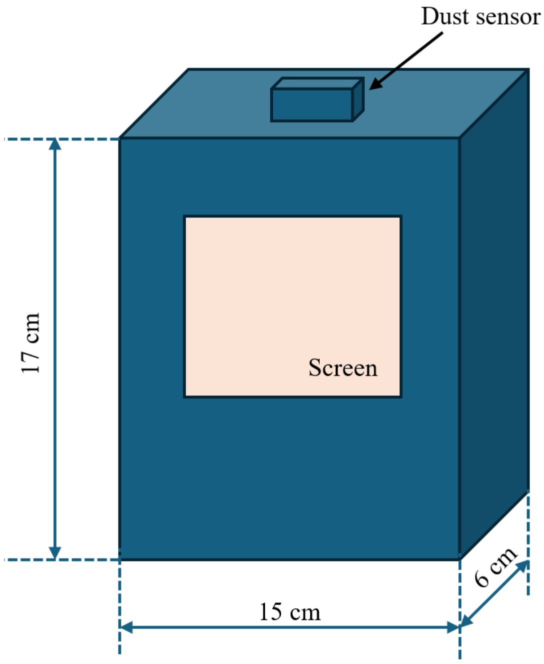

Figure 1. Dimensions of the prototype
Şekil 1. *Prototipin boyutları*

voltage output that corresponds to dust density. It is designed to deliver reliable measurements as shipped therefore calibration is not necessary (Sharp, 2024).

SHT11 temperature and humidity sensor was used to read and record the temperature and relative humidity values inside the barn (Sensirion AG, Staefa ZH, Switzerland). This sensor measures air temperature and relative humidity with high accuracy and sends it to the microprocessor unit. It is capable of preventing interference and data loss problems due to its single path connection and use of standard bandwidth signals. The SHT11 sensor is placed inside the box of the prototype device and its sensing component is suspended out of a small hole opened in the box. To ensure the reliability of the sensor data, it was fixed by soldering directly to the relevant terminals on the Arduino with 10-cm cables, as stated in the technical manual. Dust and temperature/humidity sensors are shown in Figure 3.

The operation of the developed system and the materials used are schematized below (Figure 2).

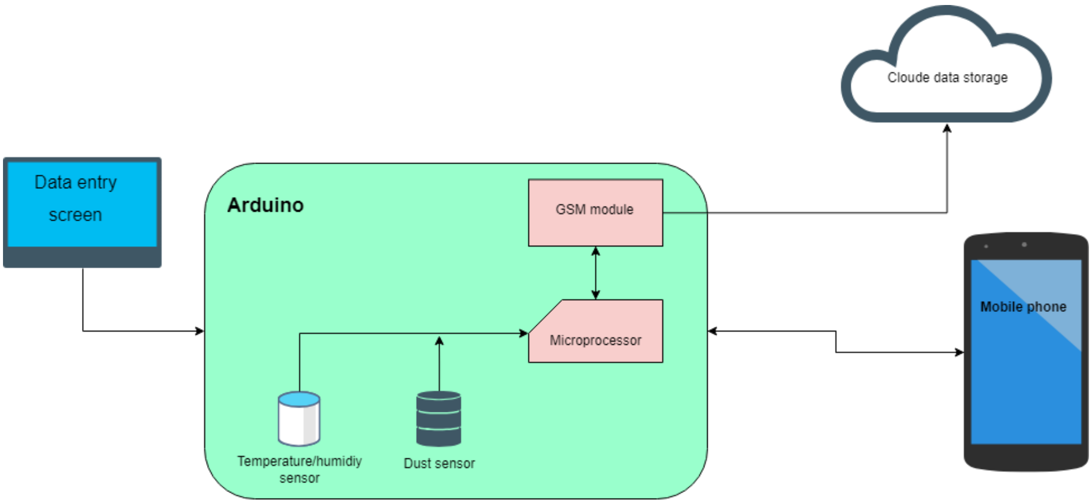

Figure 2. Schematic representation of the prototype
Şekil 2. *Prototipin şematik gösterimi*

The optical sensor integrated into the system is used to measure dust concentration (Sharp Corporation, Japan). There is an infrared transmitter and phototransistor on the sensor. It works by measuring the reflection of the emitted infrared ray from dust particles. The dust sensor, which has a very low current consumption (20mA max.) and can operate with a supply voltage of up to 7 V, has a sensitivity of 0.5 V $1mg^{-1}$ m^{-3} and provides analog voltage output proportional to the dust density. The Sharp GP2Y1010AU0F dust sensor generates an analog voltage output that corresponds to dust density. It is designed to deliver reliable measurements as shipped therefore calibration is not necessary (Sharp, 2024).

SHT11 temperature and humidity sensor was used to read and record the temperature and relative humidity values inside the barn (Sensirion AG, Staefa ZH, Switzerland). This sensor measures air temperature and relative humidity with high accuracy and sends it to the microprocessor unit. It is capable of preventing interference and data loss problems due to its single path connection and use of standard bandwidth signals. The SHT11 sensor is placed inside the box of the prototype device and its sensing component is suspended out of a small hole opened in the box. To ensure the reliability of the sensor data, it was fixed by soldering directly to the relevant terminals on the Arduino with 10-cm

cables, as stated in the technical manual. Dust and temperature/humidity sensors are shown in Figure 3.

a) Dust sensor b) Temperature/humidity sensor

Figure 3. Sensors used in the prototype
Şekil 3. *Prototipte kullanılan sensörler*

Arduino GSM Shield-Simcom/Sim800C unit was used to ensure that the data from the both sensors were sent to the internet server via the mobile network. Because of its ability to operate in four different bands and its internal antenna, data interruption is prevented in areas where the network signal is weak. By adding the DS1307 RTC Module to the entire system, it was possible to send the actual date and time information to the microprocessor and ensure the accuracy of the time stamp of the data received from the sensors.

The platform provided by the microprocessor card was used for the software. The software recorded the dust concentrations of the data received from the dust sensor on the SD card depending on the entered time intervals. The data created in the microprocessor was also transmitted to the Web-based data storage system via the GSM module. The threshold value to be used for dust concentration was taken into account as 0.38 mg m^{-3} (Takai et al., 1998).

The Temperature Humidity Index (THI) equation was used as the temperature-related threshold value. Various indices have been available over the years to measure heat stress, which occurs as a result of the combination of many environmental factors. Since temperature and humidity are both controllable and easily measurable environmental factors, the most common of these indices is THI (Herbut & Angrecka, 2012). The equation for dairy cattle heat stress is given below (NRC, 1971).

$$THI=(1.8\times Ti+32)-(0.55-0.0055\times RHi)\times(1.8\times Ti-26) \quad (1)$$

In the equation, THI is the temperature humidity index; Ti is indoor air temperature (^{o}C) and RHi is indoor relative humidity (%).

In this context, the values obtained from the temperature/humidity sensor were used in the embedded software in the device to calculate THI. When the specified threshold values for dust and THI are exceeded, an SMS was sent to the mobile phone to provide a warning. Considering that the THI value will always remain between 72-90 in shelters that do not have air conditioning systems, especially in hot seasons, 90 will be taken into account as the threshold value within the scope of this project.

The developed prototype can measure temperature/relative humidity, THI and dust concentration values within a one minute time interval. All these values can be constantly checked from the internet database via any computer, phone or smart device with an internet connection.

The platform provided by the microprocessor card (Arduino) was used for the software that collects and processes sensor data. MySQL, a relational database, was used to store data online. The database system, installed on a server computer open to the Internet, is constantly running to respond to incoming requests. A web-based application has been developed to access data in the database. Since its a web-based database, it can be run on any device with internet access without requiring installation. JavaScript and derivative programming environments were used for this application.

2.2. System testing and data collection

Before the developed system was installed in the trial barn, it was tested in the Digital Agriculture Laboratory at Çanakkale Onsekiz Mart University, Faculty of Agriculture. Error handling was done by testing the performance of the sensor data, GSM module and software at the lab.

A stalled-dairy cattle barn with a capacity of 60 cows located in Durali Village, Çan District, Çanakkale Province was chosen as the operation where the system was tested. The barn had a floor area of 25 × 30 = 750 m^2 and a ridge height of 6 m. The average milk yield in the farm was 20 lt day^{-1}. Since the aim of the project was to design a device that can be used by small family businesses, a barn suitable for the capacity and barn type commonly used in the region was selected. An interior picture of the barn showing the prototype device is given in Figure 4.

When choosing where to mount the device, a location was chosen where it would not be affected by air currents and would be as close to the center of the barn as possible. The height above the ground was mounted approximately 2.5 m above the ground so that animals could not reach it.

Figure 4. The prototype device in operation in the barn
Şekil 4. *Prototip cihazın barınakta çalışması*

2.3. Data analysis

The data was primarily used for modeling purposes. It was then checked whether the relationship between recorded temperature, relative humidity and dust sensor data is statistically significant. The dust concentration was estimated using temperature and relative humidity values for this barn. Multiple regression (MR) and artificial neural networks (ANN) algorithms were used for this purpose. Before data analysis, the raw dust sensor data were processed and concentration values were calculated (Equations 2 and 3).

$$D_C = 0.1642V_S - 0.09 \quad (2)$$

$$V_S = \frac{5}{1024} \times R_S \quad (3)$$

In equations; D_C, dust concentration (mg m^{-3}); V_S, sensor voltage; R_S, raw sensor value; 5/1024 coefficient required to calculate sensor data in the 0-5 volt range.

When the database was examined, it was seen that the dust concentration remained within the detection range of the sensor for only 50 minutes between the dates during the study. It was observed that these values were generally exceeded in the 5^{th} and 6^{th} months, when temperature and humidity were relatively higher. This shows that the dust problem is more intense in hot and humid weather. In this context, the relationship between temperature and humidity was intended to be determined by MR and ANN analysis using the available data. Considering technical specifications of the air quality sensor, it can be seen that the dust concentration cannot be calculated when the voltage value is less than 0.6. Therefore, dust concentration was considered zero at values less than 0.6 volts.

The device was designed to record data once per minute. A total of 75,594 minutes of data were collected during the trial period. Using this data, the relationship between dust concentration in response to temperature and relative humidity inputs was examined. However, during the project, unexpected fluctuations in sensor data occurred in cases such as problems arising from the power supply or the intervention of the producer or his children into the device. In this case, the web database, which is constantly under control, enabled these unwanted measurements to be detected in a timely manner. However, there are still values recorded that can be considered as outliers. For this purpose, outlier analysis of the data was performed before multiple regression analysis. The cloud-based Google Colab data analysis platform was used in both outlier and MR/ANN analysis. In the coding Python programming language was employed.

Outliers were determined by the Boxplot method. A boxplot is a statistical plot used to visualize the distribution of a data set. It is frequently used in outlier analysis. The basic components of a boxplot are as follows:

Box: The box represents the quartiles of the data set. The lower boundary of the box is called the first quartile (Q1), and the upper boundary is called the third quartile (Q3). The horizontal line inside the box represents the second quartile or median of the data set (Q2 or median).

Outliers (Whiskers): The lines at the top and bottom of the box show the overall distribution of the data set. These lines usually start at a certain distance from the boundaries of the box and represent minimum and maximum values. However, this distance can be determined by a certain number of standard deviations or a certain percentage.

Outliers: Outliers are values that differ significantly from other data points, usually beyond the whiskers. Boxplot uses a certain threshold value to identify outliers. This threshold is usually determined based on the overall spread of the data set.

After performing outliers analysis the new dataset was used in MR/ANN analysis.

3. Results and Discussion

The prototype device was installed in the barn on 10.01.2023, and 75,594-minute readings were made in a period of approximately 6 months until 09.06.2023. The data downloaded from the web-based database was converted into an Excel file and the necessary statistical analyzes were performed. The database included dust concentration (mg m^{-3}), temperature (C), relative humidity (%) and temperature humidity index readings as explained above.

In order to evaluate the effects of temperature and humidity on dust concentration a multiple regression analysis was conducted. A screenshot of the multiple regression analysis results performed in Google Colab is given in Figure 5.

```
                            OLS Regression Results
==============================================================================
Dep. Variable:                   Dust   R-squared:                       0.699
Model:                            OLS   Adj. R-squared:                  0.699
Method:                 Least Squares   F-statistic:                 8.233e+04
Date:                Wed, 29 Nov 2023   Prob (F-statistic):               0.00
Time:                        09:40:53   Log-Likelihood:                -73304.
No. Observations:               70793   AIC:                         1.466e+05
Df Residuals:                   70790   BIC:                         1.466e+05
Df Model:                           2
Covariance Type:            nonrobust
==============================================================================
                 coef    std err          t      P>|t|      [0.025      0.975]
------------------------------------------------------------------------------
const         32.8524      0.019   1730.193      0.000      32.815      32.890
Temp           0.2427      0.001    379.877      0.000       0.241       0.244
Hum            0.0087      0.000     42.573      0.000       0.008       0.009
==============================================================================
Omnibus:                    19604.374   Durbin-Watson:                   0.664
Prob(Omnibus):                  0.000   Jarque-Bera (JB):           112039.988
Skew:                           1.213   Prob(JB):                         0.00
Kurtosis:                       8.665   Cond. No.                         408.
==============================================================================

Notes:
[1] Standard Errors assume that the covariance matrix of the errors is correctly specified.
```

Figure 5. Multiple regression analysis results
Şekil 5. *Çoklu regresyon analizi sonuçları*

According to the results of multiple regression analysis, the change in dust concentration depending on temperature and relative humidity can be calculated with the following equation (4).

$$D_C = 0.2427T + 0.0087RH + 32.8524 \quad (4)$$

In equation; D_C, dust concentration (mg m^{-3}); T, inside air temperature (C); RH, inside relative humidity (%).

It can be seen that the coefficient of determination (R^2), which explains the effects of the independent variables on the dependent variable, is about 0.7, which shows that temperature and relative humidity have a significant effect on the dust concentration. Again, if we look at the standard errors of the coefficients in Figure 4 (std err), it shows that these values are quite small, meaning that the accuracy of the predictions is high. Summarizing the analysis results, it shows that the multiple regression equation can be used to determine the dust concentration.

An ANN algorithm was also used to evaluate the performance of temperature and relative humidity in determining dust concentration in Google Colab. Numpy, pandas, sklearn, tensorflow and matplotlib libraries were used in modeling. 70% of the data was used in model development and the remainder was used in testing the model. By trial and error, the parameters that yielded the highest R^2 value in the test data were determined. The parameters of the ANN model developed are given in Table 1.

Table 1. ANN model parameters
Çizelge 1. *YSA model parametreleri*

Parameter	Value
Number of neurons in input layer	2
Number of hidden layers	3
Number of neurons in each input layer	8
Number of neurons in output layer	1
Activation functions in hidden layer	Relu
Activation function in output layer	Linear
Number of epochs	100
Model optimizer	Adam
Mean Squared Error on Test Set	0.45
R-squared on Test Set	0.71

When the results obtained from both methods are compared, it is seen that there is no significant difference between them. Therefore, both approaches can be used to determine dust concentration depending on temperature and relative humidity.

Although the primary purpose of this study was to observe and model dust concentration, THI values, which reveal the combined effect of temperature and relative humidity, were also calculated. The results were evaluated using the limits given in Table 2.

Table 2. Stress categories and precautions that can be taken (Xin & Hamson, 1997)
Çizelge 2. *Stres kategorileri ve alınabilecek önlemler (Xin & Hamson, 1997)*

THI value	Meaning	Precautions that can be taken
THI < 72	No stress	-
72< THI< 80	Alarm	Animal behavior is carefully observed. Their breathing is checked. Cooling fans are turned on and animals are given plenty of water
80 < THI < 90	Beginning of danger	If possible, evaporative cooling systems such as sprinklers and fan-pads are activated or the interior is tried to be cooled with water. Meanwhile the animals are carefully observed
THI > 90	Emergency	Animals are moved as little as possible, for example they are not taken to market. In addition to the measures in the danger category, animals are not fed during the hottest hours of the day and the light level inside the shelter is reduced to reduce animal activity and therefore heat production.

Considering Table 2, it may be a correct approach to take the threshold value required for the developed system to send SMS as THI = 72. However, due to the lack of adequate air conditioning systems in barns in Türkiye, especially in small family businesses, this value is easily exceeded for most of the year. Although the study period in which data were collected in the barn (January - June) generally coincided with the winter months, it was observed that the threshold value of 72 was exceeded 64 hours in total (Figure 6). It is obvious that the threshold value will be exceeded almost throughout the day during most of the spring, summer and fall months. Therefore, the threshold value for SMS was taken as 90, which is the beginning of the emergency. Since the developed prototype can be adapted to an air conditioning system, it has a high potential to be used as an automation device.

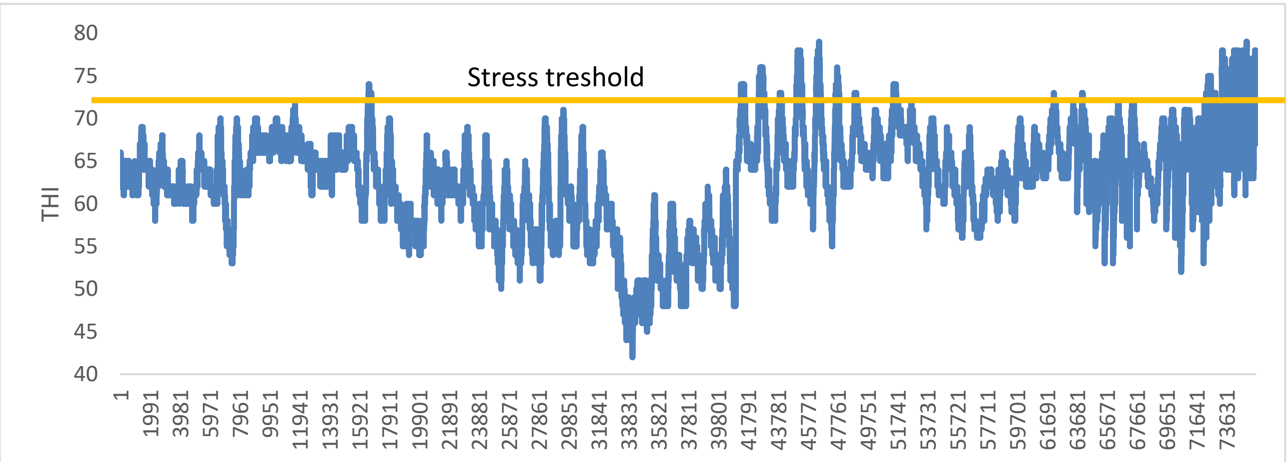

Figure 6. THI values and critical threshold value
Şekil 6. *THI değerleri ve kritik eşik değeri*

4. Conclusion

In this study, a prototype device developed to monitor dust-related air quality in small family operations and the applied methods are reported. In addition, the system has been adapted to control heat stress, which causes low productivity in dairy cattle. Using the collected data, the predictability of dust concentration with temperature and relative humidity values was also tested. For this purpose, performance of MR and ANN algorithms have been evaluated. The analysis of the data showed that both approaches can estimate dust concentration as a parameter of temperature and relative humidity. The cost of the developed prototype device is approximately $250 according to the 2023 exchange rate. This is a very reasonable cost for an automation system that will control dust, temperature and humidity. It offers an economical solution for small family businesses with a small number of animals, especially in Turkey. As a result of this study, it has been seen that being able to access today's technology at very low costs will make significant contributions to finding solutions to agricultural problems.

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Trend Analysis of the Flow and Water Quality Data for the Broad River Basin, South Carolina, USA

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Abstract: Water quality is vital for human health and the protection of natural ecosystems, and demand for quality water is increasing day by day. It is known that changes in precipitation and temperature patterns due to climate change directly or indirectly affect water quantity and quality. In order to understand the potential effects of climate change on water resources, it is very important to know the changes in flow and water quality data over time. Trend analysis methods are the most used methods for this purpose. In this study, monthly, seasonal, and annual changes of dissolved oxygen (DO), water temperature (WT), discharge (Q), and specific conductance (SC) parameters, which are measured and recorded daily between 1987 and 2022 at four monitoring stations in the Broad River Basin (South Carolina, USA), were investigated using the Mann-Kendall test and innovative trend analysis (ITA) methods. Electrical conductivity (EC) values, calculated by considering SC and WT data, were considered. The Mann-Kendall test and ITA identified significant trends in 32.4 and 64.6% of the 272-time series analysed, respectively. It was determined that ITA was more sensitive in identifying decreasing trends. While the spatially and temporally varying trends in the river DO concentration and EC values were associated with human activities, it was concluded that the increasing trends in WT values and decreasing trends in Q values may be due to climate change on precipitation and air temperature parameters. This study, based on long-term data sets, illuminates the global concerns about the impacts of climate change on water quality and provides important findings that will guide sustainable management of water resources and measures to be taken against climate change. It is the first study to examine long-term trends of water quality parameters for the basin.

Keywords: Broad River, innovative trend analysis, Mann-Kendall test, water quality

Broad Nehri Havzası, için Akış ve Su Kalitesi Verilerinin Trend Analizi, Güney Karolina, ABD

Öz: Su kalitesi, insan sağlığı ve doğal ekosistemlerin korunması bakımından hayati öneme sahip olup, kaliteli suya olan ihtiyaç her geçen gün artmaktadır. İklim değişikliği sebebiyle yağış ve sıcaklık rejimlerinde meydana gelen değişimlerin su miktarını ve kalitesini doğrudan veya dolaylı olarak etkilediği bilinmektedir. İklim değişikliğinin su kaynakları üzerindeki potansiyel etkilerinin anlaşılabilmesi için akım ve su kalitesi verilerinin zaman içerisindeki değişimlerinin bilinmesi oldukça önemlidir. Trend analizleri bu amaçla en çok kullanılan yöntemlerdir. Bu çalışmada, Broad Nehri Havzası'nda (Güney Carolina, ABD) seçilen dört gözlem istasyonunda 1987-2022 yılları arasında günlük olarak ölçülmüş ve kaydedilmiş çözünmüş oksijen (DO), su sıcaklığı (WT), debi (Q) ve özgül iletkenlik (SC) parametrelerinin aylık, mevsimlik ve yıllık değişimleri Mann-Kendall testi ve yenilikçi eğilim analizi (ITA) yöntemleri kullanılarak araştırılmıştır. Çalışmada SC ve WT verileri dikkate alınarak hesaplanan elektriksel iletkenlik (EC) değerleri kullanılmıştır. Analiz edilen 272 zaman serisinin %32.4'ünde Mann-Kendall testi, %64.6'sında ise ITA yöntemi anlamlı eğilimler belirlemiştir. ITA yöntemi azalan eğilimleri belirlemede daha hassas olduğu tespit edilmiştir. DO konsantrasyonları ve EC değerlerindeki mekâna ve zamana bağlı olarak değişiklik gösteren eğilimler insani faaliyetler ile ilişkilendirilirken, WT değerlerindeki artma ve Q değerlerindeki azalma eğilimlerinin iklim değişikliğinin yağış ve hava sıcaklığı parametreleri üzerindeki etkilerinden kaynaklanabileceği sonucuna varılmıştır. Uzun süreli veri setlerine dayanan bu çalışma iklim değişikliğinin su kalitesi üzerindeki etkilerine dair küresel endişelere ışık tutmakta ve su kaynaklarının sürdürülebilir yönetimi ve iklim değişikliğine karşı alınacak önlemler konusunda rehberlik edecek önemli bulgular sunmaktadır.

