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Overview

"Soil Studies (SoilSt)" is the successor to the "Soil Water Journal (Toprak Su Dergisi)" which has been published since 2012. Based on the experience and strengths of its predecessor, SoilSt has been developed to create a truly international forum for the communication of research in soil science. SoilSt is a refereed academic journal has been published free of charge and open accessed by Soil, Fertilizer and Water Resources Central Research Institute. The journal will be published 2 issues (July & December) starting from 2022. It covers research and requirements of all works within the areas of soil.

Aims and Scope

Soil Studies is an international peer reviewed journal that aims to rapidly publish high-quality, novel research of studies on fertility, management, conservation, and remediation, physics, chemistry, biology, genesis, and geography of soils. In addition, the main purpose of Soil Studies is to reveal the influences of environmental and climate changes on agroecosystems and agricultural production. In this context, Soil Studies publishes international studies address these impact factors through interdisciplinary studies. In the journal, articles on hypothesis-based experimental observation of the interactions of all components of agricultural ecosystems, field trials, greenhouse or laboratory-based studies, economic impact assessments, agricultural technologies, and natural resources management will be accepted within the peer-reviewed process. Topics include, but are not limited to:

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- Soil mineralogy and micromorphology
- Soil ecology and agroecosystems
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- Crop water relations, crop yields and water productivity
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- Organic and inorganic fertilization in relation to their impact on yields
- Quality of plants and ecological systems

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Agro-morphological and enzymatic responses of onion to salinity stress under deficit irrigation conditions

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Abstract

In this study, the responses of onion to drought and salinity stress were investigated. In the research, a total of 15 treatments were designed including water stress levels (I_1 : non-stress, I_2 : %25 water stress, I_3 : %50 water stress) and different NaCl concentrations (S_1 : 0.3 dS/m-control, S_2 : 2.5 dS/m, S_3 : 5.0 dS/m, S_4 : 7.5 dS/m, S_5 : 10.0 dS/m). During the experiment, 9.25, 7.66 and 6.07 L/pot irrigation water was applied to I_1 , I_2 and I_3 treatments, respectively. As a result of the study, both drought and salinity stress showed destructive effects on the agro-morphological parameters of onion. Especially salinity levels above 7.5 dS/m triggered significant decreases in yield and yield components in onion. Onion yield decreased by 17.3% in S_2 , 30.3% in S_3 , 37.5% in S_4 and 56.2% in S_5 compared to the control group (S_1) according to salinity levels. When the effects of water stress and salinity stress were evaluated separately, the highest membrane damage (58.4%) was determined in severe water stress treatment (I_3), and the highest H_2O_2 content was determined in severe salinity stress treatment S_5 . On the other hand, antioxidant enzymes such as catalase (CAT), peroxidase (POD) and superoxide dismutase undertook more significant tasks under salinity stress compared to water stress and these enzymes reached maximum values especially under severe salinity stress. Increasing salinity stress decreased the water productivity (WP) of onion, whereas increasing water stress provided significant increases in WP values. According to the obtained data; the use of water with electrical conductivity above 2.5 dS/m in irrigation shows that there will be significant yield decreases in onion. On the other hand, it was clearly understood from the results of this study that 25% to 50% water saving can be achieved by deficit irrigation in onion cultivation in water stressed environments.

Introduction

Water is an indispensable natural resource for agricultural production, and on a global scale, population growth and drought are increasing the demand for clean water resources year by year. Drought is generally defined as a decrease in precipitation and creates serious pressure on water-

dependent sectors such as agriculture ([Türkeş, 2014](#); [Erişmiş, 2023](#)). Especially in arid and semi-arid regions, increasing temperatures and irregular precipitation with climate change are increasing the frequency and severity of drought events, and all of these threaten food security ([Partigöç and Soganci, 2019](#); [Özüpekçe,](#)

2021). Drought adverse effects the development and production of plants in approximately 45% of the world's agricultural areas (Asraf and Foolad, 2007; Yavuz et al., 2021).

On the other hand, salinity as much as drought, is an important environmental stress factor that negatively affects the physiological functions and general development of crops. In particular, the use of saline irrigation water reduces the water uptake of plants from the soil, which has negative effects on photosynthesis and general plant health (Kal et al., 2023; Borromeo et al., 2023; Machado and Serralheiro, 2017). Salt stress can prevent water uptake by creating high osmotic pressure in the root system of plants, which can lead to damage to cellular components (Kahraman, 2024; Yetişsin and Karakaya, 2022). In arid and semi-arid areas, both irrigation water and irrigated areas should be regularly checked in terms of salinity (Smedema and Shiati, 2022; Fernández-Cirelli et al., 2009). For this purpose, it is necessary to know the quality of water resources and determine the salt tolerance threshold values of plants irrigated with these waters (Castellanos et al., 2016). Investigating the salt resistance of plants is important in order to select and grow plants that can produce economically, especially in areas where soil salinity cannot be reduced below a certain level (Kotuby et al., 1997; Zhang and Shi, 2013). In this context, studies are carried out to determine the effects of salinity stress on plants and their morpho-physiological and biochemical response criteria are evaluated (Zhang et al., 2014; Mukherjee et al., 2014; Kostopoulou et al., 2015; Wang et al., 2016; Arora and Bhatla, 2017; Chen et al., 2017; Li et al., 2017; Yavuz et al., 2022).

Salinity and drought stress trigger the defense mechanisms of plants and significantly affect antioxidant enzyme activities. Under stress conditions, the activities of enzymes such as SOD, CAT and POD increase (Jebara et al., 2005; Gondim et al., 2012) and increase the defense of plants against oxidative damage by detoxifying reactive oxygen species (ROS) formed under stress conditions (Corpas et al., 2015; Yasar et al., 2020; Sachdev et al., 2021). These enzymes, whose activities change depending on the severity of stress (Ashraf and Harris 2004), catalyze the splitting of high concentrations of H₂O₂ into O₂ and H₂O, thus stabilizing the physiological functions of the plant. Each enzyme manages different steps of this process; However, CAT is unique in that it does not require any cellular reducing equivalent (Scandalios et al., 1997). Although the experienced stress is first perceived through the roots, its reflections on the leaves play a more important role in understanding the negative effects of stress on plant health (Kirecci, 2018; Yakar and Tuna, 2023). Evaluation of biochemical expressions such as chlorophyll values, carotenoids content and membrane damage are criteria that will increase success in managing abiotic stresses such as

drought and salt stress (Al-Sammarraie et al., 2020; Giordano et al., 2021).

The Konya Plain, located in Central Anatolia of Türkiye and having a semi-arid climate, has limited water resources and therefore the increase in yield and quality in plant production depends on irrigation (Yavuz et al., 2022). Onion (*Allium cepa* L.), which has been widely cultivated throughout the world for thousands of years and is an important agricultural product, has a production of 124 631 tons in 6 775 hectares of land in Türkiye (FAO, 2022). Like many crops, onion is an agricultural product that requires irrigation throughout the vegetation period, especially in arid areas. Adequate irrigation is critical for the growth and productivity of onion (Gerjes et al., 2021). Onion is sensitive to drought because it is a shallow-rooted plant (Ghodke et al., 2020), and Chaudhry et al. (2024) evaluated the effects of different salinity levels in a period without irrigation and reported that photosynthesis decreased significantly, which was reflected in the yield loss. Different researchers also stated that onion is sensitive to drought and salinity stresses (Sönmez et al., 2005; Chaudhry et al., 2024), however, studies examining the responses of onion to conditions where these abiotic stresses are applied together are limited (Ghodke et al., 2020; Khandagale et al., 2024). In this study, the use of low-quality saline waters in onion cultivation in Konya, where drought and salinity problems are experienced, was investigated. In this context, the responses of onion to salinity stress under deficit irrigation conditions were determined agro-morphologically, physiologically and biochemically.

Materials and Methods

Study location and greenhouse climate

The research was carried out in pots in a glass greenhouse at Selcuk University Faculty of Agriculture in April and May 2022 (Latitude: 38°02' N, Longitude: 32° 30' E; Altitude: 1105m). During the research, climate characteristics inside the greenhouse were measured and recorded regularly by means of a meteorological station (Davis Vantage Pro2). During the 43-day vegetation period, the average temperature of the greenhouse varied between 14.3 and 27.5 °C (aver. 20.5 °C) while relative humidity values ranged from 16.4% and 64.9% (average 38.1%) (Figure 1).

Soil properties and trial subjects used in the research

In the experiment, clay-loam textured soil with an organic matter content of 2.72%, pH 7.52, electrical conductivity (EC): 0.91 dS/m and lime content 10.7% was used. The field capacity and permanent wilting point moisture content of the soil were determined as 30.1% and 13.6%, respectively, on a weight percentage

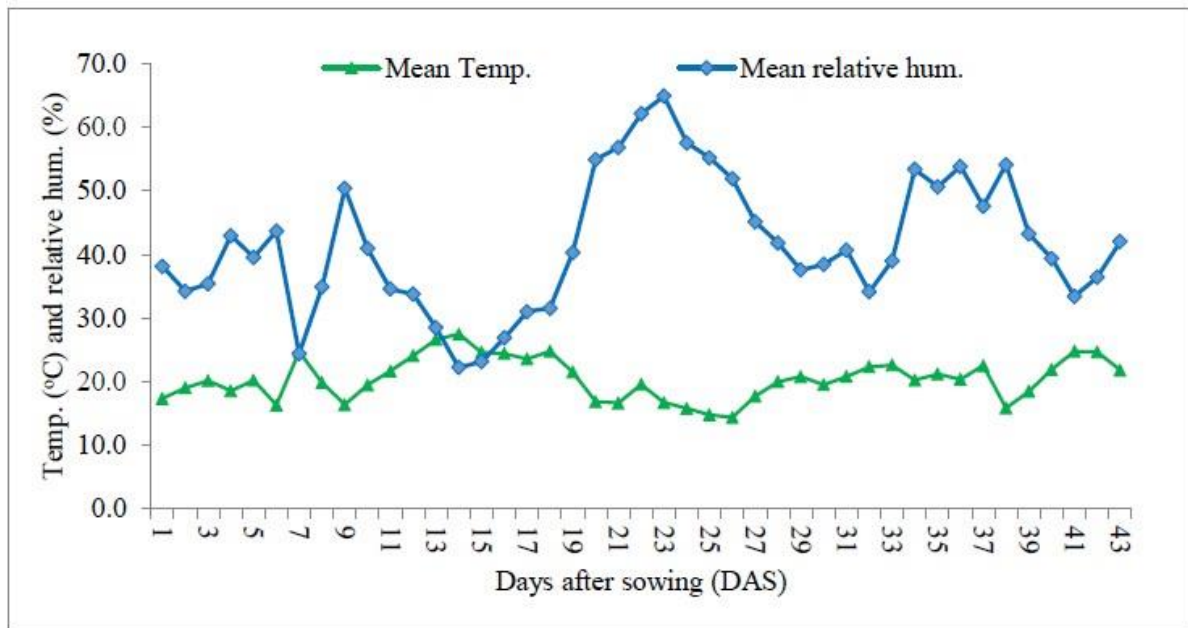


Figure 1. Temperatures and relative humidity values inside the greenhouse during the trial

basis. The soil used in the experiment does not pose a problem for onions in terms of physical and chemical properties. In the research, 12 kg of soil was weighed and placed in each of the plastic pots with a volume of approximately 12 L.

The quality of the mains water used as control is T₂S₁. Irrigation waters with different salt concentrations were created by adding NaCl salt to the tap water (EC: 0.3 dS/m). In the experiment, 15 experimental subjects including 5 different irrigation water salinities (0.3 (control), 2.5, 5.0, 7.5 and 10.0 dS/m) and 3 water levels (I₁: non-stress, I₂: 25% stress, and I₃: 50% stress) were carried out in randomized plots the factorial experimental design with 3 replications. The experimental subjects are shown in Figure 2.

Planting, harvesting and irrigation

In the study, 10 onion seeds were planted in each pot. After sowing, water with different NaCl concentrations was applied to each pot as 1400 mL. Approximately 2 days after seed planting, 1500 mL of irrigation water was applied to all pots to increase soil moisture to field capacity. Then, programmed irrigations were started and moisture losses of control pots (no water deficit and salt stress applied, S₁-I₁) were monitored daily by weighing method. In the experiment, the same amount of irrigation water as the control subject was applied to salinity subjects S₂-I₁, S₃-I₁, S₄-I₁ and S₅-I₁. After programmed irrigations started, 75% and 50% of the water applied to I₁ was given to subjects I₂ and I₃, respectively. In the study, when approximately half of the total usable water of the S₁-I₁ subject decreased, irrigation water with different NaCl

values prepared according to the trial subjects was measured and applied to all pots. The onions reached maturity 43 days after seed planting and were harvested on May 24, 2022.

Determination of actual evapotranspiration (ET_a)

In the study, ET_a for each trial subject was calculated using Equation 1.

$$ET = I - D_p \pm \Delta S \quad (\text{Eq.1})$$

ET_a = Actual evapotranspiration (L),

I = Volume of irrigation water (L),

D_p = Losses through drainage (L),

ΔS = Soil moisture content change (L) dir.

ΔS was calculated from the difference between the available soil moisture at the time of seed sowing and the available soil moisture at the time of harvest.

Water use efficiency (WP) was calculated using yield per plant (g) and ET_a (L) per plant values.

Determination of agro-morphological, physiological and biochemical properties of m onion and evaluation of the results

At harvest, the height, diameter, fresh-dry weight and number of leaves of each plant were determined for each trial. Membrane damage (MD) was determined by the method of [Lutts et al. \(1996\)](#). Carotenoid, chlorophyll values (a, b and total) in onion leaves were determined according to the method

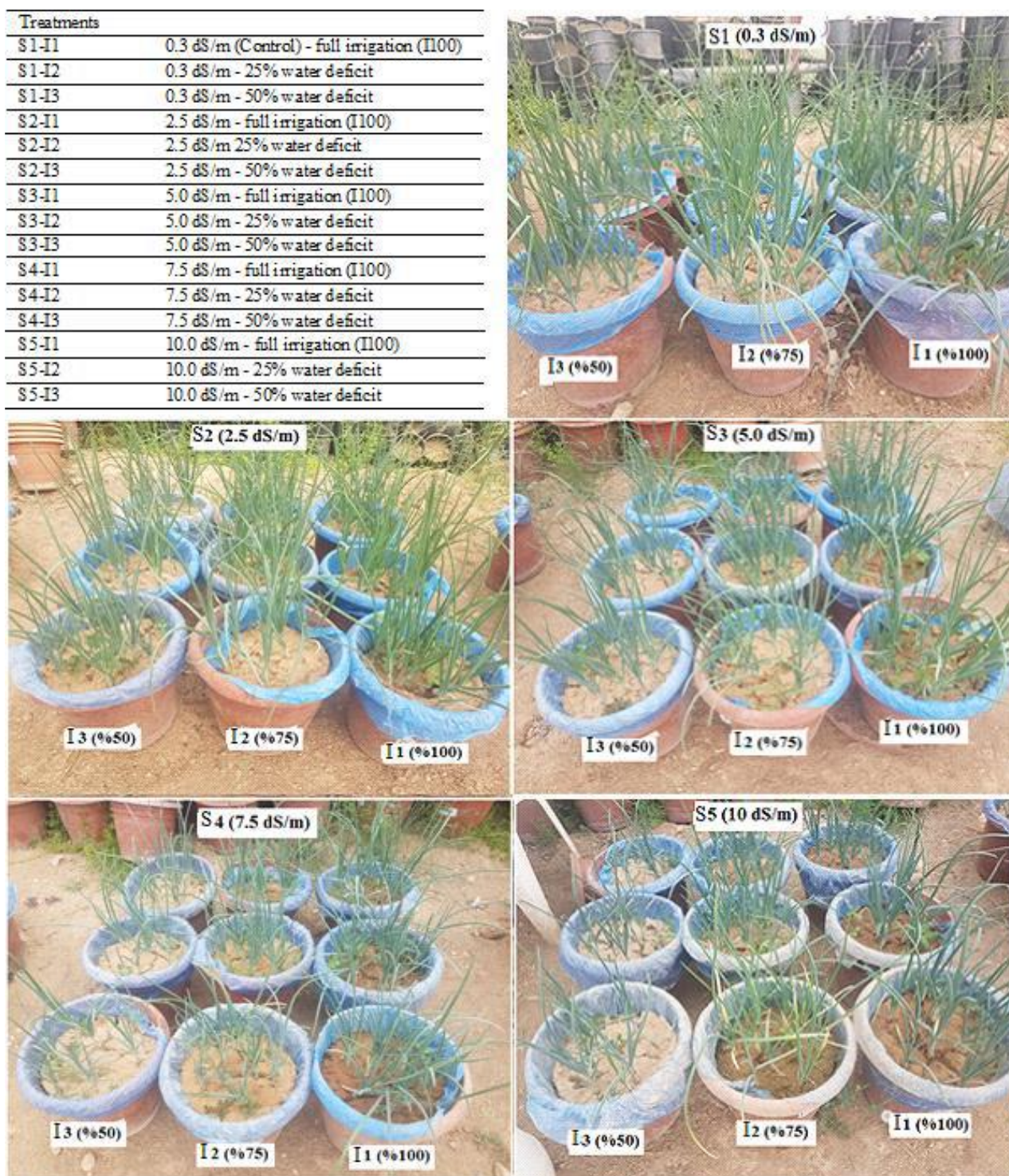


Figure 2. Plant appearances and treatments

reported by [Lichtenthaler \(1987\)](#). Proline contents were determined according to the methods reported by [Bates et al. \(1973\)](#), and protein contents according to the methods reported by [Bradford \(1976\)](#). Antioxidant enzymes were determined; SOD enzyme according to [Beyer Jr and Fridovich \(1987\)](#), POD enzyme according to [Bergmeyer et al. \(1983\)](#) and CAT enzyme according to the method suggested by [Havir](#)

and [McHale \(1987\)](#). CAT, SOD and POD values were determined as enzyme units (EU/g/leaf) per unit weight (g) of a leaf ([Öztürk et al., 2022](#)). The results obtained from the experiment were subjected to ANOVA test in the SPSS-22 program, and statistically significant parameters were grouped based on the Duncan test ($P < 0.05$).