Anahtar Kelimeler: Broad Nehri, yenilikçi eğilim analizi, Mann-Kendall testi, su kalitesi

1. Introduction

Since the Industrial Revolution, rapidly increasing fossil fuel use, deforestation and other human activities have caused greenhouse gases to accumulate in the atmosphere, causing temperatures to rise worldwide. Climate changes are occurring in the world due to global warming. Since climate is one of the main factors controlling the hydrological cycle, climate changes affect the hydrological cycle and therefore water resources (Kundzewicz, 2008). Recent studies have shown that there are significant changes in precipitation and flow patterns worldwide (Da Silva et al., 2015). The effects of these changes on water quality have also become an important research topic, as increasing temperatures and changing precipitation patterns can directly affect water resources. In order to understand the potential effects of climate change and to effectively plan and manage water resources, it is of great importance to know the changes in hydrometeorological and water quality data over time. Trend analysis is used to determine these changes.

Trend analyses are defined as statistical methods used to detect long-term changes in time series data monitored and recorded at specific intervals. These analyses are widely applied in various disciplines, including climate science, economics, and engineering. Trend analyses enable to identify irregularities in hydrological processes and to make estimations for the future (Aytekin, 2012). In recent years, due to missing data, seasonality, and skewed distributions in time series data, non-parametric methods have been increasingly used in trend analyses (Partal & Kahya 2002; Dabanlı, 2017). These methods are typically based on the ranking of data or the differences between ranked data sets (Garcia et al., 2010). In climate science, trend analyses of data such as air temperature, precipitation, discharge, and water quality play a crucial role in understanding the impacts of climate change, and how these effects evolve over time. Various studies have been conducted using different trend analysis methods. Non-parametric methods such as the Mann-Kendall test (Agbo et al., 2023; Gaddikeri et al., 2024; Likinaw et al., 2023), Spearman's Rho test (Swain et al., 2022; Vani et al., 2023), Şen's trend slope (Collaud Coen et al., 2020; Jin et al., 2021), and innovative trend analysis (ITA) (Hirca & Eryilmaz Turkkan, 2022; Tadesse et al., 2024) are commonly employed in literature. Furthermore, various parameters such as air temperature (Acar et al., 2022; Hadi & Tombul, 2018), precipitation (Da Silva et al., 2015; Koruk et al., 2023), evaporation (Javed et al., 2019; Salami et al., 2014), streamflow (Diop et al., 2018; Rogers et al., 2020), humidity (Nourani et al., 2018; Phuong et al., 2020), water temperature (Duy et al., 2022; Ouyang et al., 2021), dissolved oxygen concentration (Hashim et al., 2021; Jamian et al., 2017), hydrogen ion concentrations (Kisi & Ay, 2014), and electrical conductivity (Salvai et al., 2022; Sattari et al., 2020) have been examined.

As in the studies given above, it is important to have long-term data in order to apply trend analysis methods. The Broad River Basin, one of the sub-basins of the Santee River Basin (South Carolina, USA), has long-term monitored flow and water quality data. The basin hosts various industrial facilities, ranging from textile factories to hydropower plants. Despite the implementation of several conservation programs to address environmental threats such as increasing pollution and habitat loss, and the abundance of available monitoring data, studies focusing on water quality in the basin are quite limited. Nacar et al. (2020) modeled surface water quality using the recorded data from two monitoring stations operated in the basin. They concluded that the developed models could estimate the river DO concentrations very close to in situ measurements. Ureta et al. (2020) examined the sediment retention capacity and water yield potential of different land covers. They stated that vegetated areas provide the highest sediment retention capacity and the lowest water yield potential and prevents possible pollution and siltation of streams. Nabi et al. (2021) analysed the effects of rainfall on titanium dioxide (TiO_2) concentrations in the surface waters. They concluded that urban runoff is a major source of TiO_2 engineered particles to urban rivers, and TiO_2 engineered particles, which have high concentrations, may pose environmental risks during and following rainfall events. Maxwell et al. (2022) investigated baseflow levels and found a long-term increase in baseflow over the past millennium. In the literature review conducted by the authors, no trend analysis study on water quality variables was found.

The motivation for this study is the availability of long-term flow and water quality data in the Broad River Basin, coupled with the absence of studies applying trend analysis methods in this basin. Despite the extensive data set, the spatial and temporal variations of dissolved oxygen (DO), water temperature (WT), discharge (Q), and specific conductance (SC) have not been fully explored. This study addresses this gap by applying the Mann-Kendall test and ITA methods to evaluate relevant parameters at four flow and water quality monitoring stations in the basin, operated by the United States Geological Survey (USGS). This study aims to provide new insights into the basin's flow and water quality trends, contributing to a better understanding of the potential impacts of climate change and human activities on the water resources of the region.

There are four main sections in this study. The second section covers the study area, monitoring data, and methods. Results and discussions are presented in the third section, and the final section provides the study's conclusions.

2. Materials and Methods

2.1 Study Area and Monitoring Data

With a surface area of approximately 9.6×10⁶ km², the United States of America (USA) is one of the largest countries in the world. This vast and geographically diverse area encompasses various climate types and water resources. The USA has a complex and extensive network of rivers that are vital to ecosystem health, agriculture, industry, drinking water supply, and recreation. However, changes in precipitation patterns, increasing temperatures, glacial melting, an increase in extreme weather events, and water quality degradation due to climate change pose significant threats to water resources (Schwartz & Randall, 2003). To facilitate the identification, development, and utilization of water resources, the USA is divided into 21 hydrologic regions based on geographic features, climate conditions, and water resource distribution (Santhi et al., 2008). Monitoring studies within these hydrologic regions are conducted by the USGS. These data are critical for water resource management, flood forecasting, water quality monitoring, and environmental research. Thousands of monitoring stations are operated by the USGS across the country, and the collected data are used to ensure the sustainable management of water resources and to study the impacts of climate change.

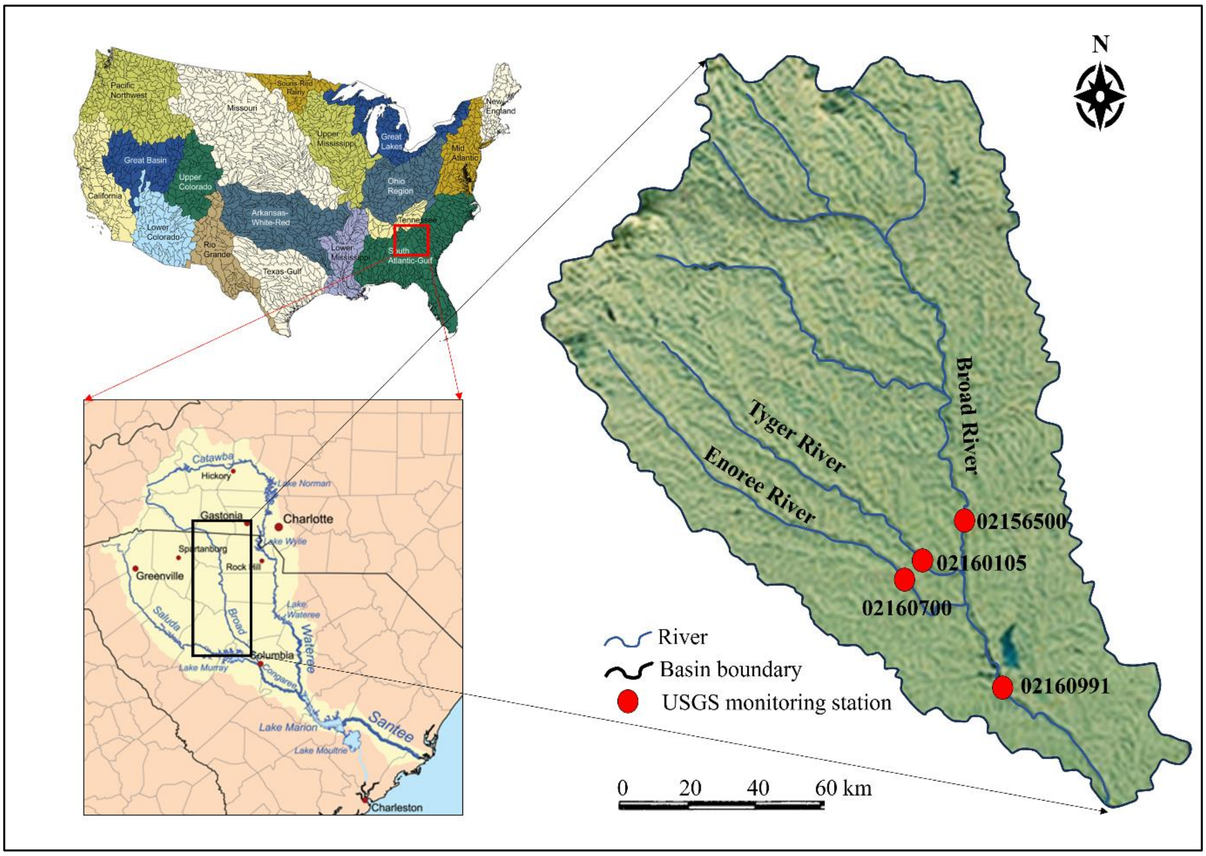

Figure 1. Flow and water quality monitoring stations, Broad River Basin, South Carolina, USA
Şekil 1. *Akım ve su kalitesi gözlem istasyonları, Broad Nehri Havzası, Güney Carolina, ABD*

The Broad River, one of the rivers in the South Atlantic-Gulf Region, originates from the eastern slope of the Blue Ridge Mountains in southwestern North Carolina (approximately 1,219 m) and flows southeastward into South Carolina. The First Broad, Second Broad, Pacolet, Tyger, and Enoree are tributaries of the Broad River (Figure 1). The First and Second Broad rivers are entirely located in North Carolina, and join the Broad River before entering South Carolina. The Broad River Basin covers an area of 9,819 km² within the borders of South Carolina. Approximately 79% of the basin is forest, pastures and row crops are found in approximately 4% and 6% of the total area, respectively. The basin has a temperate climate (Navar et al., 2019; Raschke et al., 1975).

There are 21 monitoring stations in operation, ten of which on the Broad River, five of which on the Pacolet River, two of which on the Tyger River, and five of which on the Enoree River by the South Atlantic Water Science Center in the Broad River Basin. For time series analysis, considering periods of 30 years or more is sufficient for obtaining a valid statistic (Akçay, 2018). Upon review of the stations, therefore, four stations having more than 30 years of simultaneous discharge and various water quality monitoring data, 02156500 (upstream) and 02160991 (downstream) on the Broad River, 02160105 on the Tyger River, and 02160700 on the Enoree River. For each station, daily DO, WT, Q, and SC data for the period between 1987 and 2022 were downloaded from the USGS website. Upon examination of the data, missing values were found to be less than 10%. Missing data were completed using the missForest package in R. Instead of SC data, electrical conductivity (EC) values calculated based on SC and WT data were used.

Trend analyses were conducted based on the monthly, seasonal, and annual mean values of the monitoring data. The basic statistics for the data used from each monitoring station are given in Table 1. S_x, C_s, and C_k are standard deviation, skewness coefficient, and kurtosis coefficient, respectively. C_s and C_k indicate abnormal variation in a statistical series and the values between –1.96 and 1.96 are considered normal by Field (2009). Considering the C_s and C_k values in Table 1, it is seen that the Q data for all stations and the EC data for the USGS 02158500 station show a kurtosis. Explanations of the Mann-Kendall test and ITA methods used in the study are presented under the subsections 2.2. and 2.3., respectively.

Table 1. Basic statistics for monthly mean values of discharge and water quality data in the Broad River Basin, South Carolina, USA

Çizelge 1. *Broad Nehri Havzası (Güney Carolina, ABD) akım ve su kalitesi verilerinin aylık ortalama değerlerine ait temel istatistikler*

USGS station no	Parameter	X_{min}	X_{mean}	X_{max}	S_x	C_s	C_k
02156500	DO	5.22	9.04	13.67	1.81	0.24	–1.12
	WT	4.02	17.86	30.73	7.81	–0.02	–1.41
	Q	10.60	95.23	355.41	66.99	1.45	1.97
	EC	34.48	85.75	263.62	37.06	1.58	3.38
02160105	DO	5.50	8.95	12.70	1.60	0.34	–1.13
	WT	3.70	17.00	28.90	7.20	–0.04	–1.39
	Q	2.10	22.77	100.40	17.64	1.51	2.27
	EC	32.30	84.65	235.80	30.26	1.03	1.60
02160700	DO	6.10	8.92	12.80	1.60	0.34	–1.14
	WT	3.90	16.97	28.60	7.07	–0.04	–1.38
	Q	1.90	13.95	58.80	10.34	1.57	2.50
	EC	35.90	81.76	186.60	28.08	0.85	0.42
02160991	DO	4.80	8.20	12.10	1.84	0.14	–1.22
	WT	5.50	18.96	30.30	7.29	–0.05	–1.41
	Q	16.80	146.52	610.50	111.90	1.53	2.26
	EC	42.30	82.21	147.70	21.85	0.50	–0.23

X_{min}: minimum, X_{mean}: mean, X_{max}: maximum, S_x: standard deviation, C_s: skewness coefficient, and C_k: kurtosis coefficient
DO: mg/L, WT: °C, Q: m^3/s, and EC: µS/cm

2.2. Mann-Kendall Test

The Mann-Kendall test (Mann,1945; Kendall, 1975) is the most applied non-parametric method in trend analysis studies. The test checks for unidirectional (increasing or decreasing) trends in the data set using the two-way null hypothesis H_0 (no trend) and the alternative hypothesis H_1 (Othman et al., 2016). When applying the Mann-Kendall test, the data are ordered chronologically. The S statistic is calculated using Eqs. (1)-(2), in which n is the number of data, x_i and x_j are the data values at time i and j, respectively.

When the number of data is greater than 10, the variance of S is calculated using Eq. (3), in which *p* is the number of linked groups and t_i is the number of times a data value occurs. After the variance of the series is calculated, the standard normal Z value is obtained using Eq. (4).

$$S = \sum_{i=1}^{n-1} \sum_{j=i+1}^{n-1} sgn(x_j - x_i) \qquad (1)$$

$$sgn(x_j - x_i) = \begin{cases} 1 & ; \quad x_j > x_i \\ 0 & ; \quad x_j = x_i \\ -1 & ; \quad x_j < x_i \end{cases} \qquad (2)$$

$$Var(S) = \frac{\left[n(n-1)(2n+5) - \sum_{i=1}^{p} t_i\,(t_i-1)(2t_i+5)\right]}{18} \tag{3}$$

$$Z = \begin{cases} \dfrac{S-1}{\sqrt{Var(S)}} \quad , & S > 0 \\ 0 \quad , & S = 0 \\ \dfrac{S+1}{\sqrt{Var(S)}} \quad , & S < 0 \end{cases} \tag{4}$$

If the absolute value of the calculated Z is greater than the critical z value corresponding to the specified confidence interval, it indicates the presence of a significant trend. Otherwise, it is accepted that there is no significant trend. A positive test statistic indicates an increasing trend, while a negative test statistic indicates a decreasing trend. In this study, a confidence interval of 95% was considered, and therefore the critical z value was determined as 1.96.

2.3. Innovative Trend Analysis

The ITA methodology, developed by Sen (2012) to evaluate climatic trends, is used to examine changes between consecutive time series. Since this method does not require any assumptions, it can be applied to all data series, regardless of whether they exhibit internal dependency (Dabanlı, 2017). When applying the method, the time series for which the trend is to be determined is first divided into two parts, and each half-series is sorted in ascending order. Then, a scatter plot is created, with the values of the first half-series on the x-axis and the values of the second half-series on the y-axis. The 45° trend line is added to the scatter plot. The location of the data points relative to the trend line determines the trends in the data. If the data points lie on or close to the 45° trend line, this indicates that there is practically no trend in the series. Data points located in the triangular area above the 45° trend line indicate an increasing trend, while those in the triangular area below the line indicate a decreasing trend. In cases where the scatter points are very close to the 45° trend line, the relative error (%) between the means of the two half-series is considered an important trend indicator to prevent misinterpretations from visual assessment (Sen, 2020). This value is calculated using Eq. (5).

$$\alpha = \left|\frac{\bar{X}_1 - \bar{X}_2}{\bar{X}_2}\right| \times 100 \tag{5}$$

In Eq. (5), α, $\bar{X}_1$ and $\bar{X}_2$ represent the relative error (%), arithmetic means of the first and second half-series, respectively. In practical applications, when $\alpha < 5\%$ the time series shows no significant trend. When $\alpha > 5\%$, if $\bar{X}_1 > \bar{X}_2$, the time series shows a decreasing trend, and if $\bar{X}_1 < \bar{X}_2$, it shows an increasing trend.

3. Results and Discussions

The Mann-Kendall test and ITA were performed using MATLAB 2020b software. A significance level of 5% was considered to evaluate of analysis results of DO, WT, Q, and EC data. The trend analysis results for the monthly, seasonal, and annual means of discharge and water quality data based on the Mann-Kendall test are presented in Table 2.

For the Broad River (upstream), DO concentrations showed an increasing trend in Jan, Feb, Jul, Sep, Nov, and Dec, while WT values have an increasing trend in Apr, Jun, Sep, and Oct. A decreasing trend in EC values was detected only in Aug. Additionally, WT values showed increasing trends in all seasons, while DO concentrations have increasing trends in all seasons except for Spr. No significant annual trends were detected, except for the increasing trends in DO concentrations and WT values.

For the Tyger River (tributary), a decreasing trend was found in Q values only in Mar. There was an increasing trend in DO concentrations in Aug, Sep, and Nov and WT values in all months except Jan, Feb, Mar, and Nov. In addition, it was determined that WT values had increasing trends in all seasons and DO concentrations had increasing trends in Sum and Aut. No significant trends were found for other parameters and periods. Except for the increasing trends in DO concentrations and WT values, no significant trends were detected annually.

For the Enoree River (tributary), a decreasing trend was detected in Q values only in Mar. WT values showed an increasing trend in all months except Jan, Feb, Mar, and Nov, while EC values had an increasing trend except in Aug and Dec. Seasonally, EC values showed an increasing trend in all seasons, while WT values have increasing trends in all seasons except Win. Annually, no significant trends were observed in DO and Q values, but increasing trends were detected for WT and EC values. No significant increasing or decreasing trends were found for DO concentrations in any period.

For the Broad River (downstream), Q values showed a decreasing trend in Mar, but no significant trends were observed in other months. EC values have a decreasing trend in Aug and Sep. Seasonally, no significant trends were detected except for EC values, which has a decreasing trend in the Aut, while the annual EC values also showed a decreasing trend. WT values showed an increasing trend for all months, except for Feb, Nov, and Dec. No significant increasing or decreasing trends were detected for DO concentrations on a monthly, seasonal, or annual basis.

The α values of monthly, seasonal, and annual trend analyses of discharge and water quality data according to the ITA method are given in Table 3, in which negative values mean decreasing trend while positive values mean increasing trend. For each station, 68 data series (four parameters and 17 time periods) were analysed, and an increasing trend was found for DO (20.6%), WT (41.2%), Q (8.7%), and EC (27.9%). Additionally, a decreasing trend was detected for Q (70.6%) and EC (51.5%).