Results

Irrigation water, actual evapotranspiration (ETa) and yield traits

Deficit irrigation started 9 days after seed sowing and ended 38 days later. During the experiment, 9.25, 7.66 and 6.07 L/pot (185, 153.2 and 121.4 mm) irrigation water was applied to I₁, I₂ and I₃ treatments, respectively. In this experiment, firstly plant water consumption in pots was calculated and then ETa values per plant were determined considering the number of plants. The highest ETa was found in S₁I₁ with 869 mL/plant and this was followed by S₂I₁ with 844 mL/plant. The lowest ETa was calculated in S₃I₃ treatment with 505 mL/plant, which was applied 5.0 dS/m salinity and 50% water stress (Table 1). In general, both water stress and salinity stress decreased ETa. The results regarding above-ground fresh and dry weights, under-ground fresh and dry weights, leaf number, stem diameter and plant height values are presented in Table 2. Except for the stem diameter, irrigation water level was found to be statistically non-

significant while the effect of salinity was found to be significant. In general, the data obtained from the control group were in a different Duncan group compared to the data obtained from salinity levels. As the salinity level increased, decreases were observed in agro-morphological parameters. Similar results were obtained from S₂, S₃ and S₄ treatments in some yield components, however, 10 dS/m (S₅) salinity level had destructive effects on onion. Onion yield decreased by 17.3% in S₂, 30.3% in S₃, 37.5% in S₄ and 56.2% in S₅ compared to the control group (S₁) according to salinity levels. The highest value in onion yield according to water levels was obtained in S₁ treatment with an average of 14.1 g/plant, but the differences between the treatments did not show statistical significance. Salinity had a much more negative effect on onion yield than water stress, and it was revealed that significant yield losses would occur when onion was irrigated with saline irrigation water. When both stress conditions were evaluated together, the yield value obtained from the control (I₁S₁) group was approximately three times higher than the yield value obtained from 50% water stress and 10 dS/m salinity stress application (I₃S₅).

Table 1. The total amount of irrigation water applied to the treatments, changes in the soil moisture content at the time of seed sowing and harvest (ΔS), and actual evapotranspiration (ETa).

Treatments	Irrigation water amounts (mL)	ΔS (mL)	ETa per pot (mL/10plants)	ETa per plant (mL/plant)
S ₁ I ₁	9250	565	8685	869
S ₁ I ₂	7663	471	7192	719
S ₁ I ₃	6075	758	5317	532
S ₂ I ₁	9250	815	8435	844
S ₂ I ₂	7663	883	6780	678
S ₂ I ₃	6075	942	5133	513
S ₃ I ₁	9250	916	8334	833
S ₃ I ₂	7663	925	6738	674
S ₃ I ₃	6075	1026	5049	505
S ₄ I ₁	9250	955	8295	830
S ₄ I ₂	7663	960	6703	670
S ₄ I ₃	6075	965	5110	511
S ₅ I ₁	9250	978	8272	827
S ₅ I ₂	7663	965	6698	670
S ₅ I ₃	6075	954	5121	512

Table 2. Changes in yield and yield components of onion under salinity and water stress conditions

Treatments		Aboveground Fresh weight (g/plant)	Aboveground Dry weight (g/plant)	Underground Fresh weight (g/plant)	Underground Dry weight (g/plant)	Leaf number (No/plant)	Plant stem diameter (mm)	Plant height (cm)
<u>Irrigation level</u>								
<u>(I)</u>								
I ₁		14.1	1.29	1.22	0.14	6.13	6.64 ^a	38.8
I ₂		13.2	1.31	1.14	0.12	5.95	6.67 ^a	38.4
I ₃		13.1	1.27	1.08	0.13	5.71	6.14 ^b	37.3
<u>Salinity (S)</u>								
S ₁		18.8 ^a	1.80 ^a	1.88 ^a	0.18 ^a	6.45 ^a	7.75 ^a	43.1 ^a
S ₂		15.5 ^b	1.46 ^b	1.12 ^b	0.13 ^b	6.19 ^{ab}	7.06 ^b	39.4 ^b
S ₃		13.1 ^c	1.22 ^c	1.01 ^b	0.12 ^b	6.07 ^{ab}	6.29 ^c	38.5 ^b
S ₄		11.7 ^c	1.16 ^c	1.06 ^b	0.12 ^b	5.63 ^{bc}	6.18 ^c	38.0 ^b
S ₅		8.2 ^d	0.80 ^d	0.68 ^c	0.09 ^c	5.33 ^c	5.28 ^d	32.0 ^c
<u>I x S</u>								
<u>(Interactions)</u>								
I ₁	S ₁	19.3 ^a	1.80 ^a	2.18 ^a	0.21 ^a	7.22 ^a	8.18 ^{ab}	41.2 ^{abc}
	S ₂	12.5 ^{bc}	1.08 ^{de}	0.74 ^{ef}	0.11 ^{cde}	6.22 ^{abc}	6.59 ^{de}	39.4 ^{bcd}
	S ₃	18.0 ^a	1.68 ^{ab}	1.58 ^{bc}	0.19 ^{ab}	5.89 ^{bc}	7.75 ^{abc}	41.3 ^{abc}
	S ₄	9.9 ^{cde}	0.93 ^{ef}	0.60 ^f	0.08 ^e	5.45 ^{bc}	5.11 ^g	36.6 ^{c-f}
	S ₅	10.8 ^{bcd}	0.97 ^{ef}	1.03 ^{def}	0.11 ^{cde}	5.89 ^{bc}	5.55 ^{efg}	35.5 ^{def}
I ₂	S ₁	19.1 ^a	1.83 ^a	2.12 ^a	0.19 ^{ab}	6.11 ^{bc}	8.31 ^a	45.1 ^a
	S ₂	13.8 ^b	1.44 ^{bc}	0.72 ^{ef}	0.09 ^{de}	5.78 ^{bc}	6.68 ^{cde}	35.2 ^{def}
	S ₃	12.6 ^{bc}	1.25 ^{cde}	0.95 ^{def}	0.11 ^{cde}	6.67 ^{ab}	6.45 ^{de}	39.8 ^{bcd}
	S ₄	13.0 ^{bc}	1.31 ^{cd}	1.40 ^{cd}	0.14 ^{cd}	5.55 ^{bc}	7.16 ^{bcd}	39.8 ^{bcd}
	S ₅	7.5 ^{ef}	0.73 ^f	0.50 ^f	0.08 ^e	5.67 ^{bc}	5.21 ^{fg}	32.3 ^{fg}
I ₃	S ₁	17.9 ^a	1.77 ^a	1.35 ^{cd}	0.15 ^c	6.00 ^{bc}	6.76 ^{cd}	42.9 ^{ab}
	S ₂	20.3 ^a	1.85 ^a	1.90 ^{ab}	0.20 ^a	6.56 ^{abc}	7.92 ^{ab}	43.6 ^{ab}
	S ₃	8.6 ^{def}	0.74 ^f	0.48 ^f	0.07 ^e	5.67 ^{bc}	4.68 ^g	34.3 ^{ef}
	S ₄	12.4 ^{bc}	1.25 ^{cde}	1.18 ^{cde}	0.14 ^{cd}	5.89 ^{bc}	6.28 ^{def}	37.6 ^{cde}
	S ₅	6.4 ^f	0.71 ^f	0.50 ^f	0.08 ^e	4.44 ^d	5.06 ^g	28.2 ^g
<u>Significance</u>								
Irrigation level		ns	ns	ns	ns	ns	*	ns
<u>(I)</u>								
Salinity (S)		**	**	**	**	**	**	**
<u>I x S</u>								
		**	**	**	**	*	**	**

ns: Not significant; *, P<0.05; **, P<0.01. Lowercase letters, uppercase letters, and italics indicate Duncan groups for Salinity (S), irrigation level (I), and I x S interaction, respectively.