Table 2. The Mann-Kendall test Z values of monthly, seasonal, and annual means of discharge and water quality data for the Broad River Basin, South Carolina, USA

Çizelge 2. *Broad Nehri Havzası (Güney Carolina, ABD) debi ve su kalitesi verilerinin aylık, mevsimlik ve yıllık ortalamalarının Mann-Kendall testi Z değerleri*

Parameter	Period	USGS 02156500	USGS 02160105	USGS 02160700	USGS 02160991	Parameter	Period	USGS 02156500	USGS 02160105	USGS 02160700	USGS 02160991
DO	Jan	***2.67***	0.49	0.89	–1.29	Q	Jan	–0.31	–0.98	–0.46	–0.67
	Feb	***3.36***	1.42	0.95	1.03		Feb	–0.29	–1.05	–0.49	–0.84
	Mar	1.65	0.51	0.15	0.70		Mar	–1.84	***–2.49***	***–2.10***	***–2.00***
	Apr	1.57	1.16	–0.44	–0.19		Apr	0.56	–0.22	0.08	0.08
	May	0.38	0.82	–1.95	–0.26		May	0.53	0.16	0.35	0.22
	Jun	1.21	0.54	–0.40	0.42		Jun	–0.20	–0.80	–0.25	–0.53
	Jul	***3.58***	1.59	–0.15	0.11		Jul	–0.34	–0.44	0.11	–0.64
	Aug	***2.98***	***2.60***	0.62	–0.03		Aug	–0.14	–0.48	0.10	–0.23
	Sep	***3.73***	***3.02***	1.93	0.42		Sep	–0.86	–1.85	–1.39	–1.09
	Oct	1.29	1.71	0.59	–0.64		Oct	–0.61	–1.74	–1.43	–1.10
	Nov	***3.62***	***2.91***	1.60	1.22		Nov	–0.22	–0.79	–0.46	–0.64
	Dec	***2.30***	–0.51	–0.14	0.56		Dec	0.64	0.12	0.50	0.12
	Win	***3.01***	1.38	0.38	–0.22		Win	–0.18	–0.91	–0.18	–0.64
	Spr	1.35	1.15	–0.53	0.23		Spr	–0.18	–1.05	–0.79	–0.83
	Sum	***3.06***	***2.13***	–0.04	0.30		Sum	–0.26	–0.86	–0.01	–0.45
	Aut	***4.48***	***3.18***	1.50	0.68		Aut	–0.53	–1.63	–1.02	–0.99
	Ann	***4.02***	***2.64***	0.46	0.42		Ann	–0.18	–1.27	–0.67	–0.83
WT	Jan	0.91	1.19	0.98	***2.43***	EC	Jan	–0.20	0.63	***3.42***	–0.34
	Feb	0.59	0.83	1.47	1.32		Feb	–0.15	0.71	***3.04***	–0.50
	Mar	1.65	1.88	1.70	***1.96***		Mar	0.50	1.17	***3.76***	0.01
	Apr	***2.19***	***2.84***	***3.10***	***2.81***		Apr	–0.97	–0.29	***2.70***	–0.94
	May	1.93	***3.15***	***3.34***	***2.54***		May	–1.65	–0.78	***2.57***	–1.48
	Jun	***2.49***	***3.83***	***3.76***	***3.40***		Jun	–1.19	–0.35	***2.93***	–1.83
	Jul	1.54	***3.26***	***3.37***	***2.65***		Jul	–0.99	–0.60	***2.13***	–1.27
	Aug	1.70	***3.33***	***3.20***	***2.58***		Aug	***–2.25***	–1.24	1.47	***–2.02***
	Sep	***2.66***	***3.85***	***4.46***	***2.96***		Sep	–1.48	0.07	***2.96***	***–2.32***
	Oct	***3.99***	***3.79***	***3.81***	***3.86***		Oct	–0.99	0.97	***4.09***	–1.46
	Nov	0.80	0.72	0.03	1.61		Nov	–1.38	–0.16	***2.63***	–1.89
	Dec	1.87	***2.18***	***1.99***	1.36		Dec	–1.04	–0.68	1.93	–1.48
	Win	***1.98***	***2.13***	1.79	***2.33***		Win	–0.20	0.53	***3.06***	–0.59
	Spr	***2.36***	***3.56***	***3.84***	***2.98***		Spr	–1.10	0.01	***3.09***	–1.13
	Sum	***2.30***	***4.21***	***4.48***	***3.28***		Sum	–1.65	–0.80	***2.34***	–1.92
	Aut	***3.61***	***3.94***	***3.58***	***3.54***		Aut	–1.10	0.45	***3.75***	***–1.98***
	Ann	***3.61***	***5.03***	***5.03***	***4.11***		Ann	–1.38	–0.18	***3.64***	***–2.04***

Bold italic: Significant trend for a confidence level of 95%
Critical z value is 1.96

Table 3. Relative error (%) values of innovative trend analysis for monthly, seasonal, and annual means of discharge and water quality data in the Broad River Basin, South Carolina, USA.
Çizelge 3. Broad Nehri Havzası (Güney Carolina, ABD) debi ve su kalitesi verilerinin aylık, mevsimlik ve yıllık ortalamalarının yenilikçi trend analizi yöntemi α değerleri

Parameter	Period	USGS 02156500	USGS 02160105	USGS 02160700	USGS 02160991	Parameter	Period	USGS 02156500	USGS 02160105	USGS 02160700	USGS 02160991
	Jan	***5.32***	1.80	1.85	–1.49		Jan	–4.93	***–20.75***	***–8.94***	***–16.00***
	Feb	***7.50***	4.13	3.26	2.67		Feb	***–7.60***	***–28.44***	***–16.10***	***–20.33***
	Mar	4.90	2.06	1.24	3.00		Mar	***–36.11***	***–54.84***	***–43.40***	***–54.43***
	Apr	***6.19***	1.98	0.32	0.13		Apr	***–11.14***	***–26.39***	***–17.11***	***–26.19***
	May	3.94	2.44	–1.83	–0.58		May	2.40	***–10.80***	3.51	–3.05
	Jun	4.84	0.97	0.85	1.33		Jun	***–17.28***	***–21.51***	0.60	***–25.80***
	Jul	***7.34***	4.16	2.27	1.21		Jul	***6.37***	***–14.42***	1.80	***–5.37***
	Aug	***5.96***	4.48	0.78	–0.68		Aug	***–23.93***	***–71.26***	***–42.51***	***–39.21***
DO	Sep	***6.09***	4.04	2.89	0.96	Q	Sep	***–46.90***	***–105.21***	***–70.11***	***–59.92***
	Oct	3.18	4.94	3.16	1.64		Oct	***–40.72***	***–80.06***	***–44.22***	***–54.74***
	Nov	***8.05***	***7.76***	4.27	2.71		Nov	***6.95***	***–6.34***	1.15	0.09
	Dec	4.53	0.25	0.05	0.56		Dec	***25.69***	***19.07***	***23.37***	***21.24***
	Win	***5.92***	2.34	1.74	0.75		Win	4.56	***–10.53***	–0.97	***–5.92***
	Spr	***5.05***	2.15	0.02	1.01		Spr	***–16.35***	***–33.38***	***–23.93***	***–30.64***
	Sum	***6.04***	3.19	1.29	0.66		Sum	***–11.24***	***–33.15***	***–11.26***	***–22.99***
	Aut	***5.90***	***–5.76***	3.51	1.87		Aut	***–20.89***	***–47.90***	***–28.13***	***–30.61***
	Ann	***5.67***	3.23	1.66	1.01		Ann	***–9.20***	***–26.90***	***–14.11***	***–20.31***
	Jan	***6.31***	***7.56***	***7.89***	***11.97***		Jan	***–9.16***	2.06	***17.41***	***–10.62***
	Feb	0.70	1.46	3.83	***5.72***		Feb	–3.70	2.48	***18.06***	***–8.54***
	Mar	4.92	4.54	4.96	***6.30***		Mar	–0.09	***5.73***	***18.64***	–4.89
	Apr	4.54	4.71	***6.26***	4.87		Apr	***–9.02***	–0.22	***16.28***	***–8.55***
	May	3.09	4.94	***5.73***	2.93		May	***–13.47***	***–5.87***	***15.36***	***–11.30***
	Jun	***6.12***	***6.58***	***6.51***	3.96		Jun	***–12.58***	–3.83	***20.40***	***–12.73***
	Jul	2.34	4.02	3.56	2.39		Jul	***–14.23***	***–5.75***	***17.00***	***–12.55***
	Aug	3.52	4.47	4.40	3.39		Aug	***–23.18***	***–13.94***	***11.84***	***–15.07***
WT	Sep	***5.27***	***5.74***	***6.26***	4.56	EC	Sep	***–20.20***	1.91	***22.74***	***–15.18***
	Oct	***7.61***	***7.54***	***7.47***	***6.89***		Oct	***–11.54***	***5.23***	***25.21***	***–13.53***
	Nov	0.25	–0.88	–2.75	3.80		Nov	***–16.29***	0.29	***18.37***	***–17.46***
	Dec	***10.83***	***14.76***	***13.54***	***7.17***		Dec	***–11.31***	–0.83	***14.58***	***–16.09***
	Win	***5.71***	***7.35***	***8.01***	***8.49***		Win	***–8.88***	1.50	***16.86***	***–12.30***
	Spr	4.02	4.76	***5.71***	4.42		Spr	***–8.15***	–0.69	***16.59***	***–8.48***
	Sum	3.95	***5.00***	4.79	3.23		Sum	***–16.95***	***–8.02***	***16.29***	***–13.49***
	Aut	4.98	4.92	4.72	***5.17***		Aut	***–16.20***	2.59	***22.40***	***–15.30***
	Ann	4.46	***5.26***	***5.43***	4.68		Ann	***–13.36***	–1.79	***18.21***	***–12.55***

Bold italic: Significant trend for a confidence level of 95%

The Mann-Kendall test and ITA results are presented comparatively for monthly, seasonal, and annual periods in Table 4, in which blue, red, and grey cells indicate increasing, decreasing, and no significant trend, respectively. According to the Table 4, it is seen that there are differences between the analysis results. In total, out of 272-time series (four stations, four parameters, and 17 time periods), significant trends were detected in 88 according to the Mann-Kendall test and in 154 according to the ITA. It identified numerous decreasing trends in Q and EC values, while the Mann-Kendall test mostly did not detect significant trends. This suggests that the ITA is more sensitive in detecting trends compared to the Mann-Kendall test (Achite et al., 2021; San et al., 2021).

The decreasing trends in Q values may be attributed to the effects of climate change. In the study conducted by Gentilucci et al. (2023) in the Upper Potenza Basin (Italy), it was stated that changes in precipitation due to climate change and evaporation losses caused by temperature increases were effective in the decreasing trend in the Q values of the river. The Mann-Kendall test identified more trends, particularly in WT values, compared to the ITA. This discrepancy is likely since the ITA evaluates the data, rather than separately for low, medium, and high values. Although the trends in WT values vary temporally, the increasing trends observed at all stations are likely related to rising air temperatures. This is supported by a study by Isaak et al. (2012), who found that WT in various streams in the northwestern USA were increasing due to rising air temperatures driven by climate change.

Table 4. Comparative results of trend analysis of discharge and water quality data from the Broad River Basin, South Carolina, USA

Çizelge 4. *Broad Nehri Havzası (Güney Carolina, ABD) için debi ve su kalitesi verilerinin trend analizlerinin karşılaştırmalı sonuçları*

USGS station no		02156500				02160105				02160700				02160991			
Method	Period	DO	WT	Q	EC	DO	WT	Q	EC	DO	WT	Q	EC	DO	WT	Q	EC
Mann-Kendall test	Jan	I	N	N	N	N	N	N	N	N	N	N	I	N	I	N	N
	Feb	I	N	N	N	N	N	N	N	N	N	N	I	N	N	N	N
	Mar	N	N	N	N	N	N	D	N	N	N	D	I	N	I	D	N
	Apr	N	I	N	N	N	I	N	N	N	I	N	I	N	I	N	N
	May	N	N	N	N	N	I	N	N	N	I	N	I	N	I	N	N
	Jun	N	I	N	N	N	I	N	N	N	I	N	I	N	I	N	N
	Jul	I	N	N	N	N	I	N	N	N	I	N	I	N	I	N	N
	Aug	I	N	N	D	I	I	N	N	N	I	N	N	N	I	N	D
	Sep	I	I	N	N	I	I	N	N	N	I	N	I	N	I	N	D
	Oct	N	I	N	N	N	I	N	N	N	I	N	I	N	I	N	N
	Nov	I	N	N	N	I	N	N	N	N	N	N	I	N	N	N	N
	Dec	I	N	N	N	N	I	N	N	N	I	N	N	N	N	N	N
	Win	I	I	N	N	N	I	N	N	N	N	N	I	N	I	N	N
	Spr	N	I	N	N	N	I	N	N	N	I	N	I	N	I	N	N
	Sum	I	I	N	N	I	I	N	N	N	I	N	I	N	I	N	N
	Aut	I	I	N	N	I	I	N	N	N	I	N	I	N	I	N	D
	Ann	I	I	N	N	I	I	N	N	N	I	N	I	N	I	N	D
Innovative trend analysis	Jan	I	I	N	D	N	I	D	N	N	I	D	I	N	I	D	D
	Feb	I	N	D	N	N	N	D	N	N	I	D	I	N	I	D	D
	Mar	N	N	D	N	N	N	D	I	N	I	D	I	N	I	D	N
	Apr	I	N	D	D	N	N	D	N	N	N	D	I	N	N	D	D
	May	N	N	N	D	N	N	D	D	N	N	N	I	N	N	N	D
	Jun	N	I	D	D	N	I	D	N	N	N	N	I	N	N	D	D
	Jul	I	N	I	D	N	N	D	D	N	N	N	I	N	N	D	D
	Aug	I	N	D	D	N	N	D	D	N	N	D	I	N	N	D	D
	Sep	I	I	D	D	N	I	D	N	N	N	D	I	N	N	D	D
	Oct	N	I	D	D	N	I	D	I	N	I	D	I	N	I	D	D
	Nov	I	N	I	D	I	N	D	N	N	N	N	I	N	N	N	D
	Dec	N	I	I	D	N	I	I	N	N	I	I	I	N	I	I	D
	Win	I	I	N	D	N	I	D	N	N	I	N	I	N	I	D	D
	Spr	I	N	D	D	N	N	D	N	N	N	D	I	N	N	D	D
	Sum	I	N	D	D	N	N	D	D	N	N	D	I	N	N	D	D
	Aut	I	N	D	D	I	N	D	N	N	I	D	I	N	I	D	D
	Ann	I	N	D	D	N	I	D	N	N	N	D	I	N	N	D	D

I: Increasing trend; N: No trend; D: Decreasing trend

4. Conclusions

In this study, monthly, seasonal, and annual trend analyses of discharge and water quality variables monitored in the Broad River Basin were carried out using the Mann-Kendall test and innovative trend analysis (ITA) methods, and the effects of climate change on the relevant variables were examined. A total of four monitoring stations, two of which on the main branch and the rest on the tributaries, were taken into consideration in the trend analyses. The results of the trend analyses performed using dissolved oxygen (DO), water temperature (WT), discharge (Q), and electrical conductivity (EC) data for the period 1987-2022 are given below.

Significant differences were observed in the detection of significant trends between the Mann-Kendall test and ITA methods. The Mann-Kendall test detected significant trends in 32.4% of the time series, while the ITA method detected significant trends in 64.6%. This suggests that different trend analysis methods used in similar studies may have important effects on the results and therefore, a comparative analysis of various methods would be useful.

Trend analyses reveal that there are increasing trends in WT data and decreasing trends in Q data in the basin. These trends could indicate potential challenges for water resources management in the basin. Additionally, decreasing Q indicates water scarcity and deterioration of water quality, while increasing WT may stress aquatic ecosystems, leading to issues such as reduced DO levels, altered species distribution, and habitat changes. It is thought that these trends may be related to the potential impacts of climate change.

The study contributes to the global discussion on the impacts of climate change on water quality, providing essential insights for sustainable water resource management. In order to carry out similar studies in the streams of Türkiye, long-term and continuous monitoring and data recording should be provided as a priority. Such systems are vital for informing climate change adaptation strategies and ensuring sustainable management of water resources in the future.

Methods and trends can be evaluated more comprehensively by applying current trend analysis methods as well as the Mann-Kendall test and ITA methods in examining the trends of flow and water quality data at the relevant stations in future further studies. In addition, it is thought that making scenario-based assessments and examining flow and water quality trends more comprehensively with future period estimated precipitation and temperature data may be the subject of another study. This will offer a more holistic view of the relationships between climate variables and river health.

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Water Distribution Performance Assessment for Seydişehir Irrigation Association

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Abstract: This study was conducted to evaluate water distribution performance of the Seydişehir Irrigation Association, for the years 2019–2023. Within scope of the performance assessment, various performance indicators, including the amount of water taken into the network, irrigation rate, amount of irrigation water delivered per unit area and amount of irrigation water delivered per unit of irrigated area, were used. The highest amount of water was supplied to the irrigation network in 2019, while the lowest amount was used in 2021. The highest irrigation rate occurred in 2021 as 52.85%, while the lowest rate was in 2019 as 34%. The highest amount of water delivered per hectare was in 2022, while the lowest was in 2019. The amount of irrigation water delivered per unit of irrigated area reached its peak in 2019 as 17,016.83 m³/ha, while the lowest level was recorded in 2021 as 2,637.79 m³/ha. In today's world, where every drop of water has increasing value and plays a critical role, promoting water conservation across all areas of life starting from agriculture, the sector with the highest water use-is vital for the environment, ecosystem and human life. Therefore, evaluating the performance of irrigation associations, which bear this significant responsibility of proper water resource planning in agricultural production, is of utmost importance.

Keywords: Irrigation association, Irrigation performance, Irrigation rate, Irrigation schemes

Seydişehir Sulama Birliğinde Su Dağıtım Performansının Değerlendirilmesi

Öz: Bu çalışma Konya İli, Seydişehir İlçesinde bulunan Seydişehir Sulama Birliği alanındaki sulama şebekesinin 2019-2023 yıllarına ilişkin performansını belirlemek amacıyla yürütülmüştür. Bu kapsamda performans incelemesinde şebekeye alınan su miktarı, sulama oranı, birim alana dağıtılan yıllık sulama suyu miktarı, birim sulanan alana dağıtılan yıllık sulama suyu miktarı incelenmiştir. Sulama şebekesine en fazla su 2019 yılında alınmış olup en az su ise 2021 yılında kullanılmıştır. Yıllar içerisinde en yüksek sulama oranı %52,85 ile 2021 yılında gerçekleşirken bu oranın en düşük olduğu yıl %34 ile 2019 yılına aittir. Hektar başına sağlanan suyun en fazla olduğu yıl 2022 yılı iken en az yıl 2019 yılıdır. Birim sulanan alana dağıtılan yıllık sulama suyu miktarı en yüksek 17016,83 m³/ha ile 2019 yılı iken en düşük 2637,79 m³/ha ile 2021 yılında gerçekleşmiştir. Her bir su damlasının değeri ve kritik rolünün arttığı günümüzde su tasarrufunu suyu en yoğun kullanan tarım sektöründen başlayarak hayatımızın her alanına yaymak çevre, ekosistem ve insan yaşamı açısından son derece önemlidir. Bu yüzden tarımsal üretimde su kaynak kullanımının doğru planlanması bu önemli sorumluluğu üzerine alan sulama birliklerinde performansların değerlendirilmesi büyük önem arz etmektedir.

Anahtar Kelimeler: Sulama birliği, Sulama oranı, Sulama performansı, Sulama şebekeleri,

1. Introduction

Besides economic, social and environmental impacts of ever-increasing population worldwide, climate change-induced droughts and extreme weather events increase the need for natural resources. The unbalanced distribution and scarcity of water in the world adversely affects food production. Access to food and water has become difficult simultaneously with the increasing population. Agricultural production constitutes the building block of nutrition that ensures sustainable life on earth. The first priority of policies aiming sustainable agricultural production is to increase yield per unit area. However, limited arable areas and climate change-induced droughts have revealed the need to improve potential outcomes of irrigation networks.

The surface volume of the world's water resources is 1.36 billion km³. However, only 2.55% (35 million km³) of this amount is available as fresh water. Most of the water on the earth's surface is salt water. Limited nature of these resources and increasing demands exert intense pressures on water management authorities. The primary reason for these pressures is the agricultural sector with the largest share in the inter-sectoral water distribution.

The amount of water available in underground and above-ground resources in Turkiye is 112 billion m^3 and 44 billion m^3 of this amount is used for agricultural, industrial and domestic purposes. Of the amount of water spent annually, 73% is utilized in agricultural sector and this amount is quite high. Annual amount of water per capita is around 1400 m^3 in Turkiye. With this amount, Turkiye is classified among the countries experiencing water scarcity. Therefore, it is necessary to implement realistic reforms and innovations for sustainable use of land and water resources. Application of modern irrigation technologies in the field is the leading innovation in this issue. It is possible to reduce the amount of water used in the agricultural sector from 73% to 60-65% (Candan & Çiftçi, 2018).

Modern irrigation methods should be used to increase irrigation performance levels for sustainable agricultural production. Despite all the struggles to increase the level of irrigated agriculture development, the desired levels have not been reached. Improvement efforts in irrigation are faced with the organizational problems related to operation, maintenance and management together with the high cost of facility and operation expenditures, limitation of water use for agricultural purposes due to industrial use and domestic uses. Such a case then causes a decrease in the success of irrigation systems. To achieve success, water and water management should be the primary concerns of irrigation facilities.

Sustainable water management aims to protect the existing water availability of any project site in the long term, to store it in a planned manner for future years, to distribute it according to the needs, to use it efficiently and to develop the resources, while at the same time taking an approach in harmony with the whole ecosystem. In this sense, the focus should be on the development of different water resources as well as the consumption of existing resources and long-term strategies should be developed for potential problems.

Although Türkiye is in a position that can be considered relatively sufficient among the world countries in terms of land and water resources, deficient and wrong practices are observed especially in the use of water in agriculture. The most important of these problems is that irrigation rates and irrigation yields remain far below the desired levels or are low. Current data revealed that irrigation rate in agricultural lands in Turkiye was 62% and irrigation efficiency was 42%. Such low values indicate that 58% of the water consumed in agriculture was lost and irrigation management was not effective and sustainable (Kalkınma Bakanlığı, 2013).

International organizations similar to the World Bank, playing active roles in the formation of global water policies, with their strategies supporting privatization policies in water distribution management, have had a visible impact on the spread of irrigation associations. Since the 1980s, the World Bank stated that Turkish governments should take measures to reduce the cost items for investment, facility operation and maintenance in irrigation projects. 'Irrigation Associations', defined as an "innovative" institutional structure in irrigation, and the "Accelerated Irrigation Management Transfer Program", which was put into effect under the leadership of State Hydraulic Works (DSI) in the early 1990s, became the most actual example of the measures taken with this recommendation

The irrigation facilities constructed and opened for irrigation by DSİ within the intensive efforts and high costs spent have been transferred to irrigation associations. However, serious problems have been experienced in the irrigated areas, especially due to the insufficient training level of water users and lack of utilization of modern irrigation techniques. Irrigation practices without considering crop water consumption resulted in soil erosion, salinity and alkalinity. Inability to collect water fees is also resulted in not meeting the operation and maintenance costs of the facilities. Such cases all hampered infrastructure, maintenance and repair works (Değirmenci, 2008).

Analyzing the performance levels of irrigation associations to determine whether the transfer works have achieved their objectives is of great importance in terms of irrigation management. Determination of the problems experienced in irrigation networks contributes to the improvement of irrigation system performance.

The primary objective of this study is to analyze the efficient use of water and soil resources in Konya Plain, which has an important agricultural production capacity. Water and land use efficiency parameters to assess the performance of irrigation association in the plain.

2. Material and Method

2.1. Material

In this study, Seydişehir Irrigations of Seydişehir Irrigation Association (SSB) was used as material. The study area is located in the Konya Closed Basin, on the northern foothills of the Taurus Mountains and in Seydişehir District, which has fertile lands along the Çarşamba Stream. The district, which has large and fertile lands between Lake Beyşehir in the northwest and Lake Suğla in the southeast, is approximately 1120 m above sea level.

The responsibility area of the irrigation association starts from Bektemur neighborhood of Beyşehir District and continues along the Beyşehir-Suğla-Apa (BSA) canal. The irrigations within the irrigation association consist of Gevrekli Irrigation (4438 ha), Seydişehir Gravity Irrigation (7202 ha) and Suğla Gravity Irrigation (9530 ha).

In Seydişehir district, summers are hot and dry and winters are cold and rainy. Although it has the general climate characteristics of the Central Anatolia Region, it shows a transition between the Mediterranean climate and the terrestrial climate. The hottest months of the year are July and August, while the coldest months are January and February. The average annual temperature is 11.8 ºC. Long-term (1960-2012) average rainfall is 750.3 mm (Sarı & İnan, 2011).

The soils of the study area contain all textures from heavy to very light. Heavy textured soils cover half of the cultivated areas and medium textured soils are found in 10% of the area (Anonymous, 1984).

The main irrigation water source of SSB is Beyşehir Lake. Beyşehir-Suğla-Apa (BSA) Canal starts from Beyşehir Regulator and continues until Apa Dam. Beyşehir, Seydişehir, Yalıhüyük, Ahırlı and Akören districts are irrigated by this canal. This canal, which comes to Apa Dam, which is within the borders of Çumra District, is one of the first canals of Turkiye in terms of length and flow rate (Arıaslan, 2022).

With the Bektumur Regulator on the BSA Canal, water is taken by gravity to 4438 ha area of Gevrekli Irrigation. Seydişehir Regulator and Seydişehir Gravity Irrigations (7202 ha) receive water and Saray Regulator and Suğla Gravity Irrigations (9530 ha) feed the main and tertiary canals.

The values regarding the amount of irrigated area, cropping pattern and the amount of irrigation water taken into the network in the period covering the years 2019-2023 in the field of Seydişehir Irrigation Association were obtained from DSİ IV Region KOS Brach Directorate.