Effect of salinity and drought stress on chlorophyll, carotenoid, membrane damage, H₂O₂

The effect of irrigation salinity levels (S), water stress (I) and IxS on chlorophyll values was statistically significant (Table 3). Chlorophyll values increased linearly with increasing water stress. The highest values were observed in I₃ treatments with severe water stress. In general, chlorophyll values increased up to salinity level S₄ (7.5 dS/m) but decreased at salinity

level S₅ (10.0 dS/m). For this reason, it is seen that 7.5 dS/m water salinity is the threshold value for chlorophyll content in onion and that salinity stress higher than this level has negative effects on onion. A decrease of approximately 57% was observed between the applications with the highest chlorophyll-a value (I₃S₃) and with the lowest value (I₂S₅). Similarly, there was a decrease of more than 50% in chlorophyll-b and total chlorophyll values.

Table 3. Changes in chlorophyll, carotenoid, membrane damage and H₂O₂ of onion under salinity and water stress conditions

Treatments		Chl. a (mg/g)	Chl. b (mg/g)	Total chl. (mg/g)	Carotenoids content(mg/g)	Membrane damage (%)	H ₂ O ₂ (μmol/g TA)
<u>Irrigation level (I)</u>							
I ₁		17.5 ^B	5.64 ^C	26.8 ^B	4.51 ^B	54.9 ^B	573
I ₂		16.9 ^B	6.20 ^B	26.9 ^B	4.36 ^B	54.1 ^B	526
I ₃		19.8 ^A	6.79 ^A	31.2 ^A	4.94 ^A	58.4 ^A	503
<u>Salinity (S)</u>							
S ₁		18.3 ^b	6.22 ^a	28.8 ^b	4.59 ^b	51.0 ^c	472
S ₂		18.7 ^{ab}	6.65 ^a	29.9 ^a	4.67 ^{ab}	62.3 ^a	549
S ₃		19.7 ^a	6.51 ^a	30.6 ^a	5.11 ^a	55.4 ^{bc}	494
S ₄		19.5 ^{ab}	6.54 ^a	30.4 ^a	4.96 ^{ab}	55.9 ^b	544
S ₅		14.0 ^c	5.12 ^b	21.8 ^c	3.69 ^c	54.3 ^{bc}	611
<u>I x S (Interactions)</u>							
I ₁	S ₁	21.5 ^{ab}	6.84 ^{b-e}	33.0 ^{bc}	5.61 ^{ab}	43.1 ^f	493
	S ₂	20.6 ^{abc}	6.58 ^{cde}	31.8 ^c	5.31 ^{abc}	67.6 ^{ab}	664
	S ₃	18.7 ^{cde}	5.87 ^{def}	28.6 ^d	4.53 ^{cd}	48.3 ^{ef}	548
	S ₄	16.6 ^{ef}	5.07 ^{fg}	25.0 ^e	4.51 ^{cd}	62.3 ^{bc}	510
	S ₅	10.2 ^g	3.83 ^h	15.7 ^f	2.58 ^e	52.9 ^{de}	649
I ₂	S ₁	16.5 ^{ef}	5.03 ^{fg}	24.9 ^e	4.17 ^d	47.8 ^{ef}	408
	S ₂	19.8 ^{bcd}	8.08 ^a	33.5 ^{bc}	4.80 ^{bcd}	60.1 ^{bcd}	571
	S ₃	17.6 ^{def}	5.77 ^{ef}	27.1 ^d	4.72 ^{bcd}	46.4 ^{ef}	462
	S ₄	20.9 ^{abc}	7.59 ^{abc}	33.5 ^{bc}	5.22 ^{abc}	63.1 ^{bc}	547
	S ₅	9.9 ^g	4.55 ^{gh}	15.8 ^f	2.91 ^e	53.0 ^{de}	642
I ₃	S ₁	17.1 ^{ef}	6.80 ^{b-e}	28.5 ^d	3.99 ^d	62.1 ^{bc}	516
	S ₂	15.8 ^f	5.30 ^{fg}	24.6 ^e	3.90 ^d	59.2 ^{bcd}	412
	S ₃	22.9 ^a	7.90 ^{ab}	36.3 ^a	6.08 ^a	71.4 ^a	472
	S ₄	21.1 ^{abc}	6.98 ^{bcd}	32.8 ^{bc}	5.16 ^{abc}	42.3 ^f	574
	S ₅	21.9 ^{ab}	6.97 ^{bcd}	33.9 ^b	5.58 ^{ab}	56.9 ^{cd}	543
<u>Significance</u>							
Irrigation level (I)		**	**	**	**	*	ns
Salinity (S)		**	**	**	**	**	ns
I x S		**	**	**	**	**	ns

ns: Not significant; *, P<0.05; **, P<0.01. Lowercase letters, uppercase letters, and italics indicate Duncan groups for Salinity (S), irrigation level (I), and I x S interaction, respectively.

It was observed that the content of carotenoid pigments, which play an active role in photosynthesis, increased in onion leaves compared to control group plants at all salinity levels except S₅ (Table 3). On the other hand, among the different water stress levels applied, the highest carotenoid content was in 50% stressed I₃ treatments. In other words, 50% water stress increased the carotenoid content in onion leaves. When both stress conditions were evaluated together, the highest carotenoid content was obtained in S₃I₃ treatment.

Both abiotic stress treatments and the interaction between them showed statistically significant effects

on membrane damage (Table 3). As expected, the lowest membrane damage occurred in the control group (S₁) plants. When the effect of irrigation levels is examined, it is seen that the highest membrane damage (58.4%) occurred in the severe water stress treatment. In the evaluation made in terms of irrigation water salinity according to hydrogen peroxide (H₂O₂) concentrations, the highest H₂O₂ concentration was found in S₅, which is the highest salt stress treatment. Compared to the control group plants, H₂O₂ concentration increased between 4.6% and 29.4% in irrigation water salinity treatments, but these increases did not create a statistical difference.

Table 4. Changes in protein, proline, CAT, POD and SOD of onion under salinity and water stress conditions

Treatments		Protein content($\mu\text{g/g}$)	Proline content($\mu\text{g/g}$)	CAT (EU/g leaf)	POD (EU/g leaf)	SOD (EU/g leaf)
<u>Irrigation level (I)</u>						
I ₁		45.7 ^B	34.9	1396 ^B	1044 ^A	2016 ^A
I ₂		47.1 ^B	33.6	1824 ^A	651 ^C	1717 ^C
I ₃		54.2 ^A	33.8	1414 ^B	794 ^B	1852 ^B
<u>Salinity (S)</u>						
S ₁		46.6 ^{cd}	54.1 ^a	904 ^c	785 ^b	1876 ^{bc}
S ₂		55.1 ^a	32.8 ^b	1386 ^b	588 ^c	1987 ^{ab}
S ₃		49.1 ^{bc}	28.8 ^b	1611 ^b	773 ^b	1796 ^c
S ₄		50.7 ^b	27.8 ^b	1413 ^b	776 ^b	1618 ^d
S ₅		43.7 ^d	26.9 ^b	2408 ^a	1226 ^a	2031 ^a
<u>I x S (Interactions)</u>						
I ₁	S ₁	40.8 ^f	57.2	486 ^{ef}	885 ^{cde}	2503 ^a
	S ₂	66.8 ^b	42.9	2040 ^{bc}	1008 ^{cd}	1661 ^{efg}
	S ₃	43.3 ^f	31.1	1613 ^{bc}	1360 ^b	2420 ^{ab}
	S ₄	26.6 ^g	24.1	1453 ^{bc}	501 ^{ghi}	1557 ^g
	S ₅	51.2 ^{de}	19.0	1386 ^{bc}	1466 ^b	1938 ^{de}
I ₂	S ₁	48.8 ^e	46.1	1040 ^{de}	810 ^{def}	1596 ^{fg}
	S ₂	37.9 ^f	31.6	1840 ^{bcd}	453 ^{ghi}	2162 ^{bcd}
	S ₃	49.9 ^e	36.5	1553 ^{cd}	560 ^{fgh}	1441 ^g
	S ₄	42.5 ^f	37.1	1340 ^{cd}	1104 ^c	1464 ^g
	S ₅	56.7 ^{cd}	16.8	3346 ^a	330 ^h	1923 ^{de}
I ₃	S ₁	50.2 ^e	59.1	1186 ^{de}	661 ^{e-h}	1529 ^g
	S ₂	60.6 ^c	23.8	280 ^f	305 ^h	2136 ^{cd}
	S ₃	54.2 ^{de}	18.7	1666 ^{cd}	400 ^{hi}	1526 ^g
	S ₄	83.1 ^a	22.3	1446 ^{cd}	725 ^{efg}	1835 ^{ef}
	S ₅	23.2 ^g	45.0	2493 ^b	1882 ^a	2233 ^{bc}
<u>Significance</u>						
Irrigation level (I)		**	ns	*	**	**
Salinity (S)		**	*	**	**	**
I x S		**	ns	**	**	**

ns: Not significant; *, P<0.05; **, P<0.01. Lowercase letters, uppercase letters, and italics indicate Duncan groups for Salinity (S), Irrigation level (I), and I x S interaction, respectively.