In the irrigation network between 2019-2023, sugar beet is the most cultivated plant type with 33%, followed by corn (16%), vegetables (16%) and cereals (14%) (Table 1 and 2).

Table 1. Cropping pattern of irrigated areas
Çizelge 1. *Sulanan Alanlarda Gerçekleşen Bitki Deseni*

	Gevrekli Irrigation					Seydişehir Gravity Irrigation				
Crops	**2019 Irrigated Area (da)**	**2020 Irrigated Area (da)**	**2021 Irrigated Area (da)**	**2022 Irrigated Area (da)**	**2023 Irrigated Area (da)**	**2019 Irrigated Area (da)**	**2020 Irrigated Area (da)**	**2021 Irrigated Area (da)**	**2022 Irrigated Area (da)**	**2023 Irrigated Area (da)**
Cereal	4	107	3716	2865	32	14	1452	7810	9308	95
Legumes	30	337	178	112	284,5	3006	6956	6621	2960	3355,4
Melon	7	37	80	21	14,5	416	146	322	161	86,4
Sugarbeet	8234	6814	8269	4781,23	8503,6	10434	10781	9584	9486,7	10005,7
Sunflower	1		354	17	309,5			284	1128	1687,5
Corn	739	796	726	839	581,1	2254	853	1121	1858	1513,3
Feed crops	703	725	863	757	978,9	1399	1233	1150	831	615,8
Fruits	116	96	93	55	121,5	92	92	153	162	125,3
Vegetables	3196	3664	7101	4042,9	6961,6	5576	5522	6394	4134,4	3641
Total	13030	12576	21380	13490,13	17787,2	23191	27035	33439	30029,1	21125,4

Table 2. Cropping pattern of irrigated areas
Çizelge 2. *Sulanan Alanlarda Gerçekleşen Bitki Deseni*

Suğla Gravity Irrigation						Seydişehir Irrigation Association				
BitkiDeseni	**2019 Irrigated Area (d A)**	**2020 Irrigated Area (d A)**	**2021 Irrigated Area (d A)**	**2022 Irrigated Area (d A)**	**2023 Irrigated Area (d A)**	**2019 Irrigated Area (d A)**	**2020 Irrigated Area (d A)**	**2021 Irrigated Area (d A)**	**2022 Irrigated Area (d A)**	**2023 Irrigated Area (d A)**
Cereal	388	4500	15535	14565	1441	406	6059	27061	26738	1568
Legumes	884	2534	3841	945	1847,7	3920	9827	10640	4017	5487,6
Melon	5187	4966	3984	3962	4284,3	5610	5149	4386	4144	4385,2
Sugarbeet	15412	14723	11299	10156,8	10588,9	34080	32318	29152	24424,73	29098,2
Sunflower	6	100	452	2251	3454,2	7	100	1090	3396	5451,2
Corn	7970	11799	11639	15561	12149,2	10963	13448	13486	18258	14243,6
Feed crops	2234	3239	3617	2505	2885,6	4336	5197	5630	4093	4480,3
Fruits	313	541	271	243	460,8	521	729	517	460	707,6
Vegetables	3865	7154	6449	1850,4	2405,5	12637	16340	19944	10027,7	13008,1
Total	36259	49556	57087	52039,2	39517,2	72480	89167	111906	95558,43	78429,8

2.2. Method

In this study, the water use efficiency of irrigation activities for the years 2018-2023 was analyzed. Performance indicators used in this study are provided in Table 3. The indicators used in the examination of water use efficiency are; irrigation rate, amount of irrigation water delivered per unit area and the amount of irrigation water delivered per unit of irrigated area.

Table 3. Performance indicators used in this study (Burton et al., 2000, Çakmak et al., 2004)
Çizelge 3. *Çalışmada Kullanılan Performans Göstergeleri (Burton ve ark.,2000, Çakmak ve ark.,2004)*

Performance Indicator	Required Data
Water Use Efficiency	
$IR = \frac{\text{Irrigated Area (ha)} x 100}{\text{Irrigation Area (ha)}}$	Irrigated Area Irrigation Area
$WDUA = \frac{\text{Total amount of water delivered}}{\text{Irrigation Area}}$	Daily amount of water delivered Irrigation area
$WDIA = \frac{\text{Amount of water delivered}}{\text{Irrigated Area}}$	Daily amount of water delivered Irrigated area

Note: IR: Irrigation rate; WDUA: Amount of water delivered per unit area (m³/ha); WDIA: Amount of water delivered per unit of irrigated area (m³/ha)

3. Results and Discussion

3.1. Water Use Efficiency Indicators

The irrigation works carried out by Seydişehir Irrigation Association in its area of responsibility in 2018-2023 were evaluated and water use efficiency performance criteria were calculated and given in Tables 4, 5 and 6.

3.2. Amount of Irrigation Water Taken into the Network

The amount of irrigation water taken into the irrigation area of Seydişehir Irrigation Association is 123.321.000 m³ for 7247 ha area in 2019; 109.569.999 m³ for 8915 ha area in 2020; 29.514.240 m³ for 11189 ha area in 2021; 62.366.976 m³ for 9554 ha in 2022 and 55.630.000 m³ for 7841 ha in 2023. In the last 5 years, 44746 ha area has been irrigated and the water taken to the total irrigation area is 281.789.215 m³. At the same time, while the most water was used in 2019, the least water was used in 2021.

3.3. Irrigation Rate

It is the parameter determined by the ratio of irrigated area to irrigation area. The irrigation rates of the irrigations belonging to Seydişehir Irrigation Union are given in Table 4.

Table 4. Performance indicators for water use efficiency (Irrigation-based) (DSİ)
Çizelge 4. *Su Kullanım Etkinliği Performans Göstergeleri (Sulama bazlı) (DSİ)*

Gevrekli Irrigation Irrigation Rate				Seydişehir Gravity Irrigation Irrigation Rate			
Years	**Irrigated Area (ha)**	**Irrigation Area (ha)**	**Irrigation Rate**	**Years**	**Irrigated Area (ha)**	**Irrigation Area (ha)**	**Irrigation Rate**
2019	1303	4438	29,36	2019	2319	7202	32,19
2020	1257	4438	28,32	2020	2703	7202	37,53
2021	2138	4438	48,17	2021	3343	7202	46,61
2022	1349	4438	30,39	2022	3002	7202	41,68
2023	1778	4438	40,06	2023	2112	7202	29,32
Suğla Gravity Irrigation Irrigation Rate				**Seydişehir Irrigation Association Irrigation Rate**			
Years	**Irrigated Area (ha)**	**Irrigation Area (ha)**	**Irrigation Rate**	**Years**	**Irrigated Area (ha)**	**Irrigation Area (ha)**	**Irrigation Rate**
2019	3625	9530	38,03	2019	7247	21170	34,23
2020	4955	9530	51,99	2020	8915	21170	42,11
2021	5708	9530	59,89	2021	11189	21170	52,85
2022	5203	9530	54,59	2022	9554	21170	45,13
2023	3951	9530	41,45	2023	7841	21170	37,04

For 2019, Gevrekli Irrigation had an irrigation rate of 29.36%, Seydişehir Gravity Irrigation had 32.19% and Suğla Gravity Irrigation had 38.03%. Seydişehir Irrigation Association (SSB) had an IR of 34.23% in 2019. In 2020, Gevrekli Irrigation had an irrigation rate of 28.32%, Seydişehir Gravity Irrigation had 37.53%, Suğla Gravity Irrigation had 51.99%. Seydişehir Irrigation Association (SSB) had an IR of 42.11% in 2020. In 2021; Gevrekli Irrigation had an irrigation rate of 48.17%, Seydişehir Gravity Irrigation had 46.61% and Suğla Gravity Irrigation had 59.89%. Seydişehir Irrigation Association had an IR of 52.85% in 2021. In 2022, Gevrekli Irrigation had an irrigation rate of 30.39%, Seydişehir Gravity Irrigation had 41.68%, Suğla Gravity Irrigation had 54.59% and SSB had 45.13%. In 2023, the Gevrekli Irrigation had an IR of 40.06%, Seydişehir Gravity Irrigation had and 29.32%, Suğla Gravity Irrigation had 41.45% and SSB had 37.04%. While 2021 has the highest irrigation rate, 2019 has the lowest rate. The reason for the low irrigation rates is recorded as the fact that the local people do not irrigate because they find the rainfall sufficient according to the data obtained from the DSİ Monitoring and Evaluation Reports. Eliçabuk and Topak (2016) reported that the irrigation rates between 2008 and 2013 varied between 22 - 31.5% and were quite low.

Gençoğlu and Değirmenci (2019), in their research conducted in the irrigation facility constructed and put into operation by DSİ and transferred to Kırıkhan Irrigation Association, determined that the lowest irrigation rate was 33% in 2009 and the highest rate was 89% in 2013.

In the evaluation of the irrigation rate in Çorum Irrigation Association between 2019 and 2022; it was found that the minimum irrigation rate was 27.4% in 2019 and the highest rate was 65.3% in 2021. It was emphasized that 2021 was high due to the fact that water was supplied to the off-grid area (Tanışıklı & Çakmak 2023).

3.4. Amount of irrigation water delivered per unit area (WDUA)

The amount of irrigation water delivered per unit area (WDUA) is calculated as the ratio of total amount of water taken into the network to total irrigation area (Table 5).

Table 5. Amount of water delivered per unit area (m^3/ha)

Çizelge 5. *Birim Alana Dağıtılan Yıllık Sulama Suyu Miktarı (m^3/ha)*

Years	Water taken into network (m^3)	Total Area (ha)	WDUA (m^3/ha)
2019	123.321.000	21170	582,53
2020	109.569.999	21170	517,57
2021	29.514.240	21170	1394,15
2022	62.366.976	21170	2946,01
2023	55.630.000	21170	2627,78

SSB has an irrigation area of 21170 ha. In 2019, total amount of water taken into the network was 123.321.000 m^3 and WDUA was calculated as 5825,27 m^3/ha. In 2020, total amount of water taken into the network was 109.569.999 m^3 and WDUA was calculated as 5175,72 m^3/ha. The values were 29.514.240 m^3 and 1394,15 m^3/ha in 2021, 62.366.976 m^3 and 2946,01 m^3/ha in 2022 and 55.630.000 m^3 and 2627,78 m^3/ha in 2023. As can be inferred from Table 5, the year with the highest amount of water provided per hectare is 2022 with 2946,01 m^3/ha and the year with the lowest amount is 2020 with 517,57 m^3/ha. When the total of 5 years in the Irrigation Association was analyzed, the total area was 105,850 ha. The water delivered per hectare in the irrigation area was 3593,78 m^3/ha. In the study conducted in Asartepe Irrigation Association, as a result of the 4-year evaluation, the lowest amount of annual irrigation water delivered per unit area was reported as 1375 m^3/ha in 2007 and the highest was reported as 6312 m^3/ha in 2005 (Kapan, 2010).

3.6. Amount of irrigation water delivered per unit of irrigated area (WDIA)

The amount of irrigation water delivered per unit of irrigated area (WDIA) is calculated as the ratio of total amount of water taken into the network to irrigated area (Table 5).

Table 6. Amount of water delivered per unit of irrigated area (m^3/ha)

Çizelge 6. *Birim Sulanan Alana Dağıtılan Yıllık Sulama Suyu Miktarı (m^3/ha)*

Years	Water taken into network (m^3)	Irrigated Area (ha)	WDIA (m^3/ha)
2019	123.321.000	7247	17016,83
2020	109.569.999	8915	12290,52
2021	29.514.240	11189	2637,79
2022	62.366.976	9554	6527,84
2023	55.630.000	7841	7094,76

In 2019, total amount of water taken into the network was 123.321.000 m^3 and WDIA was calculated as 17.016,83 m^3/ha on 7247 ha. In 2020, WDIA was calculated as 12.290,52 m^3/ha on 8915 ha. In 2021, WDIA was calculated as 2.637,79 m^3/ha on 11189 ha. In 2022, WDIA was calculated as 6.527,84 m^3/ha on 9554 ha. In 2023, WDIA was calculated as 7.094,76 m^3/ha on 7841 ha. When the results are analyzed, it was determined that the highest amount of water distributed per hectare was in 2019, while the least amount of water distributed was in 2021. In 2019 and 2020, it was determined that more water was taken into the network than it needed. The total irrigated area in the Irrigation Association for 5 years was 44746 ha. The water distributed per hectare of the irrigated area was 8501.37 m^3/ha. In the study covering the years 2007-2018 in Acıpayam Irrigation Association, it was reported that the amount of water distributed per unit of irrigated area varied between 4747 - 9793 m³/ha (average: 7020 m³/ha) (Cengiz & Uçar, 2021).

4. Conclusion and Recommendations

Urgent measures should be taken to realize food safety and sufficiency, which is one of the main independence criteria of the countries. The impact of climate change is felt not only in Turkiye but also all over the world. The danger of drought predicted for the coming years and the decrease in water resources are likely to cause serious problems. Effective water management policies, innovative agricultural techniques and rational sustainable water use between the sectors will be effective in reducing the danger of drought.

Lake Beyşehir, which is the water source of SSB, has not been able to store water at the desired levels due to decreasing precipitation in recent years because of drought and evaporation of water in the lake with high temperatures. Such a case has been very difficult for water users who irrigate from the lake. In the years of drought, it should be aimed to obtain maximum high yield despite minimum yield loss and deficit irrigation with a good irrigation schedule in the agricultural areas of the basin. When planning deficit irrigation, crop development periods should be taken into account and the irrigation schedules should be adjusted very well without causing yield loss in the plant.

The irrigation of Gevrekli Irrigation, Seydişehir Gravity and Suğla Gravity irrigations, for which SSB is responsible, with open canal irrigation system instead of closed irrigation systems causes serious water losses in the conveyance and distribution of water. The concrete in the open canals loses its properties over time, the lack of adequate maintenance and repair works in the canals, siltation accumulation and illegal irrigation by farmers cause intense water losses. Therefore, switching to closed irrigation systems will be effective in reducing water losses and maintenance and repair costs. Another important issue is to provide incentives for modern irrigation methods and irrigation at night or in the early hours of the morning will be effective in saving water. At the same time, it is necessary to make water users aware of irrigation when the plant needs it. Another problem caused by taking more water than needed into the network is the groundwater problem. It is thought that the development of state policies and the implementation of sanctions will be beneficial for the acquisition of this awareness.

In the light of the data obtained in this research, it was determined that the highest irrigation rate of Seydişehir Irrigation Association was in 2021 with 52.85% and the lowest rate was in 2019 with 34.23%. At the same time, the year in which the most water was taken into the network between 2019-2023 is 2019 with 123,321,000 m^3 and 7247 ha irrigated. Especially the fact that the canals of Gevrekli Irrigation are quite old and canalized irrigation is still applied in some lines causes high amounts of in-field water losses.

When the plant cultivation areas were analyzed, a decrease was observed in the last 2 years. It is thought to be the effect of the drought experienced. In the cropping pattern that has been changed due to economic concerns, it would be beneficial to turn to and support the cultivation of cereals with low plant water consumption needs instead of sugar beet and corn, which are the most cultivated plant varieties.

Irrigation planning studies are of serious importance in dry years. In irrigation areas, irrigation water allocation plans should be made according to the highest yield that can be achieved with deficit irrigation.

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Determination of the Effects of Different Salt Sources and Doses on Seedling Characteristics of a Local Common Bean (*Phaseolus vulgaris* L.) Variety

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Abstract: Salinity is a major environmental stress that adversely affects plant growth and development, leading to yield loss in agricultural production. Common bean (*Phaseolus vulgaris* L.), widely cultivated in Turkey, is also sensitive to salinity. In this study, the effects of different salt sources (Calcium Chloride, Magnesium Chloride, Magnesium Sulfate, Sodium Sulfate, Sodium Chloride) and concentrations (40, 50, 60, 70 mM) on the seedling characteristics of a local common bean variety were investigated. In the pot experiment conducted using a factorial design in randomized plots, shoot length, leaf length, leaf width, shoot fresh weight, leaf fresh and dry weight, plant water and dry matter content, and chlorophyll content were determined. The results indicate that shoot length ranged between 7.10-11.80 cm (interaction), 9.43-11.06 cm (dose), and 8.80-10.75 cm (sources); leaf length varied between 4.07-6.40 cm (interaction), 5.17-5.90 cm (dose), and 4.95-5.78 cm (sources); leaf width was between 3.67-5.43 cm (interaction), 4.46-4.87 cm (dose), and 4.12-4.91 cm (sources); shoot fresh weight ranged from 0.80-2.07 g (interaction), 1.00-1.43 g (dose), and 1.10-1.46 g (sources); leaf fresh weight varied between 0.22-0.54 g (interaction), 0.30-0.45 g (dose), and 0.29-0.42 g (sources); leaf dry weight was in the range of 0.03-0.86 g (interaction), 0.07-0.33 g (dose), and 0.06-0.31 g (sources); seedling water content ranged between 83-97% (interaction), 85-86% (dose), and 84-88% (sources); dry matter content ranged between 03-18% (interaction), 14-15% (dose), and 12-16% (sources); and chlorophyll content ranged from 37.23-48.17 SPAD (interaction), 41.15-44.15 SPAD (dose), and 40.87-44.66 SPAD (sources). These findings reveal the negative effects of salt stress on plant growth and physiology.

Key words: Dry bean, seedling development, salt concentration, salinity stress

Farklı Tuz Kaynakları ve Dozlarının Yerel Kuru Fasulye (*Phaseolus vulgaris* L.) Çeşidindeki Fide Özelliklerine Etkilerinin Belirlenmesi

Öz: Tuzluluk, bitkilerin büyüme ve gelişimini olumsuz etkileyen önemli bir çevresel strestir ve tarımsal üretimde verim kaybına yol açmaktadır. Türkiye'de yaygın olarak yetiştirilen kuru fasulye (*Phaseolus vulgaris* L.) bitkisi de tuzluluğa karşı hassastır. Bu çalışmada, farklı tuz kaynakları (Kalsiyum Klorür, Magnezyum Klorür, Magnezyum Sülfat, Sodyum Sülfat, Sodyum Klorür) ve konsantrasyonlarının (40, 50, 60, 70 mM) yerel bir kuru fasulye çeşidinin fide özellikleri üzerindeki etkileri incelenmiştir. Tesadüf parsellerinde faktöriyel deneme desenine göre yapılan saksı çalışmasında, sürgün uzunluğu, yaprak uzunluğu, yaprak genişliği, sürgün yaş ağırlığı, yaprak yaş ve kuru ağırlığı, bitki su ve kuru madde oranı ile klorofil miktarı belirlenmiştir. Sonuçlar, sürgün uzunluğunun 7.10-11.80 cm (interaksiyon), 9.43-11.06 cm (doz), 8.80-10.75 cm (tuz kaynağı); yaprak uzunluğunun 4.07-6.40 cm (interaksiyon), 5.17-5.90 cm (doz), 4.95-5.78 cm (tuz kaynağı); yaprak genişliğinin 3.67-5.43 cm (interaksiyon), 4.46-4.87 cm (doz), 4.12-4.91 cm (tuz kaynağı); sürgün taze ağırlığının 0.80-2.07 g (interaksiyon), 1,00-1.43 g (doz), 1.10-1.46 g (tuz kaynağı); yaprak taze ağırlığının 0.22-0.54 g (interaksiyon), 0.30-0.45 g (doz), 0.29-0.42 g (tuz kaynağı); yaprak kuru ağırlığının 0.03-0.86 g (interaksiyon), 0.07-0.33 g (doz), 0.06-0.31 g (tuz kaynağı); fide su içeriğinin %83-97 (interaksiyon), %85-86 (doz), %84-88 (tuz kaynağı); kuru madde içeriğinin %03-18 (interaksiyon), %14-15 (doz), %12-16 (tuz kaynağı); klorofil içeriğinin ise 37.23-48.17 SPAD (interaksiyon), 41.15-44.15 SPAD (doz), 40.87-44.66 SPAD (tuz kaynağı) arasında değiştiğini göstermektedir. Bu bulgular, tuz stresinin bitki büyümesi ve fizyolojisi üzerindeki olumsuz etkilerini ortaya koymaktadır.

Anahtar kelimeler: Fide gelişimi, kuru fasulye, tuz konsantrasyonu, tuz stresi

1. Introduction

Beans (*Phaseolus vulgaris* L.) are one of the most valuable plant species in the legume family and have been used in human nutrition for thousands of years. Bean seeds contain approximately 20-28% protein and around 60% carbohydrates. They are also rich in various vitamins (A, B, and D). These nutritional elements give beans numerous health benefits (Ceyhan et al., 2009; Pekşen and Artık, 2005). Dry bean production ranks first globally, while in our country, it follows chickpeas and lentils (Çiftci et al., 2023). In 2022, dry bean cultivation in our country covered an area of 970,520 decares, with a production volume of 270,000 tons (TÜİK, 2022).

Bean plants are susceptible to abiotic stresses such as salinity and drought. To mitigate the negative effects of these stress factors on bean plants' physiological processes, comprehensive studies on salinity and drought tolerance are being conducted in agricultural research. As a result of these studies, new strategies and varieties are being developed in areas such as genetic breeding, plant nutrition, and irrigation management, aiming to prevent yield and quality losses in bean cultivation.

Globally, over 800 million hectares of land are affected by salinity issues. Approximately one-fifth (32 million hectares) of the 150 million hectares of dry farming land worldwide is at salinity risk. Similarly, 45 million hectares of the 230 million hectares of irrigated agricultural land are affected by salinity stress (Yılmaz et al., 2011; Munns, 2002). This proportion is expected to continue to increase in the coming years. Salt stress inhibits plant growth and development, lowering the osmotic potential of plant cells due to increased salt concentration and decreased water in the soil. Consequently, a series of responses occur in the plant. In plants exposed to salt stress, many physiological processes, such as germination, cell division, photosynthesis, growth, and development, are affected (Bressan, 2008). These effects vary depending on salt stress's type, duration, or intensity. This study focuses on how different salt sources and doses affect the salinity tolerance of dry bean seedlings. For this purpose, some plant characteristics were examined in dry bean seedlings subjected to salt solutions.

2. Materials and Methods

In this study, local dry bean seeds obtained from Karaköy village in Elmalı district of Antalya were used, and the study was conducted as a pot experiment under greenhouse conditions in Rize. The experiment was set up using plastic pots with a diameter of 16 cm and a depth of 13 cm. Sterile peat from a commercial brand was used in the pots. This peat has a high air ratio, is porous and stable in structure, is composed of fine particles, and offers excellent drainage properties. The pH value of the peat was 6.0, and the amount of fertilizer was determined as 1.0 g/l. Fertilizer Sources to the seedlings was carried out using a commercial brand's Liquid Chicken Manure pH Regulator containing 100% organic matter (chicken manure extract) (Güler, 2004; Taban et al., 2013; Tavalı et al., 2014) A 4% fertilizer solution was added to each pot to meet the nutritional needs of the plants. The experiment was established in a factorial experimental design with three replications in randomized plots. Five seeds were sown in each pot, and salt solutions were applied simultaneously with sowing. The salt sources used in the study were Calcium Chloride, Magnesium Chloride, Magnesium Sulfate, Sodium Sulfate, and Sodium Chloride. Each was applied at doses of 0, 40, 50, 60, and 70 mM, as 166 ml of saline solution per pot in a single Sources. Measurements were made using a precision scale and digital caliper, and the dry weight of the plants was determined by drying at 78°C for 48 hours in an oven. After examining seedling characteristics (shoot length, leaf length, leaf width, shoot fresh weight, leaf fresh and dry weight, plant water content, plant dry matter content, and chlorophyll content), statistical analyses were performed using the JMP software, and multiple comparisons were evaluated using the Tukey test (Yıldırım and Yılmaz 2022; Yıldırım and Yılmaz 2023). Chlorophyll measurement was performed with a SPAD-502 Plus chlorophyll meter from Konica Minolta. Shoot length, leaf length, and leaf width were measured with a digital caliper, and their averages were taken. Shoot fresh weight, leaf fresh and dry weight were measured with a precision scale, and their averages were taken.

3. Results and Discussion

This study examined the effects of different salt sources and doses on various development parameters of bean seedlings. The results showed that sources types and doses significantly affected the plants shoot length, leaf length, leaf width, shoot fresh weight, leaf fresh and dry weight, seedling fresh and dry weight, water and dry matter content, and chlorophyll content.

The effects of different salt sources and doses on the shoot length of bean seedlings were investigated. It was found that Sources, dose, and Sources x dose interactions did not create a statistically significant difference in shoot length (Table 1). The most extended shoot length was measured as 11.80 cm with $MgSO_4$ (50 ml/L), followed by Na_2SO_4 (50 ml/L) (11.75 cm), $MgSO_4$ (60 ml/L) (11.75 cm), and Na_2SO_4 (50 ml/L) (11.73 cm). The shortest shoot length was 7.10 cm with $CaCl_2$ (50 ml/L). Similar findings were reported by Çiftçi et al. (2011) and Eroğlu (2007), who found that salt stress reduced considerably shoot length in bean plants. In addition, Uzun Kayıs and Ceyhan (2015) and Cakır and Ceyhan (2021) reported that salt Sources negatively affected root development in lentils, Aldemir and Ceyhan (2015) in chickpeas and Aydın et al. (2019) in cowpeas. These findings align with our study.

When examining the findings related to leaf length, it was determined that Sources and dose factors created statistically significant differences in leaf length, and Sources x dose interactions had an even, more substantial effect ($p \leq 0.001$). The most extended leaf length was recorded as 6.40 cm with $MgCl_2$ (50 ml/L), followed by $CaCl_2$ (60 ml/L) (6.37 cm) and NaCl (60 ml/L) (6.33 cm) (Table 1). The shortest leaf length was measured as 4.07 cm with $CaCl_2$ (70 ml/L). The doses of $MgCl_2$ (50 ml/L), $CaCl_2$ (60 ml/L), and NaCl (50 ml/L) produced the, most extended leaf lengths, with no statistically significant differences among these sources. In the study by Erol et al. (2024), the effect of salt stress on soybeans was examined and it was found that the soybeans were examined, and it was found that the leaf area index decreased as the salt dose increased. These results demonstrate the negative impact of salt stress on leaf development and are consistent with findings in the literature.

Table 1. Effects of Different Salt Sources and Doses on Shoot and Leaf Length of Bean Seedlings
Çizelge 1. *Fasulye Fidelerinde Farklı Tuz Kaynakları ve Dozlarının Sürgün ve Yaprak Uzunluğu Üzerindeki Etkileri*

Factor		Means of Shoot Length (cm)	Factor	Means of Leaf Length (cm)
Sources	Na_2SO_4	10.75	$MgSO_4$	5.78 a
	$MgSO_4$	10.51	NaCl	5.73 ab
	$MgCl_2$	10.23	Na_2SO_4	5.53 ab
	NaCl	10.13	$MgCl_2$	5.30 ab
	$CaCl_2$	8.80	$CaCl_2$	4.95 b
Dose	60 ml/L	11.06	50 ml/L	5.90 a
	50 ml/L	10.00	60 ml/L	5.52 ab
	70 ml/L	9.85	70 ml/L	5.24 ab
	40 ml/L	9.43	40 ml/L	5.17 b
Sources * Dose Interaction	$MgSO_4$, 50 ml/L	11.80	$MgCl_2$,50 ml/L	6.40 a
	$MgSO_4$, 60 ml/L	11.75	$CaCl_2$,60 ml/L	6.37 a
	Na_2SO_4, 50 ml/L	11.73	NaCl,50 ml/L	6.33 a
	Na_2SO_4, 60 ml/L	11.57	$MgSO_4$,60 ml/L	6.15 ab
	Na_2SO_4, 70 ml/L	11.27	Na_2SO_4,50 ml/L	6.13 ab
	$CaCl_2$, 60 ml/L	11.20	NaCl,40 ml/L	5.83 ab
	$MgCl_2$, 40 ml/L	11.17	$MgSO_4$,50 ml/L	5.80 ab
	$MgCl_2$, 60 ml/L	10.73	$MgSO_4$,40 ml/L	5.75 ab
	NaCl, 70 ml/L	10.46	Na_2SO_4,70 ml/L	5.60 ab
	$MgSO_4$, 70 ml/L	10.35	Na_2SO_4,60 ml/L	5.57 ab
	NaCl, 60 ml/L	10.07	$MgCl_2$,70 ml/L	5.57 ab
	NaCl, 40 ml/L	10.04	NaCl,70 ml/L	5.55 ab
	NaCl, 50 ml/L	9.95	$MgSO_4$,70 ml/L	5.43 ab
	$MgCl_2$, 70 ml/L	9.63	NaCl,60 ml/L	5.20 ab
	$MgCl_2$, 50 ml/L	9.40	$MgCl_2$,40 ml/L	4.93 ab
	$CaCl_2$, 40 ml/L	9.37	$CaCl_2$,50 ml/L	4.85 ab
	Na_2SO_4, 40 ml/L	8.43	Na_2SO_4,40 ml/L	4.83 ab
	$MgSO_4$, 40 ml/L	8.15	$CaCl_2$,40 ml/L	4.50 ab
	$CaCl_2$, 70 ml/L	7.55	$MgCl_2$,60 ml/L	4.30 ab
	$CaCl_2$, 50 ml/L	7.10	$CaCl_2$,70 ml/L	4.07 b
General Information	CV (%)	17.83	CV (%)	13.02
	Sources	Ö.D.	Sources	**
	Dose	Ö.D.	Dose	**
	Sources * Dose	Ö.D.	Sources * Dose	***

Na_2SO_4: Sodium Sulfate; $MgSO_4$: Magnesium Sulfate; $MgCl_2$: Magnesium Chloride; NaCl: Sodium Chloride; $CaCl_2$: Calcium Chloride. N.S., Not Significant, $p \leq 0.05$: **, $p \leq 0.001$: *, CV: Coefficient of Variation

The effects of various salt concentrations on the leaf width of bean seedlings were evaluated, and a statistically significant difference was found in terms of the Sources factor ($p \leq 0.05$) (Table 2). The widest leaf length was recorded as 5.43 cm with $MgCl_2$ (50 ml/L), followed by NaCl (50 ml/L) (5.40 cm) and $MgSO_4$ (40 ml/L) (5.35 cm). The lowest leaf width was measured as 3.67 cm with $CaCl_2$ (40 ml/L).

When the effects of different salt sources and doses on the shoot fresh weight of bean seedlings were examined, no statistically significant difference was found in Sources and Sources*dosage interactions (Table 2). However, statistically significant differences were detected regarding dose ($p \leq 0.05$). Na_2SO_4 (1.46 g) provided the highest shoot fresh weight, followed by $MgCl_2$ (1.42 g) and $MgSO_4$ (1.22 g). The lowest shoot fresh weight was obtained with $CaCl_2$ (1.10 g). In studies by Karakullukçu (2008), it was reported that salt stress significantly reduced the fresh weight of plants. Fidan and Ekincialp (2020) obtained similar results and emphasized the effect of salt stress on root fresh weight. These findings are consistent with other studies in literature. Similarly, Aktaş and Kılıç (2013) investigated the effects of salt on soybean sprout (*Glycine max*) production and reported a significant decrease in shoot and root length as well as fresh weights with increasing NaCl concentration. Under 50 mM NaCl application, the shoot fresh weight decreased by 27% and root fresh weight by 60% in sensitive varieties, while these rates were approximately 15% and 6% in tolerant varieties, respectively. These findings align with previous studies, emphasizing the negative impacts of salt stress on plants.

Table 2. Effects of Different Salt Sources and Doses on Leaf Width (cm) and Shoot Fresh Weight (g) of Bean Seedlings

Çizelge 2. *Fasulye Fidelerinde Farklı Tuz Kaynakları ve Dozlarının Yaprak Genişliği (cm) ve Sürgün Yaş Ağırlığı (g) Üzerindeki Etkileri*

Factor		Means of Leaf Width (cm)	Factor	Shoot Fresh Weight
Sources	$MgSO_4$	4.91 a	Na_2SO_4	1.46
	NaCl	4.89 a	$MgCl_2$	1.42
	Na_2SO_4	4.78 ab	$MgSO_4$	1.22
	$MgCl_2$	4.58 ab	NaCl	1.18
	$CaCl_2$	4.12 b	$CaCl_2$	1.10
Dose	50 ml/L	4.87	50 ml/L	1.43 a
	60 ml/L	4.71	70 ml/L	1.42 a
	70 ml/L	4.58	60 ml/L	1.26 ab
	40 ml/L	4.46	40 ml/L	1.00 b
Sources * Dose Interaction	$MgCl_2$,50 ml/L	5.43	Na_2SO_4,70 ml/L	2.07
	NaCl,50 ml/L	5.40	$MgCl_2$,50 ml/L	1.73
	$MgSO_4$,40 ml/L	5.35	$MgCl_2$,70 ml/L	1.69
	Na_2SO_4,70 ml/L	5.23	$CaCl_2$,60 ml/L	1.64
	$MgSO_4$,60 ml/L	5.15	Na_2SO_4,60 ml/L	1.54
	$CaCl_2$,60 ml/L	5.10	$MgSO_4$,50 ml/L	1.53
	Na_2SO_4,50 ml/L	4.87	Na_2SO_4,50 ml/L	1.40
	NaCl,40 ml/L	4.83	NaCl,50 ml/L	1.36
	NaCl,70 ml/L	4.78	$MgCl_2$,40 ml/L	1.29
	Na_2SO_4,60 ml/L	4.77	NaCl,70 ml/L	1.26
	$MgSO_4$,50 ml/L	4.73	$MgSO_4$,70 ml/L	1.24
	$MgCl_2$,70 ml/L	4.67	$MgSO_4$,60 ml/L	1.21
	NaCl,60 ml/L	4.53	NaCl,40 ml/L	1.18
	$MgSO_4$,70 ml/L	4.40	$CaCl_2$,50 ml/L	1.13
	Na_2SO_4,40 ml/L	4.23	$MgCl_2$,60 ml/L	0.99
	$MgCl_2$,40 ml/L	4.20	NaCl,60 ml/L	0.92
	$MgCl_2$,60 ml/L	4.00	$MgSO_4$,40 ml/L	0.90
	$CaCl_2$,50 ml/L	3.90	Na_2SO_4,40 ml/L	0.85
	$CaCl_2$,70 ml/L	3.83	$CaCl_2$,70 ml/L	0.82
	$CaCl_2$,40 ml/L	3.67	$CaCl_2$,40 ml/L	0.80
General Information	CV (%)	12.89	CV (%)	32.12
	Sources	**	Sources	Ö.D.
	Dose	Ö.D.	Dose	**
	Sources * Dose	Ö.D.	Sources * Dose	Ö.D.

Na_2SO_4: Sodium Sulfate; $MgSO_4$: Magnesium Sulfate; $MgCl_2$: Magnesium Chloride; NaCl: Sodium Chloride; $CaCl_2$: Calcium Chloride. N.S., Not Significant, $p \leq 0.05$: **, $p \leq 0.001$: *, CV: Coefficient of Variation

Analyses of seedling water and dry matter content showed that Sources, dose, and Sources x dose interactions created statistically significant differences in these parameters ($p \leq 0.001$) (Table 4). The highest seedling water content was measured as 97% with $MgSO_4$ (40 ml/L), while the lowest was found as 82% with NaCl (60 ml/L). Similarly, the highest seedling dry matter content was measured as 18% with NaCl (60 ml/L), while the lowest was 3% with $MgSO_4$ (40 ml/L). The study by Kibar et al. (2020) also reported that salt stress decreased water content and increased dry matter content. These findings were consistent with the literature.

It was determined that Sources, dose, and Sources x dose interactions created statistically significant differences in leaf dry weight ($p \leq 0.001$) (Table 3). The highest leaf dry weight was obtained with $MgCl_2$ (60 ml/L) (0.86 g), while the lowest was observed with $CaCl_2$ (70 ml/L) (0.03 g). Differences in growth strategies among various genotypes may explain such variations in weight. Erdal et al. (2000) reported developmental regressions and reductions in plant weights in plants grown under salt stress. These findings are similar to those of our study.

When examining the effects of different salt sources and doses on chlorophyll content, it was determined that Sources and Sources x dose interactions created statistically significant differences in chlorophyll content ($p \leq 0.001$). The highest chlorophyll content was measured as 48.17 with $MgSO_4$ (40 ml/L), while the lowest chlorophyll content was found as 37.23 with $MgSO_4$ (70 ml/L) (Table 5). Acar et al. (2011) showed that chlorophyll content decreased in plants under salt stress.

Table 3. Effects of Different Salt Sources and Doses on Leaf Fresh Weight (g) and Leaf Dry Weight of Bean Seedlings

Çizelge 3. *Fasulye Fidelerinde Farklı Tuz Kaynakları ve Dozlarının Yaprak Yaş Ağırlığı (g) ve Yaprak Kuru Ağırlığı Üzerindeki Etkileri*

Factor		**Means of Leaf Fresh Weight (g)**		**Factor**	**Leaf Dry Weight (g) ortalaması**	
Sources	Na_2SO_4	0.42	a	$MgCl_2$	0.31	a
	NaCl	0.40	ab	Na_2SO_4	0.19	b
	$MgCl_2$	0.39	ab	NaCl	0.09	bc
	$MgSO_4$	0.37	ab	$MgSO_4$	0.09	bc
	$CaCl_2$	0.29	b	$CaCl_2$	0.06	c
Dose	50 ml/L	0.45	a	60 ml/L	0.33	a
	70 ml/L	0.39	ab	50 ml/L	0.11	b
	60 ml/L	0.37	ab	40 ml/L	0.07	b
	40 ml/L	0.30	b	70 ml/L	0.07	b
Sources * Dose Interaction	Na_2SO_4,70 ml/L	0.54	a	$MgCl_2$.60 ml/L	0.86	a
	$MgCl_2$,50 ml/L	0.54	ab	Na_2SO_4.60 ml/L	0.51	b
	NaCl,50 ml/L	0.50	ab	$MgCl_2$.50 ml/L	0.22	bc
	Na_2SO_4,50 ml/L	0.48	ab	NaCl.60 ml/L	0.11	c
	$MgCl_2$,70 ml/L	0.47	ab	$MgSO_4$.40 ml/L	0.10	c
	$CaCl_2$,60 ml/L	0.45	ab	$MgSO_4$.50 ml/L	0.10	c
	$MgSO_4$,50 ml/L	0.43	ab	Na_2SO_4.50 ml/L	0.10	c
	Na_2SO_4,60 ml/L	0.40	ab	NaCl.70 ml/L	0.10	c
	NaCl,40 ml/L	0.38	ab	$CaCl_2$.60 ml/L	0.10	c
	$MgSO_4$,60 ml/L	0.37	ab	NaCl.40 ml/L	0.10	c
	NaCl,70 ml/L	0.36	ab	Na_2SO_4.70 ml/L	0.09	c
	NaCl,60 ml/L	0.35	ab	$MgSO_4$.60 ml/L	0.09	c
	$MgSO_4$,40 ml/L	0.35	ab	$MgCl_2$.70 ml/L	0.08	c
	$MgSO_4$,70 ml/L	0.34	ab	Na_2SO_4.40 ml/L	0.06	c
	$MgCl_2$,40 ml/L	0.28	ab	$MgCl_2$.40 ml/L	0.06	c
	$CaCl_2$,50 ml/L	0.28	ab	$MgSO_4$.70 ml/L	0.06	c
	$MgCl_2$,60 ml/L	0.27	ab	$CaCl_2$.50 ml/L	0.05	c
	Na_2SO_4,40 ml/L	0.27	ab	NaCl.50 ml/L	0.05	c
	$CaCl_2$,70 ml/L	0.22	ab	$CaCl_2$.40 ml/L	0.05	c
	$CaCl_2$,40 ml/L	0.22	b	$CaCl_2$.70 ml/L	0.03	c
General Information	CV (%)	27.83		CV (%)	67.28	
	Sources	**		Sources	***	
	Dose	***		Dose	***	
	Sources * Dose	**		Sources * Dose	***	

Na_2SO_4: Sodium Sulfate; $MgSO_4$: Magnesium Sulfate; $MgCl_2$: Magnesium Chloride; NaCl: Sodium Chloride; $CaCl_2$: Calcium Chloride. N.S., Not Significant, $p \leq 0.05$: **, $p \leq 0.001$: *, CV: Coefficient of Variation

This finding aligns with the results of our research. Similarly, Aşcı and Zambi (2020) conducted a study investigating the effects of different salt concentrations (0, 25, 50, 75, 100, 125, and 150 mM) on chlorophyll and mineral content in some forage pea genotypes. The study determined that the genotype x salt dose interaction was statistically significant for many traits, including chlorophyll content. It was reported that salt stress decreased chlorophyll, phosphorus, potassium content, and the K/Na ratio, while calcium and sodium concentrations increased. Notably, the application of 150 mM salt significantly reduced chlorophyll content in the Töre and Ürünlü genotypes, whereas the SPAD values were not statistically affected in the Gölyazı genotype and the Çaybaşı population. Additionally, in the Turnasuyu population, SPAD values were observed to remain within the same group under salt applications up to 100 mM, but a significant decrease occurred when the dose was increased to 125 mM. Acar et al. (2011) reported that salt concentrations between 0-100 mM NaCl did not affect chlorophyll content in peas, while Najafi et al. (2006) found that 100 and 150 mM NaCl applications significantly reduced chlorophyll content compared to the control, with similar effects observed at these doses. Öztürk et al. (2012) stated that chlorophyll content in peas decreased as the salt dose increased. These findings in the literature also support the results of our study.

Table 4. Effects of Different Salt Sources and Doses on Seedling Water Content (%) and Seedling Dry Matter Content (%) of Bean Seedlings

Çizelge 4. *Fasulye Fidelerinde Farklı Tuz Kaynakları ve Dozlarının Fide Su İçeriği (%) ve Fide Kuru Madde İçeriği (%) Üzerindeki Etkileri*

Factor		Seedling Water Content (%) ortalaması		Factor	Means of Dry Matter Content (%)	
Sources	$MgSO_4$	88	a	NaCl	16	a
	$MgCl_2$	86	b	$CaCl_2$	16	a
	Na_2SO_4	85	c	Na_2SO_4	15	b
	$CaCl_2$	85	c	$MgCl_2$	15	b
	NaCl	84	e	$MgSO_4$	12	c
Dose	40 ml/L	86	a	50 ml/L	15	a
	70 ml/L	85	b	60 ml/L	15	a
	50 ml/L	85	b	70 ml/L	15	a
	60 ml/L	85	c	40 ml/L	14	b
Sources * Dose Interaction	$MgSO_4$.40 ml/L	97	a	NaCl.60 ml/L	18	a
	$MgCl_2$.50 ml/L	87	b	$CaCl_2$.40 ml/L	17	b
	$CaCl_2$.60 ml/L	86	c	$CaCl_2$.50 ml/L	17	b
	$CaCl_2$.70 ml/L	86	c	$MgSO_4$.50 ml/L	17	b
	$MgCl_2$.60 ml/L	86	c	$MgCl_2$.40 ml/L	17	b
	$MgCl_2$.70 ml/L	86	c	NaCl.70 ml/L	17	b
	Na_2SO_4.50 ml/L	86	c	Na_2SO_4.40 ml/L	17	b
	Na_2SO_4.70 ml/L	86	c	$MgSO_4$.60 ml/L	15	c
	$MgSO_4$.70 ml/L	86	c	Na_2SO_4.60 ml/L	15	c
	NaCl.40 ml/L	85	d	NaCl.40 ml/L	15	c
	$MgSO_4$.60 ml/L	85	d	NaCl.50 ml/L	15	c
	NaCl.50 ml/L	85	d	Na_2SO_4.70 ml/L	14	d
	Na_2SO_4.60 ml/L	85	d	$CaCl_2$.60 ml/L	14	d
	NaCl.70 ml/L	83	e	$CaCl_2$.70 ml/L	14	d
	$CaCl_2$.40 ml/L	83	e	$MgCl_2$.60 ml/L	14	d
	$CaCl_2$.50 ml/L	83	e	$MgCl_2$.70 ml/L	14	d
	$MgCl_2$.40 ml/L	83	e	$MgSO_4$.70 ml/L	14	d
	$MgSO_4$.50 ml/L	83	e	Na_2SO_4.50 ml/L	14	d
	Na_2SO_4.40 ml/L	83	e	$MgCl_2$.50 ml/L	13	e
	NaCl.60 ml/L	82	f	$MgSO_4$.40 ml/L	03	f
General Information	CV (%)	0.00		CV (%)	0.000001	
	Sources	***		Sources	***	
	Dose	***		Dose	***	
	Sources * Dose	***		Sources * Dose	***	

Na_2SO_4: Sodium Sulfate; $MgSO_4$: Magnesium Sulfate; $MgCl_2$: Magnesium Chloride; NaCl: Sodium Chloride; $CaCl_2$: Calcium Chloride. N.S., Not Significant, $p \leq 0.05$: **, $p \leq 0.001$: *, CV: Coefficient of Variation

4. Conclusion

This study revealed the effects of different salt types and doses applied to bean seedlings on plant growth parameters. Salt types and doses affected several growth parameters, such as shoot length, leaf length, leaf width, and shoot fresh and dry weight. $MgSO_4$ and Na_2SO_4 sources caused less damage to plant growth parameters, while $CaCl_2$ sources negatively affected plant growth. Generally, the 50 ml/L dose yielded the best results on plant growth. This study emphasizes the importance of ensuring optimal conditions when farming in saline soils. Furthermore, using salt-tolerant plant varieties could be an effective strategy to enhance agricultural productivity.