Results regarding protein, proline and antioxidant enzyme activities

According to salinity levels, the highest protein content was determined in S₂ and the lowest in S₅ (Table 4). The physiological drought that the plant was exposed to due to the increasing salinity intensity had a destructive effect on the protein structure. When considered in terms of different irrigation levels, the protein content reached the highest value in I₃ subjects applied 50% severe drought stress. When the effects of salinity and deficient irrigation were evaluated together, the highest protein content was determined in I₃S₄ treatment (83.1 $\mu\text{g/g}$). It is thought that proteins increased as a function of the defense mechanism

provided to the plant by stress physiology as a result of the increase in the interactive effect of stress. In the experiment, salinity stress had a significant effect on proline contents (Table 4). The highest proline content (54.1 $\mu\text{g/g}$) was found in the control group and up to 50% decreases in proline content were observed with the increase in salinity.

Antioxidant enzymes play an important role under stress conditions and protect plants from the negative effects of ROS. Increased salinity stress caused significant increases in CAT enzyme (Table 4). Compared to the control group plants (904 EU/g), CAT activity increased approximately 2.5 times in S₅ treatment (2408 EU/g). Among water levels, the highest CAT activity was determined in subjects with I₂

irrigation level and was statistically differentiated from the others. When the results related to POD activity were examined, the highest POD activity was found in S₅, which was the highest salt stress application, in terms of irrigation water salinity. When the effects of both stress factors were evaluated together, it was seen that the highest POD activity occurred in I₃S₅ (1882 EU/g), which was exposed to the most severe salt and water stress. When the activity of SOD enzyme was examined, it could be said that it increased with salinity stress, similar to POD, but the increase rate was not as significant as in CAT.

Water-Yield Relationships in Onion

According to salinity levels, the highest WP was calculated in S₁ application with an average of 30.2 g/L, and this application was in a different Duncan group than the others (Table 5). WP decreased by 11.1% in S₂,

31.8% in S₃, 33.0% in S₄ and 56.1% in S₅ compared to S₁. Salinity stress had a devastating effect by reducing water productivity by more than 50%, especially in S₅ application applied with 10 dS/m. The highest WP value calculated among water level applications belonged to 50% water stress application (I₃) with an average of 27.5 g/L. Significant increases in WP occurred as the irrigation water deficit rate increased. The relationships between different irrigation water salinity levels and onion yield are given in Figure 3. By evaluating the effect of salinity levels on yield, linear relationships were found between irrigation water salinity and yield with 99% confidence ($R^2=0.97$) as formulated. Relationships between irrigation water amount and yield are given in Figure 4. As can be seen from the figure, strong polynomial relationships were found between irrigation water amount and yield. Onion yield was negatively affected by salinity stress much more than water stress.

Table 5. Average values (g/L) and importance groups for water use efficiencies (WUE)

Salinity (S)	Irrigation level (I)			Average**	Relative WUE (%)	Difference (%)
	I ₁ (%100)	I ₂ (%75)	I ₃ (%50)			
S ₁ (Control)	24.7 ^{de}	29.6 ^c	36.2 ^b	30.2 ^a	100.0	0.0
S ₂ (2.5 dS/m)	15.6 ^{gh}	21.4 ^{ef}	43.3 ^a	26.8 ^b	88.9	-11.1
S ₃ (5.0 dS/m)	23.5 ^{de}	20.2 ^{ef}	18.0 ^{fg}	20.6 ^c	68.2	-31.8
S ₄ (7.5 dS/m)	12.6 ^h	21.4 ^{ef}	26.5 ^{cd}	20.2 ^c	67.0	-33.0
S ₅ (10.0 dS/m)	14.3 ^{gh}	11.9 ^h	13.5 ^{gh}	13.2 ^d	43.9	-56.1
Average **	18.2 ^C	20.9 ^B	27.5 ^A			
Relative WUE (%)	100.0	115.0	151.4			
Difference (%)	0.0	15.0	51.4			

ns: Not significant; *, $P<0.05$; **, $P<0.01$. Lowercase letters, uppercase letters, and italics indicate Duncan groups for Salinity (S), irrigation level (I), and $I \times S$ interaction, respectively. Significant: $I \times S$: **

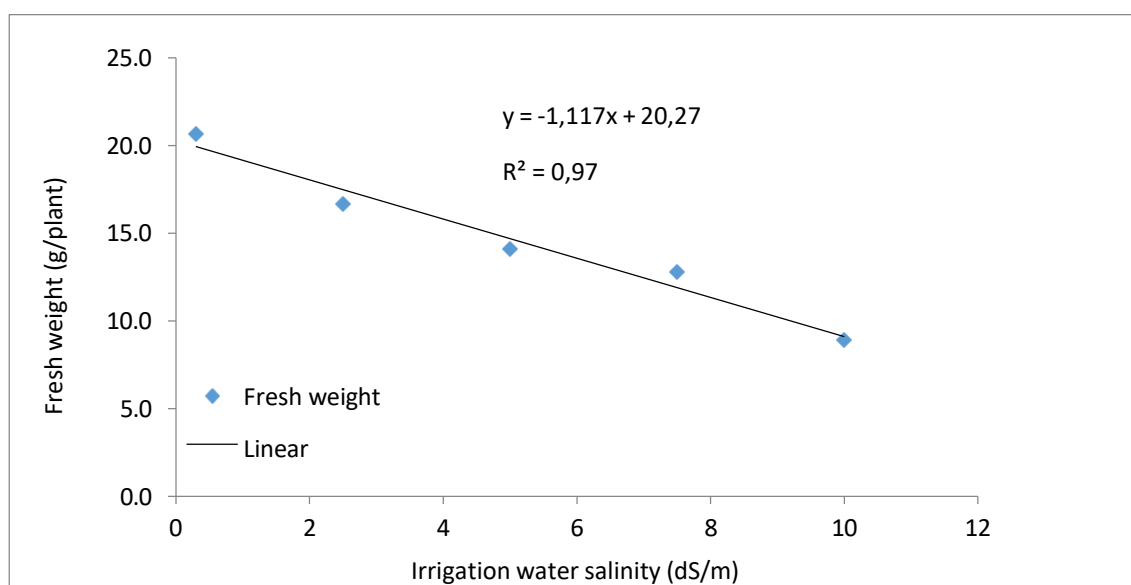


Figure 3. Relationship between irrigation water salinity and yield

Discussion

Drought and salinity, which are the main abiotic stress factors, cause significant losses in plant production (Yavuz et al., 2023). It has been reported that drought stress affects not only yield but also seed quality (El Balla et al., 2013), but salinity stress is a determinant in yield and yield parameters (Al-Harbi et al., 2002; Bekheet et al., 2006; Krasensky and Jonak, 2012). Similar to our findings, some researchers have stated that water stress causes decreases in yield in vegetable species. It has been reported that drought stress causes decreases in aboveground fresh and dry weight in lettuce (Oh et al., 2009), tomato (Çebi et al., 2018), Chinese cabbage (Shawon et al., 2020) and spinach (Yavuz et al., 2022). In the evaluation of water-yield relationships, it is expected that plants have high WP values, that is, the highest yield in return for the water consumed by the plant. Researchers evaluate plants under different stress conditions to determine these threshold values (Munoz-Perea et al., 2007; Blum, 2009; Liu et al., 2016; Yavuz et al., 2023; Seymen et al., 2024). Similarly, in our study, water productivity was determined, and the highest WP (43.3 g/L) was obtained from plants without salt stress under deficit irrigation conditions.