Table 5. Effects of Different Salt Sources and Doses on Chlorophyll Content of Bean Seedlings
Çizelge 5. *Fasulye Fidelerinde Farklı Tuz Kaynakları ve Dozlarının Klorofil İçeriği Üzerindeki Etkileri*

Factor	Chlorophyll Content		
Sources	$MgSO_4$	44.66	a
	$MgCl_2$	44.07	ab
	$CaCl_2$	43.28	ab
	Na_2SO_4	41.27	ab
	NaCl	40.87	b
Dose	60 ml/L	44.15	a
	40 ml/L	43.62	ab
	50 ml/L	42.39	ab
	70 ml/L	41.15	b
Sources * Dose Interaction	$MgSO_4$.40 ml/L	48.17	a
	$MgSO_4$.60 ml/L	47.57	ab
	$CaCl_2$.40 ml/L	46.93	abc
	$MgCl_2$.60 ml/L	46.40	abc
	$MgSO_4$.50 ml/L	45.67	abcd
	$MgCl_2$.40 ml/L	44.87	abcd
	Na_2SO_4.70 ml/L	44.20	abcd
	NaCl.60 ml/L	44.13	abcd
	$MgCl_2$.50 ml/L	43.10	abcd
	Na_2SO_4.50 ml/L	42.87	abcd
	$CaCl_2$.60 ml/L	42.60	abcd
	$CaCl_2$.70 ml/L	42.00	abcd
	$MgCl_2$.70 ml/L	41.90	abcd
	$CaCl_2$.50 ml/L	41.57	abcd
	NaCl.70 ml/L	40.40	abcd
	NaCl.40 ml/L	40.20	abcd
	Na_2SO_4.60 ml/L	40.07	abcd
	NaCl.50 ml/L	38.73	bcd
	Na_2SO_4.40 ml/L	37.93	cd
	$MgSO_4$.70 ml/L	37.23	d
General Information	CV (%)	6.90	
	Sources	***	
	Dose	**	
	Sources * Dose	***	

Na_2SO_4: Sodium Sulfate; $MgSO_4$: Magnesium Sulfate; $MgCl_2$: Magnesium Chloride; NaCl: Sodium Chloride; $CaCl_2$: Calcium Chloride. N.S., Not Significant, $p \leq 0.05$: **, $p \leq 0.001$: *, CV: Coefficient of Variation

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Beneficial Effects on Growth Performance of Brown Shrimp (*Penaeus aztecus*) Fed Dietary Inulin and Vitamin C

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Abstract: This study evaluated the effects of different dietary doses of inulin, vitamin C and combinations on the growth performance of brown shrimp (*Penaeus aztecus*). The experiment was conducted over a period of 84 days using 27 tanks (20 juvenile shrimp/tank, initial weight: 0.85 g) with a total of 540 individuals. Nine dietary treatments were formulated: I0C0 (control, basal diet); I3C0 (0.3% inulin); I4C0 (0.4% inulin); I0C0.1 (0.1% vitamin C); I0C0.2 (0.2% vitamin C); I3C0.1 (0.3% inulin + 0.1% vitamin C); I4C0.1 (0.4% inulin + 0.1% vitamin C); I3C0.2 (0.3% inulin + 0.2% vitamin C); and I4C0.2 (0.4% inulin + 0.2% vitamin C). Shrimp fed with the I4C0.2 diet exhibited the highest growth performance, achieving the best values for final body weight, weight gain, specific growth rate, protein efficiency ratio, and feed conversion ratio. While the I4C0.2 group showed superior feed efficiency, intermediate improvements were observed in the I3C0.2, I4C0.1, and I0C0.2 groups, which also demonstrated significantly higher performance metrics compared to the control ($P < 0.05$). These findings highlight the potential of inulin and vitamin C as effective feed additives for improving growth performance in brown shrimp, either individually or in combination. The remarkable synergistic effects observed in the I4C0.2 group suggest that this combination promotes better growth and feed efficiency. Future studies should explore the physiological and molecular mechanisms underlying these effects to optimize the application of inulin and vitamin C as functional feed additives.

Keywords: Prebiotic, dietary additives, shrimp, growth, sustainability

İnülin ve C Vitamini ile Beslenen Kahverengi Karideslerin (*Penaeus aztecus*) Büyüme Performansı Üzerine Faydalı Etkiler

Öz: Bu çalışma, farklı diyet dozlarında inülin, C vitamini ve bunların kombinasyonlarının kahverengi karideslerin (*Penaeus aztecus*) büyüme performansı üzerindeki etkilerini değerlendirmiştir. Deneme, 84 gün boyunca toplam 540 birey ile 27 tankta (20 karides/tank, başlangıç ağırlığı: 0.85 g) yürütülmüştür. Dokuz diyet grubu formüle edilmiştir: I0C0 (kontrol, bazal diyet); I3C0 (%0,3 inülin); I4C0 (%0,4 inülin); I0C0.1 (%0,1 C vitamini); I0C0.2 (%0,2 C vitamini); I3C0.1 (%0,3 inülin + %0,1 C vitamini); I4C0.1 (%0,4 inülin + %0,1 C vitamini); I3C0.2 (%0,3 inülin + %0,2 C vitamini); ve I4C0.2 (%0,4 inülin + %0,2 C vitamini). I4C0.2 diyetiyle beslenen karidesler, final ağırlık, ağırlık kazancı, spesifik büyüme oranı, protein etkinlik oranı ve yem değerlendirme oranı açısından en yüksek büyüme performansını sergilemiştir. I4C0.2 grubu üstün yem etkinliği gösterirken, I3C0.2, I4C0.1 ve I0C0.2 gruplarında da orta düzeyde iyileşmeler gözlenmiş ve bu gruplar kontrol grubuna kıyasla anlamlı olarak daha yüksek performans değerleri sergilemiştir ($P < 0,05$). Bu bulgular, inülin ve C vitamininin kahverengi karideslerde büyüme performansını iyileştiren etkili yem katkı maddeleri olarak potansiyelini vurgulamaktadır. I4C0.2 grubunda gözlenen dikkate değer sinerjik etkiler, bu kombinasyonun daha iyi büyüme ve yem etkinliğini desteklediğini göstermektedir. Gelecekteki çalışmalar, bu etkilerin altında yatan fizyolojik ve moleküler mekanizmaları araştırarak inülin ve C vitamininin işlevsel yem katkıları olarak uygulanmasını optimize etmeyi hedeflemelidir.

Anahtar Kelimeler: Prebiyotik, diyet katkı maddeleri, karides, büyüme, sürdürülebilirlik

1. Introduction

Crustaceans, particularly penaeid shrimps, hold a significant position in the global aquaculture industry (Haris et al., 2024). Among these, Pacific white shrimp (*Penaeus vannamei*) leads global shrimp production, followed by tiger shrimp (*Penaeus monodon*). However, the dominance of a few species underscores the need for increasing species diversity in aquaculture to ensure food security and enhance resilience in global food systems (FAO, 2024; Chan et al., 2024). Addressing this need has spurred research efforts to identify alternative penaeid species that hold promise for aquaculture development.

The brown shrimp (*Penaeus aztecus*), a penaeid species native to the Western Atlantic, has emerged as a viable candidate due to its high economic value and adaptability. Recent studies have investigated its potential in aquaculture systems, reporting encouraging findings regarding its growth performance and market potential (Al-Badran et al., 2019; Uludağ & Aktaş, 2021; Durmuş & Aktaş, 2021; Genc et al., 2024a, 2024b). These studies form a basis for exploring how nutritional strategies could optimize the cultivation of brown shrimp and unlock its full potential.

Functional feed additives have gained attention for their role in enhancing productivity and sustainability in aquaculture (Karataş 2024; Kaya et al., 2024). Prebiotics, such as inulin, are known to selectively promote the growth of beneficial gut bacteria, improving nutrient absorption and supporting growth (Gibson et al., 2004; Rohani et al., 2022). Similarly, vitamin C plays a critical role in physiological processes, including growth promotion and maintaining homeostasis (Darias et al., 2011). Studies have consistently demonstrated that optimal dietary levels of vitamin C enhance growth metrics such as final body weight, weight gain, and specific growth rates in various aquatic species (Kong et al., 2021; Cai et al., 2022; Song et al., 2023).

Efforts have been accelerated to strengthen the concept of sustainability in the aquaculture sector. In this context, the use of environmentally friendly feed additives that meet the needs of aquatic animals, prioritize welfare, and support reliable and responsible production is emphasized. Feed additives under investigation are expected to align with the United Nations Sustainable Development Goals (SDGs), which represent a holistic approach to addressing urgent challenges and achieving measurable progress by 2030. The introduction of functional, animal-compatible growth promoters into the feed industry has the potential to help the aquaculture sector minimize waste, improve health and welfare, ensure clean water and sanitation, and mitigate the effects of climate change—all while preventing harm to aquatic life and the environment. These contributions directly support SDGs such as Responsible Consumption and Production. Additionally, this approach could foster partnerships for global and local goals, increasing the likelihood of advancing Decent Work and Economic Growth (United Nations, 2015).

This study is the first application of an alternative shrimp species, *P. aztecus*, for culture, and therefore focuses primarily on growth parameters. In this context, we focus on the potential synergy between inulin and vitamin C, two well-documented feed additives, when combined in basal diet. Although their individual benefits are well-established, limited research has explored their combined effects at varying doses. By investigating these synergistic interactions, this study aims to bridge a critical knowledge gap and provide insights into optimizing the growth performance of brown shrimp.

2. Material and methods

2.1. Basal and supplemented diets

A basal diet and experimental diets supplemented different doses of inulin (Orafti®) and vitamin C (Aromel®) to the basal diet were prepared separately and in combinations. The diets were named as follows: I0C0 (basal diet); I3C0 (basal diet + 0.3% Inulin); I4C0 (basal diet + 0.4% Inulin); I0C0.1 (basal diet + 0.1% vitamin C); I0C0.2 (basal diet + 0.2% vitamin C); I3C0. 1 (basal diet + 0.3% Inulin + 0.1% vitamin C); I4C0.1 (basal diet + 0.4% Inulin + 0.1% vitamin C); I3C0.2; (basal diet + 0.3% Inulin + 0.2% vitamin C) and I4C0.2 (basal diet + 0.4% Inulin + 0.2% vitamin C). The ingredients and proximate composition of the basal diet are presented in Table 1.

Table 1. The composition and proximate analysis of the basal diet

Çizelge 1. *Bazal diyetin bileşimi ve besin madde kompozisyonu*

Ingredients	Basal diet (%)
Fish meal	24
Soy protein concentrate	2
Soybean meal	8
Collagen	1
Corn gluten	7
Pea protein concentrate	2
Wheat flour	47.1
Fish oil	3.7
Soy oil	3.7
Vitamins mineral premix	1
Vitamin C	0.1
Cholesterol	0.4
Total (~)	100
Proximate composition (%)*	
Crude protein	35.33±0.10
Crude fat	10.63±0.23
Ash	5.89±0.19
Crude cellulose	1.57±0.06
Moisture	10.82±0.41

2.2. Experimental culture conditions

The study was conducted in the Ankara University fisheries unit using 27 fiberglass tanks with a water volume of 45 L each. Shrimps were obtained from Iskenderun Technical University and transported to Ankara in plastic tanks (200 L) under aerated conditions. After transportation, the shrimps were fed with the basal diet for 15 days to acclimate to the experimental conditions. Five hundred and forty juvenile shrimp were randomly distributed in triplicates to the experimental tanks (20 individuals each). The shrimp were fed at 08.00, 13.00, and 18.00 h every day during the experiment. The feeding regime was 6% of body weight for days 1-14, 5.4% for days 15-28, 4.8% for days 29-42, 4.2% for days 43-56, 3.6% for days 57-70 and 3% for days 71-84. During the trial, water temperature, salinity, pH and dissolved oxygen indices (YSI® 556, YSI Inc., Yellow Springs, OH, USA) were measured daily at 28.60±0.24°C, 34±1 ppt, 7.99±0.10, 6.49±0.24 mg/L, respectively.

2.3. Growth parameters

At the end of the trial, the Equations 1-5 were used to calculate the growth indices of the shrimps.

Weight gain (WG; g/shrimp) = Final weight (g) − Initial weight (g) (1)

Specific growth rate (SGR; %/day) = ((*ln* (final weight) − *ln* (initial weight))/days) x 100 (2)

Protein efficiency ratio (PER) = Weight gain (g) / Protein intake (g) (3)

Feed conversion ratio (FCR; %) = Feed intake (g) / Weight gain (g) (4)

Survival rate (SR; %) = (Shrimp final number – shrimp initial number) x 100 (5)

2.4. Statistics

The data sets were analyzed using the SPSS 27 statistical software (Chicago, IL, USA) for parametric analyses. Before performing parametric tests such as analysis of variance and Tukey tests, assumptions were evaluated. Normality assumptions were checked by examining skewness and kurtosis statistics for all groups, and the homogeneity of variances was assessed using the Levene test. The data were reported as mean ± standard deviation. A P-value of less than 0.05 was considered statistically significant.

3. Results

Over an 84-day period, the growth performance of brown shrimp subjected to dietary treatments revealed statistically significant differences between groups (Tukey test, Table 2). Each zootechnical parameter demonstrated varying effects of dietary inulin and vitamin C supplementation.

For final body weight (FBW), the I4C0.2 group achieved the highest value (9.59±0.20 g), significantly outperforming all other groups ($p < 0.05$). This was followed by the I3C0.2 (9.27±0.03 g) and I4C0.1 (8.97±0.04 g) groups, which also exhibited significantly higher FBW compared to the control group (7.78±0.01 g). Intermediate improvements were observed in the I4C0 (8.50±0.09 g) and I0C0.2 (8.70±0.18 g) groups, which formed a statistically similar subset. The lowest FBW was recorded in the control group, representing shrimp fed a basal diet without supplementation.

Weight gain (WG) followed a similar pattern, with the I4C0.2 group achieving the highest WG (8.74±0.20 g), significantly exceeding all other groups ($p < 0.05$). This was followed by the I3C0.2 (8.42±0.03 g) and I4C0.1 (8.13±0.05 g) groups, both of which showed substantial gains compared to the control group (6.94±0.01 g). Intermediate WG values were observed in the I4C0 (7.66±0.09 g) and I0C0.2 (7.85±0.18 g) groups, which formed another distinct subset. The control group exhibited the lowest WG.

Specific growth rate (SGR) was highest in the I4C0.2 group (2.88±0.02), which significantly surpassed all other groups ($p < 0.05$). The I3C0.2 (2.84±0.01) and I4C0.1 (2.82±0.01) groups also demonstrated significantly higher SGR values compared to the control group (2.64±0.01). Moderate but statistically significant increases in SGR were recorded in the I4C0 (2.75±0.02) and I0C0.2 (2.77±0.03) groups. The control group exhibited the lowest SGR.

Protein efficiency ratio (PER) was highest in the I4C0.2 group (1.80±0.04), significantly exceeding all other groups ($p < 0.05$). High PER values were also observed in the I3C0.2 (1.73±0.01) and I4C0.1 (1.68±0.01) groups, forming a statistically distinct subset compared to the control group (1.45±0.00). Intermediate values were recorded in the I4C0 (1.58±0.02) and I0C0.2 (1.62±0.04) groups, which were statistically similar but significantly higher than the control. The control group exhibited the lowest PER.

The feed conversion ratio (FCR) was most efficient in the I4C0.2 group (1.59±0.04), indicating optimal feed utilization. This value was significantly better than all other groups ($p < 0.05$). The I3C0.2 (1.65±0.01) and I4C0.1 (1.70±0.01) groups also demonstrated improved FCR values, outperforming the control group (1.97±0.00). Intermediate improvements were observed in the I4C0 (1.81±0.02) and I0C0.2 (1.77±0.04) groups. The control group exhibited the least efficient FCR, highlighting the absence of dietary supplementation.

In terms of survival rate (SR) did not exhibit significant differences between groups ($p > 0.05$). The highest SR values (73.33±7.64% to 73.33±12.58%)

were observed in the I3C0.2, I4C0.1, I3C0.1, and I0C0.2 groups. The I4C0.2 group (73.33±10.41%) showed slightly lower SR values but remained statistically similar to these groups. The control group recorded the lowest SR (65.00±5.00%), suggesting that dietary treatments may have marginally improved resilience or health conditions, although these effects were not statistically significant.

Table 2. Growth parameters of brown shrimp grown in tanks with different feed additive groups for 84 days
Çizelge 2. *Farklı yem katkı gruplarında 84 gün boyunca tanklarda yetiştirilen kahverengi karideslerin büyüme parametreleri*

P/G*	I0C0	I3C0	I4C0	I0C0.1	I0C0.2	I3C0.1	I4C0.1	I3C0.2	I4C0.2
IBW	0.85±0.01	0.85±0.01	0.84±0.02	0.85±0.01	0.86±0.02	0.85±0.02	0.85±0.02	0.85±0.01	0.85±0.02
FBW	7.78±0.01^{a}	7.89±0.07^{a}	8.50±0.09^{b}	7.80±0.02^{a}	8.70±0.18^{bc}	8.63±0.10^{b}	8.97±0.04^{c}	9.27±0.03^{d}	9.59±0.20^{e}
WG	6.94±0.01^{a}	7.04±0.06^{a}	7.66±0.09^{b}	6.95±0.03^{a}	7.85±0.18^{bc}	7.78±0.10^{b}	8.13±0.05^{cd}	8.42±0.03^{d}	8.74±0.20^{e}
SGR	2.64±0.01^{a}	2.65±0.01^{a}	2.75±0.02^{b}	2.65±0.01^{a}	2.77±0.03^{bc}	2.77±0.02^{b}	2.82±0.01^{cd}	2.84±0.01^{de}	2.88±0.02^{e}
PER	1.45±0.00^{a}	1.47±0.01^{a}	1.58±0.02^{b}	1.45±0.01^{a}	1.62±0.04^{bc}	1.61±0.02^{b}	1.68±0.01^{cd}	1.73±0.01^{d}	1.80±0.04^{e}
FCR	1.97±0.00^{e}	1.94±0.02^{e}	1.81±0.02^{d}	1.97±0.01^{e}	1.77±0.04^{cd}	1.78±0.02^{d}	1.70±0.01^{bc}	1.65±0.01^{ab}	1.59±0.04^{a}
SR	65.00±5.00	66.67±7.64	68.33±10.41	70.00±5.00	71.67±12.58	73.33±7.64	73.33±7.64	73.33±12.58	73.33±10.41

*****Parameters and groups:** I0C0 (baal diet); I3C0 (basal diet + 0.3% Inulin); I4C0 (basal diet + 0.4% Inulin); I0C0.1 (basal diet + 0.1% vitamin C); I0C0.2 (basal diet + 0.2% vitamin C); I3C0. 1 (basal diet + 0.3% Inulin + 0.1% vitamin C); I4C0.1 (basal diet + 0.4% Inulin + 0.1% vitamin C); I3C0.2; (basal diet + 0.3% Inulin + 0.2% vitamin C) and I4C0.2 (basal diet + 0.4% Inulin + 0.2% vitamin C). **IBW**, initial body weight (g); **FBW**, final body weight (g); **WG**, weight gain (g); **SGR**, specific growth rate (%/day); **PER**, protein efficiency ratio; **FCR**, feed conversion ratio; **SR**, survival rate (%). The data correspond to the mean ± standard deviation. Means within the same row with different superscript letters indicate significant differences at the level of $P < 0.05$. No significant differences were observed among groups for IW and SR ($P > 0.05$).

4. Discussion

Dietary prebiotics have been extensively studied as growth promoters in aquatic organisms, including shrimp (Wee et al., 2024). For example, dietary supplementation with 0.2–0.4% inulin significantly improved the growth performance of *Penaeus vannamei* after an 8-week feeding trial (Zhou et al., 2020). Similarly, in a 56-day trial, dietary inulin at doses of 4 and 8 mg g^{-1} enhanced growth parameters in *P. vannamei* (Li et al., 2021). Beyond prebiotics like inulin, dietary vitamin C has also been widely recognized for its ability to promote growth in aquatic species such as red swamp crayfish (*Procambarus clarkii*) (Kong et al., 2021) and Pacific white shrimp (*P. vannamei*) (Niu et al., 2009). These studies highlight the potential of functional feed additives to improve aquaculture productivity. Consistent with these findings, the present study demonstrated that dietary supplementation with inulin or vitamin C significantly enhanced growth performance in brown shrimp. The most remarkable finding, however, was that the combination of these supplements provided superior results compared to their individual use. Shrimp fed diets supplemented with 0.4% inulin and 0.2% vitamin C (I4C0.2) exhibited significantly higher growth performance, feed efficiency, and survival rates compared to the control group. Other groups, including I4C0, I3C0.1, I3C0.2, I4C0.1, and I0C0.2, also showed notable improvements, highlighting the versatility of these dietary supplements, particularly when combined or administered at optimal doses.

The superior growth indices observed in the I4C0.2 group likely stem from the synergistic effects of inulin and vitamin C in enhancing nutrient absorption and utilization. This combination appears to promote better growth rates and feed efficiency, a finding consistent with prior studies suggesting that combining functional feed additives can yield synergistic benefits by positively influencing gut microbiota and improving overall health conditions (Yamamoto et al., 2020; Li et al., 2021). As a prebiotic, inulin supports the proliferation of beneficial gut microbiota, improving digestive efficiency and general health (Escamilla-Montes et al., 2021; Wang et al., 2024). Vitamin C, meanwhile, contributes to growth by supporting metabolic processes, immune function, and stress resilience (Ibrahem et al., 2010).

The enhanced specific growth rate (SGR) and protein efficiency ratio (PER) observed in the I4C0.2 group further emphasize the advantages of combining inulin and vitamin C. This combination not only promotes better growth but also facilitates more efficient protein utilization, which is critical for reducing production costs in penaeid aquaculture. Additionally, the low feed conversion ratio (FCR) in the I4C0.2 group supports the hypothesis that this combination improves the shrimp's capacity to convert feed into body mass, thereby optimizing production performance. The improved survival rate (SR) in the I4C0.2 group suggests a potential for enhanced resilience against environmental stress or disease, possibly by maintaining gut health and immune balance.

However, this hypothesis warrants further investigation through detailed studies.

These present study findings underscore the potential of dietary inulin and vitamin C as effective feed additives for enhancing growth in brown shrimp, either individually or in combination. The synergistic effects observed in the I4C0.2 group are particularly noteworthy and highlight the value of integrating these functional additives into aquaculture feeds. Future research should focus on elucidating the physiological and molecular mechanisms underlying these benefits to optimize their application as functional feed additives.