Some researchers think that fluctuations in chlorophyll contents occur as a result of inhibition of chlorophyll biosynthesis and enzymes, and pigments resulting from increased chloroplast membrane permeability, which are broken down to a degree that disrupts metabolic functions (Foyer and Shigeoka, 2011). Basahi et al. (2014) reported that drought stress increased chlorophyll a content in lettuce, but had no effect on chlorophyll b content. In our study, chlorophyll values increased with drought stress but were not statistically different from each other at low

salinity stress levels. Carotenoids are pigments that protect chlorophyll from excessive light intensity in leaves, have antioxidant properties, and are auxiliary to photosynthesis (Smirnoff, 2005; Keyvan, 2010). In parallel with our findings, abiotic stress conditions and severity increased the carotenoid content of spinach (Yavuz et al., 2022) and lettuce (Basahi et al., 2014).

The combined effect of stress factors increased membrane damage, but this increase did not occur regularly and significantly. In a research on salinity stress in blackberry, it was reported that salinity increased membrane damage (Arikan et al., 2018). Similarly, in a study on spinach, it was reported that increasing the salt content of irrigation water increased membrane damage (Yavuz et al., 2022). In this study, among water stress levels, the highest membrane damage was found in the treatment applied with 50% water deficit. It was reported that membrane damage increased significantly in treatments applied with severe water stress (80% water deficit) in spinach (Seymen, 2021). Similarly, it was reported that water stress increased membrane damage in lettuce during the last harvest period (Yavuz et al., 2021).

H₂O₂, an important ROS species, is a compound that causes oxidative stress (Cruz et al., 2013). There was an increase in H₂O₂ content in onion leaves in salinity applications, but this increase was not found to be statistically significant. In the study, it is thought that the reason why H₂O₂ concentration did not increase under water stress conditions is due to the increase in enzyme activities that clean ROS triggered by oxidative stress. These enzymes play important roles in determining the level of effectiveness of each stage of plant responses to stress (Chen et al., 2017). Plants have a magnificent antioxidant defense system consisting of enzymatic and non-enzymatic components that keep ROS under control in order to

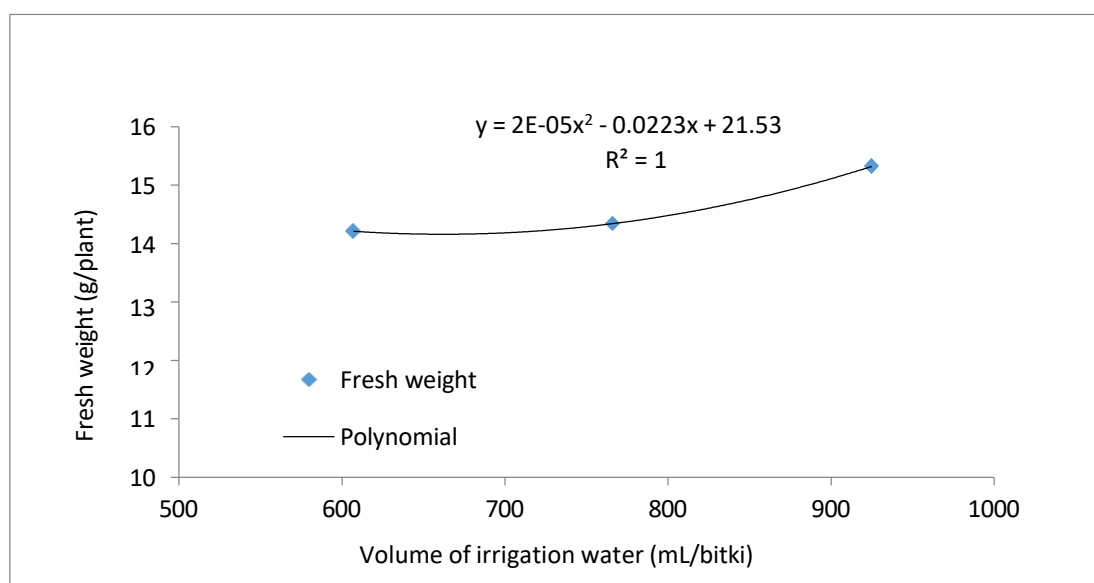


Figure 4. Correlation between irrigation water amount and yield

avoid stress or to continue with minimal damage under oxidative stress (Reddy et al., 2004). The most important ROS-cleaning mechanisms of plants are superoxide dismutase (SOD), peroxidase (POD) and catalase (CAT). The functioning and timing of these enzymes in plant metabolism are vital in maintaining the balance of superoxide radical and H_2O_2 , which cause damage in plant cells under stress conditions. SOD is responsible for the defense system by catalyzing the conversion of superoxide to H_2O_2 and O_2 , POD leads in stress signaling, and CAT is highly effective in scavenging ROS that increase during stress (Mittler, 2002; Mishra and Sharma, 2019; Zulfiqar and Ashraf, 2021). The results obtained from the present study, in line with the literature, show that CAT, POD and SOD fulfill their duties under stress conditions applied to onion.

Conclusions

Onion showed more negative responses to salinity stress than water stress, and it was observed that saline irrigation water would cause significant yield losses in onion. In general, both increasing water salinity and increasing water stress decreased the water consumption of the onion. When the joint effect of irrigation water salinity and irrigation levels ($T \times S$) was evaluated, there were significant yield reductions (about 66.8%) at full and restricted irrigation levels in subjects treated with 10 dS m^{-1} irrigation water salinity. Although chlorophyll values (chlorophyll-a, chlorophyll-b, and total chlorophyll) increased with salinity stress (at T1, T2, T3, and T4 levels), no statistically significant difference was found. Enzymatic (CAT, SOD, POD) and non-enzymatic (protein, proline) responses of onion under stress conditions were evaluated as statistically significant. The metabolism of onion, which reacts to different salt levels, increased enzyme activities significantly to avoid stress. Here, CAT enzyme responded with the highest increase to the signals created by the applied stress conditions. Based on membrane damage values, both stressors were found to be statistically significant. Although the effect of salinity levels fluctuated, the highest membrane damage values were observed at 50% water stress, as expected. Abiotic stress factors such as salinity and drought are important factors affecting plant production in arid and semi-arid areas. Determining the responses of plants to irrigation water salinity under deficit irrigation conditions in such areas is very important for sustainable agricultural production. When the data obtained from the study are evaluated together, it shows that the use of water with an electrical conductivity above 2.5 dS/m in irrigation will cause significant yield decreases in onion. It is obvious that irrigation waters containing very high salt content, especially 7.5 and 10 dS/m , will cause serious yield reductions in onion. On the other hand, it is clearly

understood from the results of this study that farmers who have clean water resources and do not have sufficient water can save irrigation water by applying 25% water stress in green onion cultivation in greenhouse.

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Author Contribution

E.A.: Data curation, Investigation (Master's student). **D.Y.:** Methodology, Investigation, Formal analysis, Data curation, Validation (Thesis advisor). **S.K.:** Writing- Original draft, Data curation, Investigation. **N.Y.:** Writing- Original draft, Investigation. **M.S.C.:** Writing- Original draft.

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Conflict of Interest

No potential conflict of interest was reported by the authors.

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Does azelaic acid priming increase the germination ability of cucumber (*Cucumis sativus* L.) seeds under salt stress?

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Abstract

The aim of this study was to evaluate the effects of Azelaic Acid (AzA) pretreatment on germination of cucumber seeds under salt stress conditions. The experiment was conducted according to a complete randomized experimental design with five different salinity levels (0 mM NaCl; (Control: distilled water), 20 mM, 40 mM, 60 mM and 80 mM NaCl) and 3 different doses of AzA (0 mM, 0.25 mM, 0.5 mM) with 3 replicates. Cucumber variety Beith Alpha was used as plant material. Germination and growth parameters (germination rate, radicle length, wet weight, dry weight, salt tolerance index, root length, shoot length) were determined. According to the results of analysis of variance, the effect of AzA pretreatment on germination rate, salt tolerance index and shoot length of cucumber was found to be insignificant, while it affected radicle length, wet weight, dry weight and root length ($p \leq 0.01$, ≤ 0.05). NaCl levels significantly affected all parameters except germination rate and dry weight. When AzA \times NaCl effects were evaluated together, salt tolerance index was affected at different levels of significance, while there was no significant effect on other parameters. AzA pretreatment at the dose of 0.25 mM was significant for many parameters. Consequently, under salt stress conditions, we can say that AzA, when used at appropriate doses, has a positive effect on germination ability. For various plants grown in areas experiencing salinity problems, priming or foliar application is recommended to determine the role of AzA in stress physiology.

Introduction

Salinity stress is one of the most significant abiotic stress factors affecting agricultural land, and it is reported to negatively impact approximately 932 million hectares of agricultural land (Er et al., 2021). It is estimated that approximately 50% of the world's arable land will be affected by this negative stress factor by 2050 (Bartels and Sunkar, 2005). Salt stress limits plant growth by increasing soil osmotic pressure, specific ion toxicity, and irregularity in nutrient uptake (Läuchli and Epstein, 1990). Increasing salt

concentration in the rhizosphere creates osmotic stress and inhibits water uptake by plants. This condition, referred to as "physiological drought," threatens agricultural sustainability (Yavuz et al., 2022). Salt stress affects plant physiology and biochemistry, causing a significant decrease in crop yield (Ostaci, 2024). In salinity conditions, the osmotic stress observed is caused by an imbalance in the availability of water around plant roots, high concentrations of soluble substances, which significantly limits plant growth and biomass yield (Slabu, 2009). In plants under salt stress, reduced cell extension and stomatal closure

are among the first signs of stress ([Lindberg and Premkumar, 2023](#)). The second stage of salt stress is when the ion concentrations accumulated in plants increase to toxic levels. Accumulated sodium (Na^+) and chloride (Cl^-) cause harmful effects on metabolism, depending on the type of plant and its stage of development ([Munns, 2011](#)). Salt tolerance varies among cucurbits on a scale ranging from sensitive tolerance to moderate tolerance ([Naseer et al., 2022](#)) and it has been reported that the effect of salinity during the germination stage is more harmful than during other stages of product growth ([Irik and Bikmaz, 2024](#)). Melons, which belong to the Cucurbitaceae family, are considered to be moderately tolerant to salinity, but it has been reported that they are significantly affected by the salt factor during germination, shooting, and early seedling stages ([Huang et al., 2012; Pinheiro et al., 2019](#)).

Cucumber (*Cucumis sativus* L.) is one of the most important vegetables worldwide and belongs to the Cucurbitaceae family. Its shallow roots are highly sensitive to salt stress, which causes a decrease in growth and yield ([Wang et al., 2021](#)). Like other salt-sensitive plants, salt stress in cucumbers can cause reduced germination, root growth, and water uptake, and in extreme conditions, plant death ([Liu et al., 2021](#)). The assessment of salinity effects can be an important marker in researching the salt tolerance of cucumber varieties.

Various techniques have been researched for years to improve the adaptability of seeds during germination and increase their tolerance to salinity stress conditions. One of these is the seed priming technique. Seed priming involves exposing seeds to chemicals or stress factors in advance. This process increases the plant's resistance and ability to detect stress signals ([Borges et al., 2014](#)). Many compounds are being studied for their efficiency in increasing the applicability of these and similar anti-stress techniques.

Azelaic acid (AzA), which is commonly used in medicine, cosmetics, and pharmacology, is a compound that has been tested in plants under normal conditions and biotic stress (pathogen) conditions ([Yu et al., 2013; Cecchini et al., 2019](#)). In a study, it has been reported that AzA is already in the plant's biochemical cycle ([Jung et al., 2009](#)). This molecule, known to accumulate in the root system ([Mukhtarova et al., 2011](#)), is a preparatory molecule in the resistance mechanism of plants. In addition, AzA, one of the components that make up the signal transduction pathway, is reported to contribute to the rapid accumulation of salicylic acid (SA) in cases of infection or oxidation. ([Dinler and Cetinkaya, 2024](#)). [Haghighi and Sheibanirad \(2018\)](#); applied 0, 8, 10, and 24 mg l^{-1} azelaic acid exogenously to tomato plants under salinity conditions of 0, 100, 150, and 200 mM, and found that, particularly at 8 mg l^{-1} AzA dose-maintained gas exchange capacity at an optimal level up to 100 mM salinity, induced osmotic balance, and reduced the effects of salinity. In a

different study, the effect of priming corn seeds with AzA under salt stress conditions on germination and early seedling development was investigated, and it was concluded that 1 mM AzA applied to seeds could regulate water uptake, particularly under low and medium salinity conditions, and increase total biomass ([Güleç et al., 2025](#)). Despite these noteworthy results, studies on the effects of AzA against abiotic stress factors are rare.

In this context, this study aimed to evaluate the effects of AzA priming on the germination of cucumber seeds under salinity stress conditions, which is an important abiotic stress factor.

Materials and Methods

In the study, three different AzA doses (0 mM, 0.25 mM, 0.5 mM) and five different salt (NaCl) levels (0; control, 20, 40, 60, and 80 mM) were used for the cucumber variety Beith Alpha. Cucumber seeds were soaked in AzA solutions of different concentrations using 0 mM, 0.25 mM, 0.5 mM doses. The seeds were then dried at +25 °C for 12 hours. After pretreatment, the seeds were sterilized in a 0.5% (v/v) sodium hypochlorite solution for 10 minutes and then rinsed three times with distilled water. Sterilized seeds were placed on double-layered Whatman filter paper in petri dishes (90 mm x 15 mm). Ten seeds were placed in each Petri dish. NaCl solutions prepared in five different concentrations were applied to each Petri dish at a rate of 10 ml. In the study, the number of germinated seeds was counted at the same time every day. Until the end of the study (10th day), NaCl solutions were added to the seeds in the petri dishes every 48 hours (depending on the humidity conditions). At the end of the 24th hour of the experiment, the germination rate (GR) on plants randomly selected from each treatment ([Pour et al., 2021](#)), and radicle length (RL), fresh weight (FW), dry weight (DW), and salt tolerance index (STI) ([Kusvuran et al., 2015](#)) values were determined at the end of the 48th hour. Afterwards, the root lengths (RL) and shoot lengths (SL) of the plants grown until the 10th day were measured at the end of the 10th day. The effect of three different AzA priming doses on cucumber seeds under five different NaCl concentration conditions was analyzed according to a randomized design. Differences between the means of variation sources in terms of germination and seedling growth characteristics were determined using Duncan's multiple comparison test at the 5% significance level.

Results

According to the variance analysis results in Table 1, AzA and NaCl treatments significantly affected the mean germination rate (GR) at 24 hours, while the effect of the AzA X NaCl interaction was insignificant. In

terms of AzA doses, the highest mean germination rate (82.67%) was obtained with the 0.25 mM AzA priming treatment, which was 35.5% higher than the control at 24 hours. This was followed by 0.5 mM AzA priming with 68.67%, and the lowest mean germination rate was obtained from the control group with 53.33% (Table 1). The improving effect of the 0.25 mM AzA dose on the mean germination rates is shown in Figure 1a. When examined in terms of NaCl treatments, a decrease in germination rates occurred due to the increase in salt stress (Figure 1b). The highest mean germination rate was obtained from the control group (0 mM NaCl) at 86.67%, while the lowest value was obtained from the 80 mM NaCl stress level at 37.78%, representing a 56.4% decrease (Table 1).

Although the interaction between AzA and NaCl didn't show a statistically significant effect on the germination rate, significant differences were observed when the treatments were evaluated separately.

Accordingly, the highest germination rate was 100% in the 0.25 mM AzA and 0 mM NaCl interaction, while the lowest mean was 30% in the 0 mM AzA (Control) and 80 mM NaCl interaction (Table 1).

According to measurements taken at the end of 48 hours, the effect of AzA priming and NaCl stress treatments on radicle length (RL) means was significant ($p \leq 0.001$), while the effect of AzA X NaCl interaction was found to be insignificant (Table 1). In terms of AzA priming doses, the mean radicle lengths were found to be 22.48% and 16.99% higher in the 0.25 mM AzA (3.78 mm) and 0.5 mM AzA (3.53 mm) treatments, respectively, compared to the control (2.93 mm) (Table 1; Figure 2a). Increases in stress (NaCl) levels pressured mean radicle lengths (Figure 2b). The highest mean radicle length was 4.29 mm in the control group without salt treatment, while the lowest mean radicle length was 2.67 mm in the 80 mM NaCl treatment (Table 1).