This study, conducted on brown shrimp (*P. aztecus*), a species tested for its performance as a candidate for aquaculture production, demonstrates its alignment with broader topics supporting sustainable aquaculture. Perhaps most importantly, the adoption of these functional feed additives is consistent with global sustainability initiatives. The United Nations' Sustainable Development Goals (SDGs) emphasize the importance of addressing food security, environmental sustainability, and responsible production systems (United Nations, 2015). By improving feed efficiency, growth performance, and resilience in aquatic species, these strategies directly support SDGs such as Zero Hunger (Goal 2), Good Health and Well-being (Goal 3), Life Below Water (Goal 14), and Responsible Consumption and Production (Goal 12). Additionally, FAO's Blue Transformation roadmap underscores the need for sustainable aquaculture practices to meet the growing global demand for aquatic products while maintaining ecological integrity (FAO, 2022). The findings of the present study, which demonstrate improvements in feed efficiency and survival rates in shrimp farming, can be considered consistent with these goals, contributing to sustainable aquaculture practices. Furthermore, the European Green Deal promotes carbon-neutral production practices and sustainable food systems to combat climate change and environmental degradation (European Commission, 2019). The preliminary data from this study suggest that the use of environmentally friendly feed additives can enhance resource efficiency while supporting the reduction of the environmental footprint of aquaculture systems.

5. Conclusion

In conclusion, based on the results of this study, the combined supplementation of inulin and vitamin C is recommended for penaeid shrimp farming. These findings align with international goals that prioritize improving global welfare, encouraging the development of sustainable aquaculture systems that are efficient, resilient, and environmentally responsible through the use of functional feed additives.

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The Effects of Agricultural Drone Use on Agricultural Production, Yield and Profitability: The Case of Samsun Province

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Abstract: It is necessary to switch to digital farming practices to increase the efficiency in agricultural production and to meet the increasing population's food needs. Digital agricultural practices provide significant advantages to farmers at many stages of production. This study analysed the effects of agricultural drone use, a digital farming practice, on farmers' production, yield, and income in paddy production in Samsun province. The results of the study showed that the use of agricultural drones increased gross profit by 107.31% and net profit by 254.61%. In addition, the increase in relative profit from 1.20% to 1.73% revealed that drones increased farmers' competitiveness in some aspects such as homogenous planting, optimum fertilization, and spraying. Although it is not possible to say that all of the increased profit is due to the use of drones, it can be said that this change is largely due to the use of drones. It should be kept in mind that similar analyses should be repeated to fully understand the effects of agricultural drones and that comprehensive training programs and legal regulations should be reviewed for the effective use of this technology.

Keywords: Agricultural drone, digital farming practices, production costs, productivity, profitability.

Zirai Dron Kullanımının Tarımsal Üretim, Verim ve Karlılık Üzerine Etkileri: Samsun İli Örneği

Öz: Artan nüfusun gıda ihtiyacını karşılamak amacıyla tarımsal üretimde verimi artırmak için, dijital tarım uygulamalarına geçiş gerekliliği ortaya çıkmaktadır. Dijital tarım uygulamaları üretimin pek çok aşamasında çiftçilere önemli avantajlar sağlamaktadır. Bu çalışmada, Samsun ilinde çeltik üretiminde, dijital tarım uygulamalarından olan zirai dron kullanımının çiftçilerin üretim, verim ve gelirleri üzerindeki etkileri incelenmiştir. Araştırma sonuçları, zirai dron kullanımının brüt karı %107.31, net karı %254.61 artırdığını göstermiştir. Ayrıca, nispi karın %1.20'den %1.73'e yükselmesi, dronların sağladığı homojen ekim, optimum gübreleme ve ilaçlama gibi avantajlarla çiftçilerin rekabet gücünü artırdığını ortaya koymuştur. Her ne kadar artan karın tamamının dron kullanımından kaynaklandığını söylemek mümkün olmasa da bu değişimin büyük oranda dron kullanımına bağlı olduğu söylenebilir. Zirai dronların etkilerini tam olarak anlamak için benzer analizlerin tekrarlanması gerektiği ve bu teknolojinin etkin kullanımı için kapsamlı eğitim programları ile yasal düzenlemelerin gözden geçirilmesi gerektiği unutulmamalıdır.

Anahtar Kelimeler: Dijital tarım uygulamaları, karlılık, üretim maliyetleri, verimlilik, zirai dron.

1. Introduction

Due to the rapid increase in world population, great challenges occur in meeting food demand. According to United Nations data, the world population is expected to reach approximately 10 billion people by 2050 (Anonymous, 2023). This situation questions the adequacy of current agricultural production methods and shows that food production should be increased by about 50 percent (Hafeez et al., 2023). The agricultural sector plays a critical role in ensuring food security, which is one of the basic needs of humanity. However, increasing population and urbanization lead to a decrease in agricultural areas and natural resources (Özgüven et al., 2021). Under these conditions, increasing productivity in agricultural production is of great importance. While the farming sector faces many challenges such as climate change, environmental degradation, and price fluctuations, the spread of modern technologies in agriculture offers opportunities to overcome these challenges (Pakdemirli et al., 2021).

To increase productivity, factors such as the use of chemical inputs, mechanization, development of irrigation facilities, and expansion of arable areas should be taken into consideration (Hazneci & Arslanoğlu, 2021; Keleş et al., 2023). At this point, innovative applications in digital agricultural technologies have also started to play an important role in agricultural production. In accordance with the EU aims to be climate-neutral 2050, the European Union announced the European Green Deal, which encourages the use of digital technologies in agriculture. In line with these targets, Turkey aims to invest in environmentally friendly technologies and reduce dependence on resource use with the 'Green Deal Action Plan' (Ercan et al., 2019).

Digital agriculture refers to the tools that collect data in a digital environment store, analyse, and transfer this data to the end user (Anonymous, 2021). Data are collected in new-generation technologies such as sensors and drones (Öztekin et al., 2023). Thanks to digital agriculture, it is possible to obtain real information about the planting and harvesting time, meteorological forecasts, soil quality, the number of workers required, and costs in advance (Anonymous, 2021).

In recent years, unmanned aerial vehicles (UAVs) have been adopted by various sectors by facilitating costly and labour-intensive processes (Macit, 2023). The agriculture sector is one of them (Türkseven et al., 2016). Agricultural drone technology, one of the UAVs, contributes to agricultural research and applications by using remote sensing and data analytics. These technologies, which help to improve the working and living standards of farmers in agriculture, are supported by software and hardware developments in Turkey (Yangal et al., 2022). Agricultural drones are mostly used for crop monitoring, sowing and planting, weed control, insect control, plant health, plant analysis, weather forecasts, and yield monitoring (URL 1, 2023).

There are studies on digital farming technologies and the use of agricultural drones in the world. (Alimuzzaman, 2015; Deepak, 2018; Ghazali et al., 2022; Hafeez et al., 2023; Kushvaha et al., 2021; Mhetre et al., 2020; Mogili & Meetali et al., 2020; Puri et al., 2017; Shahrooz et al., 2020; Sharma et al., 2021). When the studies on digital agriculture applications in Turkey to date are examined, digital agriculture (Aldağ et al., 2018; Dayıoğlu et al., 2020; Kılavuz & Erdem, 2019; Kirmikil & Ertaş, 2020; Pakdemirli et al., 2021; Şahin, 2022; Tutkunca & Haydar, 2022; Varnalı, 2024), smart agriculture applications (Çokuysal, 2021; Ercan et al, 2019; Kaya, 2019; Yılmazer & Tunalıoğlu, 2024), agriculture 4.0 (Aday & Aday, 2020; Gökkur, 2019; Yıldız, 2021; Yüksel, 2020), agricultural drone technology (Kaya & Goraj, 2020; Macit, 2023; Özgüven et al., 2021; Türkseven et al., 2016; Ünal, 2024), but no studies focusing on the effects of digital applications in agriculture on agricultural production process and yield. were found. The research aims to reveal the technical and economic effects of agricultural drone use on enterprises, which is one of the digital agriculture applications. In the study, the effects of agricultural drone use on production, yield, costs, profitability, and the willingness of producers to continue the use of agricultural drones were examined. In this respect, the study aimed to reveal with numerical analyses whether the use of drones has positive effects on yield and profitability as mentioned in the literature.

2. Material and Method

2.1. Material

The main material of the research was the data obtained from the questionnaires conducted with the producers using agricultural drones in Bafra and Çarşamba districts of Samsun province. The questionnaires included questions on socio-demographic information, structural and economic characteristics, agricultural drone use, and problems encountered. In addition, data from the Ministry of Agriculture and Forestry and information obtained from observations in the field were also included in the study. In addition, previous domestic and foreign studies on the subject were also utilized.

2.2. Method

2.2.1. The method applied in data collection

Only the Bafra and Çarşamba districts constitute 31.33 percent of the total agricultural lands in 17 districts in Samsun. At the same time, the intensity of agricultural drone use was determined in these two districts. Therefore, Bafra and Çarşamba districts were selected for the study. In the research, according to the complete census method, all enterprises using agricultural drones in Bafra and Çarşamba districts were interviewed. In the region, 27 paddy producers who rented agricultural drones from Helimore Aviation, R&D, Design, Engineering, Technology Manufacturing Industry Limited Company, which was established in cooperation with Ondokuz Mayıs University Technopark in 2019, were interviewed. In addition, 10 paddy producers who rented drones from Tekno Tarım Aviation Company, which just started drone rental activities in 2023, and 23 producers who rented drones from Sonagtech Sönmez Agricultural Technologies Company were interviewed. Research data were obtained from face-to-face interviews with a total of 60 producers. The surveyed producers have been producing paddy for many years. However, most of the producers have rented drones to carry out their agricultural activities between the last 1 and 3 years. The location of. the research area is shown in Figure 1 (Özçelebi & Yılmaz, 2020).

Figure 1. Location of the research area
Şekil 1. *Araştırma alanının konumu*

2.2.2. The method applied in the analysis of the data

In the summer of 2023, the enterprises utilizing agricultural drone application was visited with the officials of the company renting drones and these processes were monitored in the field by the researchers. With the surveys conducted with farmers before and after the application, both the input-output coefficients of the production activity and the effects of agricultural drone application on the production processes and yields of farmers were determined. Within this direction, the possible effects of agricultural drone use on farmer incomes were determined by comparing the production carried out with traditional methods and that carried out using agricultural.

As a result of the surveys and observations made in the field, it was observed that agricultural drones are intensively used in paddy production activities in the research region. All of the producers who rent agricultural drones in the region rent drones to use in paddy production. For this reason, firstly, the amount of inputs used in production and the outputs obtained as a result of the use of agricultural drones in paddy production activities were determined. If the same production activity is carried out with traditional methods, the inputs used and the outputs obtained are compared and the effect of drone use on input and output coefficients is revealed. After determining the input and output coefficients, calculations were made based on the market prices of variable inputs. Differences between the use of agricultural drones and traditional methods were examined and their effects on yield and costs were analysed. In addition, the economic impact of drone use was evaluated by calculating the gross, net, and relative profits of the activities.

Production costs are divided into two fixed and variable costs. While fixed costs include costs that do not depend on the amount of production, variable costs vary depending on the amount of production (Erkuş & Demirci, 1985). Variable costs were calculated one by one in the production carried out with the use of agricultural drones and compared with the production carried out with traditional methods. Variable costs included elements such as soil preparation, sowing, spraying, fertilization, seed, fuel oil, irrigation, labour, harvesting, transportation, and marketing. Within the scope of the study, the revolving fund interest, which is included in variable costs, was calculated at half of the interest rate (16.15%) determined by the Türkiye Ziraat Bankası for crop production loans. Fixed costs consist of general administrative expenses and land rent. Administrative expenses were calculated as 3% of variable expenses, and land rent was determined according to the declarations of the producers.

As a result of the study, it was determined that the use of agricultural drones only made a difference in planting, fertilizing, and spraying processes in paddy production. Other production stages (soil preparation, seed, irrigation, harvesting, drying, transportation) were carried out in the same way in both production using drones and traditional production. Therefore, the coefficients for traditional paddy production were determined and costs were calculated by multiplying them with 2023 market prices. Planting, fertilizing, and spraying activities resulting from the use of drones were calculated and analysed with new coefficients.

A profitability analysis was performed to calculate the economic return of agricultural drone use to enterprises, and gross profit, net profit, and relative (proportional) profit were calculated with the help of the equations given below. Gross Production Value (GPV) was obtained by multiplying the product price by the yield values per decare (Açıl & Demirci, 1984; Cinemre, 2013; Hazneci & Arslanoğlu, 2021; Hazneci et al., 2022; Kıral et al., 1999; Tanrıvermiş, 2000). All producers in the research region benefit from fuel and fertilizer support. Fuel-fertilizer support was determined as 271 TL per decare (Anonymous, 2023a) and was added to GVV while conducting the analysis.

Gross profit = GPV - Variable Costs

Net profit = GPV - Total Production Costs

Relative profit = GPV / Total Production Costs

In addition, the reasons for starting and continuing production with agricultural drones, the problems encountered, and the opinions of the producers on the use of agricultural drones were also revealed in the research.

3. Results and discussion

All of the producers interviewed in the study were male. The age of the producers varied between 30 and 72 and the average age was calculated as 55.13. The years of education of the producers varied between 5 and 16, and it was determined that the average education of the producers was 8.81 years. The average agricultural land size of the analysed farms is 24.5 hectares. It was observed that the smallest land size was 80 ha and the largest land size was 600 ha among the enterprises analysed in the research region. 43 business managers who participated in the research are only engaged in agriculture. While 6 people work as civil servants along with agriculture, 11 people work as tradesmen along with agriculture. It was determined that all of the enterprise managers in the research region have social security. 57% of the producers are covered by BAĞ-KUR, 32% by SGK, and 11% by Retirement Fund (Table 1)

Within the scope of the research, the effects of the use of agricultural drones on the production process were personally observed in the field and determined by interviewing farmers one-on-one. It has been determined that the use of agricultural drones differs from traditional methods in sowing, fertilization, and agricultural control of paddy crops. In the sowing stage, traditional methods usually require the use of human or agricultural machinery. In these methods, seed placement and sowing depth control usually depend on certain standards. In contrast, agricultural drones used in the research area were observed to analyse the characteristics of the fields using sensitive sensors and data analytics and ensure that the seeds are placed at optimum depth and spacing. In this way, homogenous planting is achieved and waste of resources is prevented

Table 1. Socio-demographic characteristics of enterprises

Çizelge 1. *İşletmelerin sosyo-demografik özellikleri*

	Mean Frequence (N)	Standard Deviation / Percentage (%)
Age (years)	55.13	10.95
Education Period (years)	8.81	3.72
Land Size (ha)	24.53	12.89
Occupation		
Farmer	43	71.67
Civil Servant	6	10
Tradesman	11	18.33
Total	60	100
Social Security		
SGK	19	31.67
BAĞ-KUR	34	56.67
Retirement Fund	7	11.66
Total	60	100

In the process of fertilization, traditional methods are usually done with a fertilizer spreader. This method causes some areas to be over- or under-fertilised. This leads to both fertilizer wastage and negative effects on soil fertility. However, agricultural drones use sensitive sensors to determine plant needs in different parts of the field and perform point fertilization accordingly. Thus, it has been observed that fertilizer is used more effectively and environmental impacts are minimized.

Traditional methods in agricultural control are generally done by spraying with a pulveriser. This method causes the same amount of spraying of healthy areas and areas where plant pests are dense. For this reason, some areas are sprayed more than necessary or not sprayed enough. In contrast, agricultural drones use advanced sensors and imaging technology to precisely identify pest hotspots. By applying pesticides only to the areas where they are needed, they optimize chemical use, reduce costs, and minimize environmental impacts, resulting in more effective pest control. According to the research findings, it was determined that productivity in paddy production increased significantly as a result of agricultural drone application (Table 2). Although it is not possible to attribute the entire increase in yield to the use of agricultural drones, it can be said that it is largely due to these practices.

Table 2. Cost and profitability indicators of paddy production activities before and after the use of agricultural drones

Çizelge 2. *Zirai dron kullanımı öncesi ve sonrasında çeltik üretim faaliyetinin maliyet ve karlılık göstergeleri*

Cost and Profitability Measurements (TL/ha)*	Before Agricultural Drone Use			After Agricultural Drone Use		
	Amount	Ratio 1 (%)**	Ratio 2 (%)***	Amount	Ratio 1 (%)**	Ratio 2 (%)***
Deep plowing and labor	4202.26	5.64	4.13	4202.26	6.02	4.34
2nd plowing and labor	2826.92	3.80	2.78	2826.92	4.05	2.92
Rice levee and labour	1779.47	2.39	1.75	1779.47	2.55	1.84
Raking and labour	655.28	0.88	0.64	655.28	0.94	0.68
Sowing and labour	10796.67	14.50	10.62	9290.83	13.32	9.59
Harvesting and labour	5941.00	7.98	5.84	5941.00	8.52	6.13
Drying and labour	3114.89	4.18	3.06	3114.89	4.47	3.22
Transport and labour	894.88	1.20	0.88	894.88	1.28	0.92
Irrigation and labour	4182.00	5.62	4.11	4182.00	6.00	4.32
Fertilisation and labour	22188.07	29.79	21.82	21737.23	31.16	22.44
Agricultural pest control and labour	7535.00	10.12	7.41	5433.33	7.79	5.61
Revolving fund interest	10354.81	13.90	10.18	9699.38	13.90	10.01
Total variable costs	*74471.25*	*100.00*	*73.22*	*69757.47*	*100.00*	*72.03*
General administration expenses	2234.14	8.20	2.20	2092.72	7.72	2.16
Land lease	25000.00	91.80	24.58	25000.00	92.28	25.81
Total fixed costs	*27234.14*	*100.00*	*26.78*	*27092.72*	*100.00*	*27.97*
Total production costs	*101705.39*		*100.00*	*96850.19*		*100.00*
Yield (kg/da)	6926.70			8040.80		
Sale price	17.17			20.50		
Gross value of production	118931.44			164836.40		
Support income	2710.00			2710.00		
Gross profit	47170.19			97788.93		
Net profit	19936.05			70696.21		
Relative profit (%)	1.20			1.73		

*1$ = 28.06 TL (24.10.2023)
** Ratio 1: Rates in variable and fixed costs
*** Ratio 2: Rates in total production costs

The differences between traditional farming methods and the use of agricultural drones are significant only in sowing, fertilization, and pest control processes. It was determined that the costs did not change in other cost items. While the sowing cost was 10796.67 TL/ha before the use of agricultural drones, this cost decreased to 9290.83 TL/ha afterward. Fertilization cost decreased from 22188.07 TL/ha to 21737.23 TL/ha and agricultural pest control cost decreased from 7535 TL/ha to 5433.33 TL/ha. These cost reductions indicate a 6.3% decrease in total variable costs (Table 2).

An increase was observed in yield after the use of agricultural drones. While the yield was 692.67 kg/ha before the use of agricultural drones (Anonymous, 2023a; Anonymous, 2023b), this yield increased to 804.08 kg/ha after the use of drones. However, an increase was also observed in the sale price. According to the statements received from the producers who did not use agricultural drones, the average sales price was 17.17 TL/kg, while this price increased to 20.50 TL/kg after the use of agricultural drones. The reason for the increase in sales price is due to the increase in yield. Agricultural drones enable more effective monitoring and management of agricultural lands, enabling homogenous planting, optimum fertilization, and agricultural control. According to the statements received from farmers, these factors positively affected productivity and consequently sales prices by about 19% (Table 2).

According to the research, while the total production cost was 101705.39 TL/ha before the use of agricultural drones, it was determined that these costs decreased by approximately 5% to 96850.19 TL/ha after the use of agricultural drones. While the gross profit was 47170.19 TL/ha before the use of agricultural drones, it was observed that the gross profit increased by 107.31% to 97788.93 TL/ha after the use of agricultural drones. In a study conducted in Çanakkale province, it was stated that paddy producers earned a gross profit of 12724.40 TL/ha with the support income. (Semerci, 2021). Net profit increased by 254.61% from 19936.05 TL/ha to 70696.21 TL/ha. According to the results of the research, relative profit increased from 1.20 percent to 1.73 percent. Accordingly, while the farmer earns 1.20 TL for 1 TL invested in traditional paddy production, he earns 1.73 TL for 1 TL invested in paddy production using agricultural drones (Table 2) In a study conducted in a nearby geography (Sinop province), the proportional profit of paddy production was found to be 1.56 (Yuzbasioglu & Abaci, 2023).

Forty-five percent of the paddy producers participating in the research have been using agricultural drones for 3 years. About 17% of the producers have been renting drones for 2 years, while 38% of the producers rented drones in the last production year (Table 3). The enterprises decided to use agricultural drones with the advice of their colleagues. It was determined that all of the enterprises were satisfied with the use of agricultural drones and wanted to continue using this technology. Even all of the enterprises want to buy agricultural drones.

Table 3. Duration of use of agricultural drones by enterprises
Çizelge 3. *İşletmelerin zirai dron kullanım süreleri*

Year	Frequency	%
1	23	38.33
2	10	16.67
3	27	45.00
Total	60	100

The advantages and disadvantages of agricultural drone use are shown in Table 4. The general opinion of the surveyed enterprise managers is that agricultural drone technology has an important potential in agricultural activities. According to the interviewed business managers, since the use of agricultural drones offers optimum planting, fertilization, and spraying in unit areas, the continuity of this technology is considered inevitable. It has been observed that agricultural drone applications provide a reduction in the use of inputs (seed, fertilizer, pesticide) and time-saving, although labour costs are higher than traditional agricultural practices. Due to the mentioned contributions to the production process, the use of agricultural drones has become attractive among enterprises. Although the enterprises do not encounter any difficulties while using agricultural drones, some limitations are noteworthy. In particular, it was observed that there are obstacles such as high customs duty and the lack of night flight permits for agricultural drones. It was concluded that the enterprises are interested in digital agriculture practices and expect support from the government in this regard. In general, the enterprises stated that with the help of agricultural drone technology, productivity in agricultural production increased, costs decreased and it will be used more widely in the agricultural sector in the future (Table 4).

Table 4. Advantages and disadvantages of agricultural drone
Çizelge 4. *Zirai dron kullanımının avantaj ve dezavantajları*

Advantages	Frequency	%	Disadvantages	Frequency	%
Offers yield mapping capability	24	40	High customs duty	28	47
Positive impact on occupational health and safety	33	55	Legal obstacles	50	83
Environmentally friendly application	35	58	Lack of night flight permits	60	100
Provides precise and homogenous sowing	60	100	Restrictions in adverse weather conditions	60	100
Provides optimum fertilization and spraying	60	100	High customs duty		
Reduces input costs	60	100			
Saves labour and time	60	100			
Increases agricultural yield	60	100			

4. Conclusion

As a result of the research, it was determined that the use of agricultural drones, which is one of the digital agriculture practices, contributed to the increase in yield and accordingly to the increase in agricultural income of the enterprises. In Turkey, agricultural drones are at the centre of digital farming practices and make significant contributions to agricultural production processes. As a result of the research, it was determined that these technologies increase agricultural productivity by making homogeneous and precise spraying in critical production stages such as sowing, fertilization, and spraying. In addition, the use of agricultural drones will contribute to achieve environmental sustainability goals by preventing waste of resources. As a result of the research, it was also determined that agricultural drones equipped with high-resolution cameras and sensors provide detailed maps of agricultural lands and provide farmers with data on plant health, water, and nutrient deficiencies. This data facilitates more informed decision-making, reducing crop losses and optimizing production processes. In accordance with the study results, it was concluded that product quality and paddy yield increase especially thanks to homogenous planting and fertilisation. This situation will increase the competitiveness of enterprises. In addition, it was observed in the research that the use of agricultural drones directly contributes to both cost reduction and environmental sustainability by optimizing the use of chemicals.

The effectiveness of agricultural drones in agricultural production was also reflected in their impact on the profitability of businesses. According to the research results, the increase in gross profit was achieved through the efficiency and cost optimization of agricultural drone technology. The increases in gross and net profits showed that agricultural drones offer a low-cost and high-efficiency solution. The increase in the relative profit rate revealed that the profitability and business efficiency obtained from each unit of production have increased. Farmers happily adopted this technology and continued to use it. Studies conducted particularly in the Black Sea region have shown that the use of agricultural drones has increased significantly during the transition period from 2022 to 2023, and that this technology is expected to become even more widespread in the coming years. However, similar analyses need to be repeated at regular intervals to clearly understand the long-term effects of this technology. It is expected that educational programs on the use of digital technologies and the improvement of legal regulations on the subject will accelerate the technological transformation in the agricultural sector, allowing for the wider adoption of agricultural drones and other digital technologies.

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Assessment of InDel Variations in CPT1A and PRDM6 Genes Across Three Anatolian Goat Populations

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Abstract: Identifying candidate genes associated with growth traits in livestock and incorporating them into Marker Assisted Selection (MAS) studies, in addition to classical selection methods, can increase genetic progress and thus profitability. Carnitine Palmitoyl Transferase 1a (*CPT1a*) and PR domain family 6 (*PRDM6*) are among the candidate genes reported to be associated with growth traits in goats. This study aimed to identify polymorphisms in the CPT1a and PRDM6 genes related to growth traits in Kıl (HAI), Honamlı (HNM), and Kabakulak (KBK) goats, and to discuss their potential use in MAS studies. The II genotype of the *CPT1a* gene was not found in the HNM or KIL goats, whereas the KBK population included all three genotypes (II, ID, and DD). The frequency of the ID genotype ranged from 0.25 (HAI) to 0.40 (KBK), while the DD genotype frequency ranged from 0.37 (KBK) to 0.75 (HAI). For the *PRDM6* gene, the KBK population was found to be monomorphic, containing only the DD genotype. Although the II genotype was not detected in the HNM and HAI breeds, the ID genotype frequencies were calculated as 0.05 and 0.07, respectively. The study's findings point out that the *CPT1a* gene can be used in MAS studies for HNM, KBK, and HAI goats, whereas the *PRDM6* gene cannot be utilized.

Keywords: *CPT1a*, *PRDM6*, goat, indel, polymorphism

Üç Anadolu Keçisi Populasyonunda CPT1A ve PRDM6 Genlerindeki InDel Varyasyonlarının Değerlendirilmesi

Öz: Çiftlik hayvanlarında büyüme özellikleri ilişkili aday gen bölgelerinin belirlenerek klasik seleksiyon çalışmalarına ek olarak yapılacak Marker Destekli Seleksiyon (MAS) çalışmalarında kullanımı genetik ilerleme hızını ve karlılığı artırılabilir. Keçilerde büyüme özellikleri ile ilişkili olduğu bildirilen aday genler arasında Karnitin Palmitoil Transferaz 1a (*CPT1a*) ve PR domain family 6 (*PRDM6*) genleri de vardır. Gerçekleştirilen çalışmada Kıl (HAI), Honamlı (HNM) ve Kabakulak (KBK) keçilerinde büyüme özellikleri ile ilişkili *CPT1a* ve *PRDM6* genleri üzerindeki indel varyantların neden olduğu polimorfizmlerin araştırılması ve MAS çalışmalarında kullanım olanaklarının tartışılması amaçlanmıştır. *CPT1a* geni için HNM ve KIL keçilerinde II genotipi tespit edilemezken, KBK populasyonunda her üç genotip de (II, ID ve DD) tespit edilmiştir. ID genotip frekansının 0.25 (HAI) ile 0.40 (KBK) aralığında, DD genotip frekansının ise 0.37 (KBK) ile 0.75 (HAI) aralığında değiştiği tespit edilmiştir. *PRMD6* geni için KBK populasyonu monomorf (yalnızca genotip DD) bulunmuştur. HNM ve HAI ırklarında II genotipi tespit edilemezken, ID genotip frekansları sırasıyla 0.05 ve 0.07 olarak hesaplanmıştır. Çalışmadan elde edilen sonuçlar HNM, KBK ve HAI keçilerinde *CPT1a* geninin MAS çalışmalarında kullanılabileceğine ancak *PRDM6* geninin kullanılamayacağına işaret etmektedir.

Anahtar Kelimeler: *CPT1a*, *PRDM6*, keçi, indel, polimorfizm

1. Introduction

Being famous for their higher adaptability to extreme environmental conditions, goats have been an indispensable part of humankind due to their direct and indirect benefits such as providing valuable animal-derived products (meat and milk) and utilizing grasslands efficiently (Karslı & Demir 2024). In Türkiye, goat breeding is mainly practiced with local populations such as Hair (HAI), Honamlı (HNM), and Kabakulak (KBK) across mountainous and forestry regions by making a significant contribution to the incomes of smallholder farmers (Günlü & Alaşahan 2010). Of these goats, HAI, which has the highest population size (nearly 90% of the total goat population) among native breeds and developed adaptation to the harsh climate of Anatolia, is raised in villages near mountains and forests for a dual purpose (Güngör et al. 2021). As highlighted by Demir (2024), unlike the HAI breed, the rest of the local Anatolian goat hold a lower population size. KBK, which is reared in Muğla and Antalya provinces, is believed to be a variety of HAI (Karsli et al. 2020) while it shows differences in terms of several phenotypic traits such as body size, fertility, and economically important yields (Demir 2024). HNM, on the other hand, is bred by smallholder farmers living across the Taurus Mountains of the Mediterranean region covering Antalya, Isparta, and Konya provinces (Daskiran et al. 2018).

The agricultural sector has been negatively affected by numerous factors such as increasing human populations, declining animal populations, and climate change (Demir et al. 2021) whereas farmers exert great efforts to design selection studies to increase the quantity and quality of milk and meat production to meet the demand of the growing societies. Like many other economically important traits, meat yield is affected by the combination of numerous environmental factors and genes due to the nature of quantitative inheritance (Núñez-Torres & Almeida-Secaira 2022). In theory, improving both environmental factors and considering candidate genes in selection studies seems promising to increase meat production in livestock. However, as highlighted by Atay et al. (2023), enhancement of environmental factors may not be practical or affordable by the farmers since it is not inherited by the next generations. Moreover, the improvement of environmental factors should be repeated for the following production seasons. In contrast, selection studies based on candidate genes, called marker-assisted selection (MAS), are an efficient approach to improve quantitative traits including meat yield for the current and future generations. In this approach, animals are genotyped via accurate molecular tools such as polymerase chain reaction (PCR) and subjected to mating programs to create a population with desired genotype combinations regarding multiple genes affecting the trait of interest. Several genes and their receptor regions including insulin-like growth factor (Naicy et al. 2017; Lazar et al. 2018; Alex et al. 2023), growth hormone (Sharma et al. 2013; Yan et al. 2022; Rashijane et al. 2022), and leptin (Sarmah et al. 2020) were previously reported to have direct effects on growth traits in goats. Recently, Carnitine palmitoyltransferase 1A (*CPT1A)* and PR/SET domain family 6 (*PRDM6*) genes were reported to be not only associated with growth traits in goats but also the insertion-deletion (InDel) variations on these genes could be integrated into MAS studies to improve meat yield (Wang et al. 2019: Li et al. 2021). Although no studies monitoring variations of *CPT1A* and *PRDM6* genes in native Turkish goat breeds are available in the literature, these genes could be utilized in MAS programs to improve genetic gain regarding meat yield in local Anatolian goats. Hence, this study aims i) to investigate genetic variations in *CPT1A* and *PRDM6* in HAI, HNM, and KBK goats via PCR technique and) to assess their usefulness in MAS studies to enhance meat yield.

2. Material and Methods

2.1. Animal sampling and DNA extraction

A total of 204 animals from HAI (n=56), HNM (n=78), and KBK (n=70) were sampled in different regions of Antalya province (Figure 1). This study was approved by Eskisehir Osmangazi University Animal Experiments Local Ethic Committee (Protocol No: HAYDEK-968/2023).

Figure 1. The goat breeds used study (a: HNM, b: KBK, and c: HAI goats)
Şekil 1. Çalışmada kullanılan keçi ırkları (a: HNM, b: KBK, ve c: HAI keçileri)

2.2. PCR amplification and genotyping

The PCR protocols (Table 1) reported by Li et al. (2021) and Manjutha et al. (2023) to investigate 12 bp length of InDel variations were followed to amplify

CPT1A and *PRDM6* genes, respectively, in three Anatolian goats.

A 25 µl PCR reaction was prepared by mixing 50 ng/µl template DNA, 12.50 µl EcoTech 2X Master Mix, 10 pmol/µl of each primer, and 6.50 µl ddH_2O. The thermal cycler was initiated at 95 ºC for 5 mins followed by 35 cycles of 45 sec at 95 ºC for denaturation, 45 sec at 60 ºC for annealing, and 45 sec at 72 ºC for extension. Final elongation was optimized at t 72 ºC for 10 mins. Amplified PCR fragments were visualized via 3.5% agarose gel electrophoresis to genotype each animal. In this step, DNA fragments with 179 and 191 bp lengths were coded as I and D alleles, respectively, for the *CPT1A* gene. Similarly, the PCR bands at 275 and 287 bp length were considered as I and D alleles, respectively, for the *PRDM6* gene.

Table 1. An overview of primers to amplify *CPT1A* and *PRDM6* genes and expected band sizes
Tablo 1. *CPT1A* ve *PRDM6* genlerini çoğaltmak için kullanılan primerler ve beklenen bant büyüklükleri

Gene	Primer sequence (5'-3')	Expected band sizes (bp)	Kaynak
CPT1a	F: ACAGTCACTCCTGCTGCCAATA R: CGCTGCTCTGCGTGTCATTAT	DD = 179 ID = 191, 179 II = 191	Li vd. (2021)
PRDM6	F: GGATACAGGACAGTGTGGGC R: CAACTCACTGAGCAAGGGGT	DD = 275 ID = 275, 287 II = 287	Manjutha vd. (2023)

2.3. Statistical analysis

Popgene V.1.32 (Yeh et al. 1997) program was utilized to calculate allele frequency, genotype frequency, observed heterozygosity (H_O), expected heterozygosity (H_E), and chi-square (χ^2) test to assess deviation from Hary-Weinberg equilibrium (HWE) in three Anatolian goats in terms of *CPT1A* and *PRDM6* genes. Besides, genetic distance values among populations were estimated by the Popgene V.1.32 (Yeh et al. 1997) program and processed into MEGA 11 (Tamura et al. 2021) software to construct the UPGMA dendrogram.

3. Results

In this study, InDel variations of *CPT1A* and *PRDM6* genes were studied in three Anatolian goats by traditional PCR method in which the agarose gel images are given in Figures 2 and 3 while the genetic diversity parameters are summarized in Table 2.

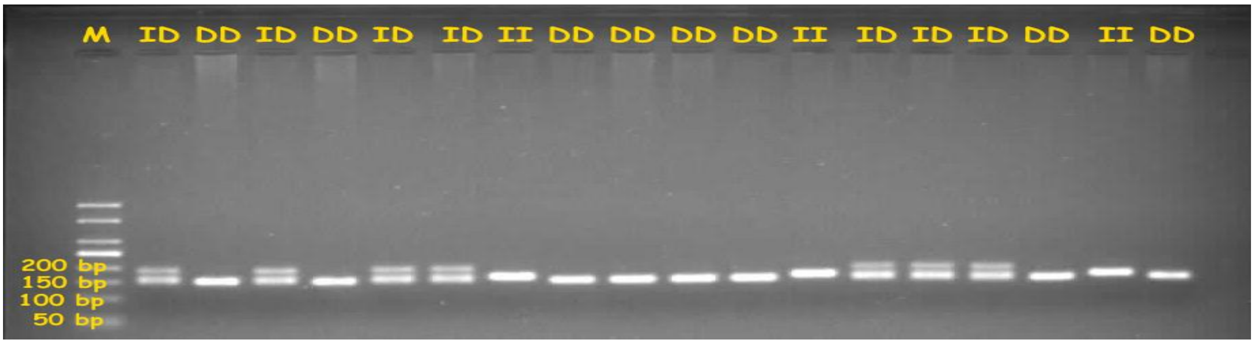

Figure 2. Agarose gel image for 19 randomly chosen animals for the *CPT1A* gene variation
Şekil 2. *CPT1A* geni için rastgele 19 hayvandan seçilen agaroz jel görüntüsü

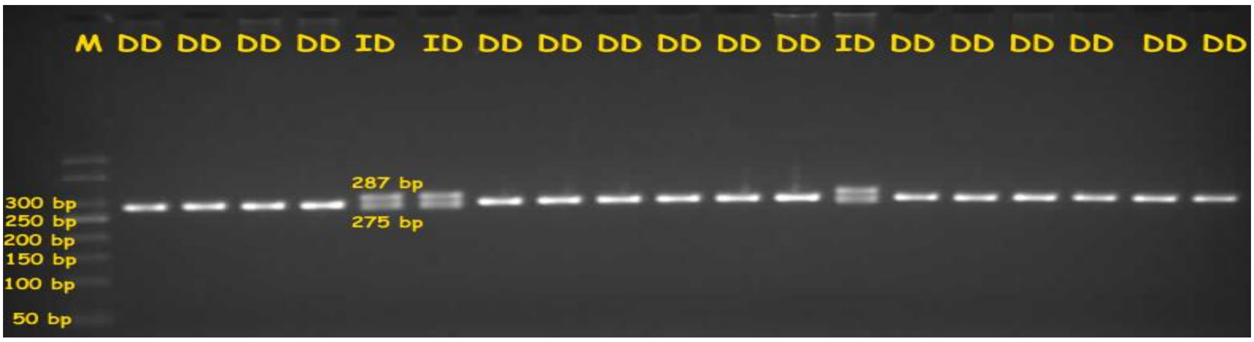

Figure 3. Agarose gel image for 19 randomly chosen animals for the *PRDM6* gene variation
Şekil 3. *PRDM6* geni için rastgele 19 hayvandan seçilen agaroz jel görüntüsü

Table 2. Gene, genotype frequencies and genetic diversity parameters for studied genes
Tablo 2. Çalışılan gen bölgeleri için gen, genotip frekansları ve genetik çeşitlilik parametreleri

			Gene frequency		Genotype frequency			Genetic diversity			HWE
Gene	Population	n	I	D	II	ID	DD	Ho	He	Ne	χ^2
CPT1A	HNM	74	0.13	0.87	0.00	0.26	0.74	0.74	0.77	1.28	1.61
	HAI	56	0.12	0.88	0.00	0.25	0.75	0.75	0.77	1.28	1.14
	KBK	70	0.43	0.57	0.23	0.40	0.37	0.60	0.50	1.96	2.35
PRDM6	HNM	78	0.03	0.97	0.00	0.05	0.95	0.95	0.95	1.05	0.06
	HAI	56	0.03	0.97	0.00	0.07	0.93	0.92	0.92	1.07	0.07
	KBK	70	0.00	1.00	0.00	0.00	1.00	1.00	1.00	1.00	-

$\chi^2_{0.05;1}$: 3.84; a: significant deviation from HWE.

All populations showed polymorphism in terms of the *CPT1A* gene variation in which I allele frequency was estimated at 0.13, 0.12, and 0.43 in HNM, HAI, and KBK goats, respectively (Table 2). D allele frequency turned out to be higher than the I allele across all studied populations. No animals with II genotype were detected in HNM and HAI breeds, while all possible genotypes (II, ID, and DD) were observed in KBK goats (Table 2). 16 animals (0.23) from the KBK population carried the II genotype. ID genotype frequencies ranged from 0.25 in HAI to 0.40 in KBK goats whereas DD genotype frequencies varied between 0.37 (KBK) and 0.75 (HAI) (Table 2). The observed value of heterozygosity was higher than the expected one in only KBK goat. Ne values ranged from 1.28 (HNM and HAI) to 1.96 (KBK) across three Anatolian goats in terms of the *CPT1A* gene variation (Table 2).

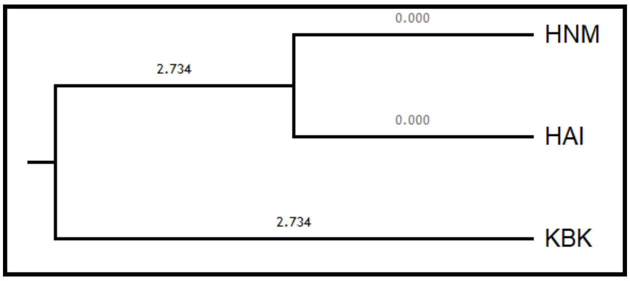

Figure 3. A genetic distance-based UPGMA dendrogram across three Anatolian goat breeds.
Şekil 3. Üç Anadolu keçi ırkı arasında genetik mesafeye dayalı bir UPGMA dendrogramı.

When it comes to the *PRDM6* variations, no polymorphisms were detected for the KBK goat in which all animals turned out to carry the DD genotype. Besides no II genotype was detected across three studied goat populations for the *PRDM6* gene. ID genotype frequencies were estimated at 0.05 and 0.07 in HNM and HAI goats, respectively (Table 2). I and D allele frequency was calculated as 0.03 and 0.97 in HNM and HAI goats, respectively. Similar expected and observed heterozygosity values were detected in all goat populations regarding the *PRDM6* gene. No significant deviation from HWE was detected in HNM and HAI goat in terms of the *PRDM6* variations (Table 2).

The UPGMA dendrogram drawn based on pairwise genetic values among populations is given in Figure 3 in which HNM and HAI clustered together while KBK constituted a separate branch.

4. Discussion

No studies focusing on revealing genetic variations of *CPT1A* and *PRDM6* genes in native Turkish goat breeds were detected in the literature. On the other hand, the number of studies from different countries is still scarce. For example, Li et al. (2021) investigated associations between 12 bp length InDel variations of the *CPT1A* gene and growth traits in Guanzhong dairy (GZ), Hainan black (HNB), Fuqing (FQ) ve Nubian (NB) goats. The authors highlighted those animals with ID and II genotypes showed superior values of growth traits compared to animals carrying DD genotype (Li et al. 2021). Additionally, II genotype frequencies were reported to be 0.21, 0.70, 0.43, and 0.28 for GZ, HNB, FQ, and NB breeds, respectively (Li et al. 2021). ID genotype frequencies were reported to range from 0.29 (HNB) to 0.58 (GZ) in Chinese goats (Li et al. 2021). The current study on three native Turkish goats showed consistent results with the findings reported for II and ID genotype frequencies in four Chinese goat breeds (GZ, HNB, FQ, and NB). Indeed, ID genotype frequency varied between 0.25 (HAI) and 0.40 (KBK) across three Anatolian goats. Although no animals with the II genotype were detected regarding the *CPT1A* genes, the frequency of the ID genotype turned out to be sufficient in HNM and HAI goats. That no animals with the II genotype were detected in the HNM breed was surprising since it is famous for its superior growth traits and live weight. Similarly, the detection of the II genotype in KBK goats is surprising. However, it is noteworthy that possession of a higher body weight could make KBK genetically different from the HAI breed.

A 12 bp length of genetic variations of *PRDM6* genes was previously investigated in Shaanbei White Cashmere and associated with several growth-related parameters (Wang et al. 2020). The authors reported that animals with the DD genotype showed superior values in terms of chest depth and chest width while the II genotype was advantageous for body length. The frequency of II, ID, and DD genotypes in Shaanbei White Cashmere goats was reported as 0.57, 0.37, and 0.06, respectively while the D allele frequency (0.25) was lower than the I allele (0.75) (Wang et al. 2020). Another study carried out by Manjutha et al. (2023) revealed that the D allele frequency (ranging from 0.84 to 0.96) was higher than the I allele frequency (varied between 0.01 and 0.05) in two native Indian goats (Attapady Black and Malabari). The current study showed similar findings reported for native Indian goats (Manjutha et al. 2023) while consistent results were observed compared to Shaanbei White Cashmere goats reared in China (Wang et al. 2020). The reason underlying of these differences may be explained by the origins of the studied goat breeds in which Shaanbei White Cashmere goats were derived from *Capra hircus laniger*. On the other hand, HAI, KBK, HNM, Attapady Black, and Malabari are descendants of hair goats.

Today, KBK goats are considered a variety of the HAI breed (Aslan et al. 2022) but this information has been questioned by several studies. For example, using 20 microsatellite loci, Karsli et al. (2020) highlighted that KBK is genetically closer to HNM rather than HAI in phylogenetic analyses. A similar finding was also reported by Karsli and Demir (2020) based on the variations related to *CSN1S1* and *CMTM2* genes. In contrast, Aslan et al. (2022) highlighted that KBK was genetically closer to HAI than the HNM breed according to polymorphisms of 9 microsatellite loci. Unlike the results of previous studies, this study showed that KBK was genetically from both HAI and HNM breeds. However, the current study focused on two genes which do not represent the whole genome. Therefore, future studies based on next-generation sequencing and array technologies are required for determination of whether KBK is a variety of HAI breed or not.

5. Conclusion

The increasing trend in the human population implies that meat demand will also increase in the future. Therefore, the development of selection strategies based on genes related to meat yield will play a vital role in meeting increasing demand in the agriculture sector. Considering these facts, this study aimed to investigate a 12 bp length of InDel variations of *CPT1A* and *PRDM6* genes in three Anatolian goats via PCR technique. The desired genotype for *CPT1A* (II) was detected only in KBK goats while a sufficient number of animals with ID genotype were detected across all populations. This finding indicates that MAS studies could be carried out in KBK whereas II genotypes may be obtained in HAI and HNM breeds via mating ID genotypes. On the other hand, no animals with the II genotype were detected in KBK goats for the *PRDM6* gene whereas the frequencies of the ID genotype were insufficient across three Anatolian goats. Therefore, the *PRDM6* gene was not useful for MAS studies due to a lack of genetic variations. Since meat yield shows quantitative inheritance, future studies may focus on revealing the other candidate genes which could be subjected to MAS studies in local Anatolian goats.

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Author contributions

YA: Lab Analysis, Data analysis, Data collection, **VA:** Lab Analysis, Data analysis, Data collection **ED:** Methodology, Draft writing, **TK:** Supervision, Methodology, Funding acquisition, Draft writing;

Ethical Permission

This study is reviewed and approved by the Eskişehir Osmangazi University Animal Experiments Local Ethic Committee (Protocol No: HAYDEK-968/2023).

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