Table 1. Mean values and importance groups for the parameters examined

Treatments	GR ¹ (24 h)	RL (48 h)	FW (48 h)	DW	STI	RL	SL
Priming (P)							
0 mM AzA (Control)	53.33 ^c	2.93 ^b	0.077 ^b	0.026 ^b	64.51	6.37 ^b	5.85
0.25 mM AzA	82.67 ^a	3.78 ^a	0.094 ^a	0.029 ^a	70.98	7.67 ^a	6.58
0.5 mM AzA	68.67 ^b	3.53 ^a	0.081 ^b	0.026 ^b	70.61	5.96 ^b	5.53
NaCl (S)							
0 mM (Control)	86.67 ^a	4.29 ^a	0.095 ^a	0.028	0.00	9.63 ^a	8.39 ^a
20 mM	78.89 ^{ab}	3.80 ^b	0.088 ^b	0.028	93.27 ^a	7.22 ^b	6.30 ^b
40 mM	72.22 ^{bc}	3.40 ^c	0.088 ^b	0.027	92.51 ^b	6.31 ^{bc}	6.92 ^b
60 mM	65.56 ^c	2.90 ^d	0.078 ^c	0.027	82.67 ^c	4.99 ^d	4.53 ^c
80 mM	37.78 ^d	2.67 ^d	0.071 ^d	0.025	75.05 ^d	5.17 ^d	3.79 ^c
P × S							
0 mM × 0 mM	86.67	4.13	0.091	0.026	0.00	8.20	8.10
0 mM × 20 mM	60.00	2.97	0.078	0.028	86.27 ^e	7.47	6.20
0 mM × 40 mM	50.00	2.81	0.076	0.028	83.82 ^g	5.57	7.30
0 mM × 60 mM	40.00	2.48	0.075	0.026	82.97 ^h	5.40	4.17
0 mM × 80 mM	30.00	2.24	0.063	0.021	69.49 ^k	5.20	3.47
0.25 mM × 0 mM	100.00	4.54	0.104	0.031	0.00	9.90	8.30
0.25 mM × 20 mM	93.33	4.29	0.100	0.030	96.14 ^c	7.57	6.37
0.25 mM × 40 mM	90.00	3.74	0.103	0.029	99.79 ^a	8.83	7.73
0.25 mM × 60 mM	83.33	3.28	0.084	0.026	80.81 ⁱ	5.80	5.77
0.25 mM × 80 mM	46.67	3.03	0.081	0.028	78.14 ⁱ	6.23	4.73
0.5 mM × 0 mM	73.33	4.18	0.089	0.026	0.00	10.80	8.77
0.5 mM × 20 mM	83.33	4.16	0.087	0.025	97.39 ^b	6.63	6.33
0.5 mM × 40 mM	76.67	3.63	0.084	0.025	93.91 ^d	4.53	5.73
0.5 mM × 60 mM	73.33	2.94	0.075	0.027	84.22 ^f	3.77	3.67
0.5 mM × 80 mM	36.67	2.73	0.069	0.025	77.52 ^j	4.07	3.17
Sources of Variance							
F value (P)	23.063 ^{**2}	22.660 ^{**}	33.988 ^{**}	5.948 ^{**}	2.826	5.031 ^{**}	2.987
F value (S)	22.563 ^{**}	30.676 ^{**}	20.836 ^{**}	1.676	45.199 ^{**}	13.581 ^{**}	21.251 ^{**}
F value (P × S)	2.111	1.203	1.227	1.359	3.639 ^{**}	1.711	0.871

¹: GR (%): germination rate (24th hours); RL (cm): radicle length (48th hours); FW (g): fresh weight (48th hours); DW (g): dry weight; STI (%): salt tolerance index; RL (cm): root length (10 days); SL (cm): shoot length (10th days).

²: *p < 0.05, ** p < 0.01. Different lowercase letters within a column or row indicate significant differences at the 0.05 level according to Duncan's multiple range test.

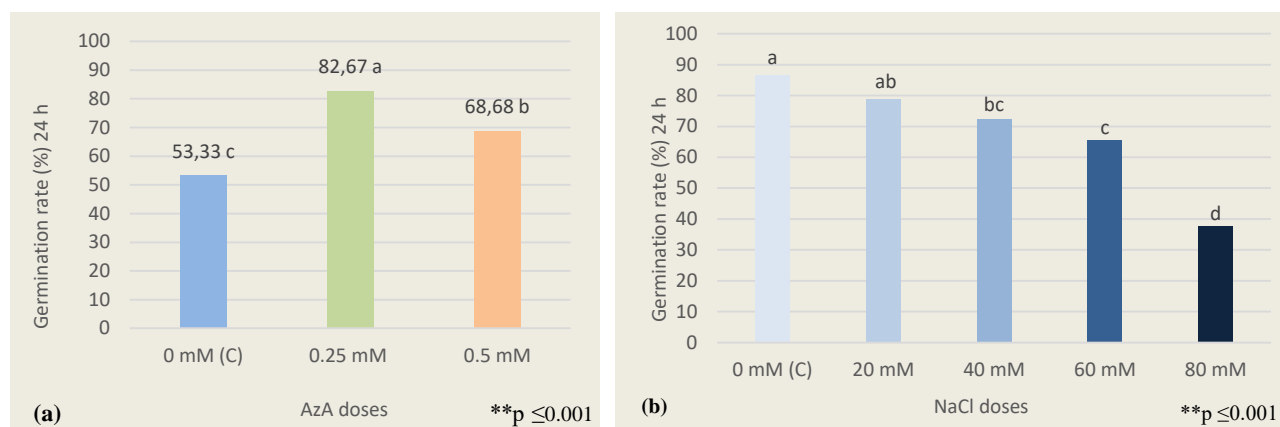


Figure 1. Effect of azelaic acid (a) and salinity stress (b) doses on the mean germination rate (GR) values

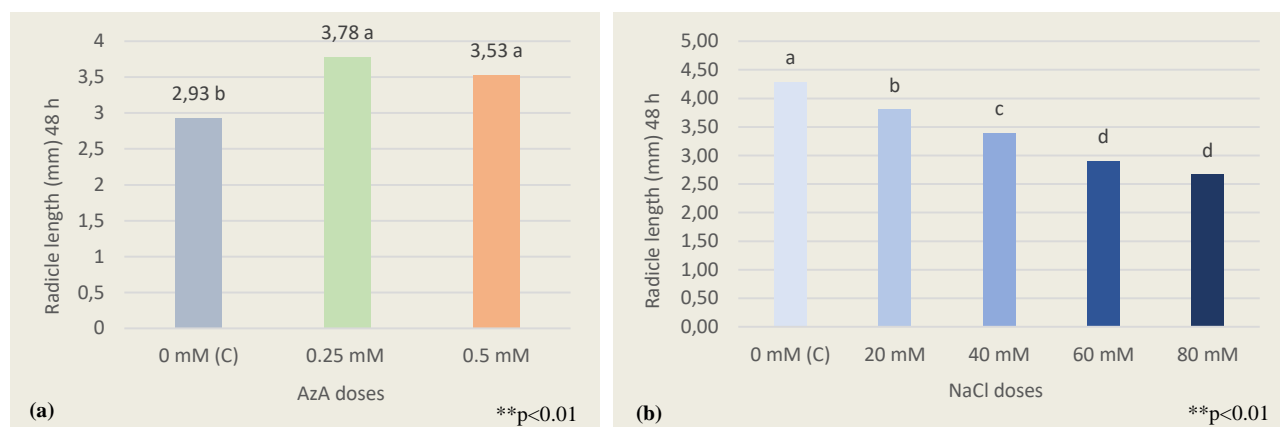


Figure 2. Effect of azelaic acid (a) and salinity stress (b) doses on the mean radicle length (RL) values

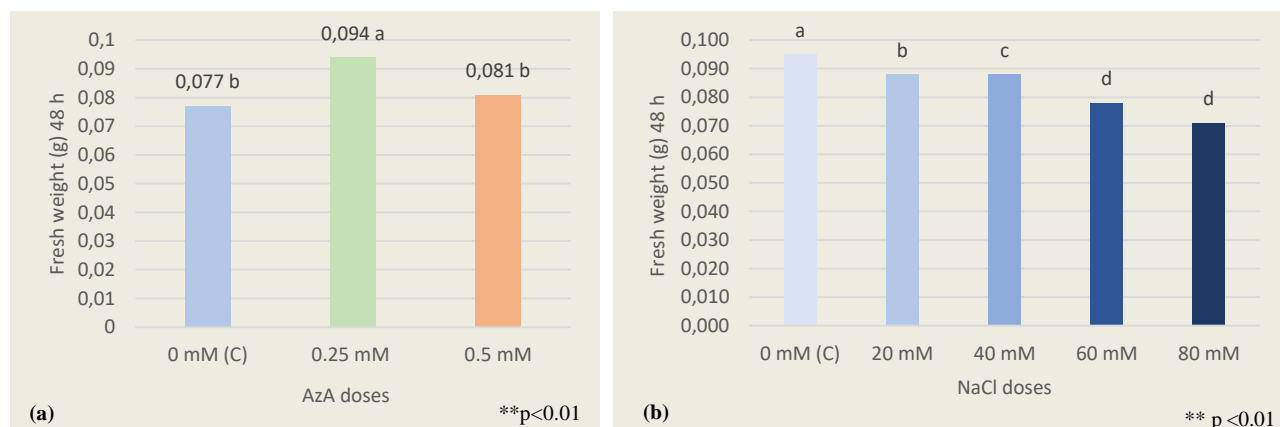


Figure 3. Effect of azelaic acid (a) and salinity stress (b) doses on the mean fresh weight (FW) values

The effect of AzA and NaCl treatments on fresh weights was significant ($p < 0.01$), while the effect of the interaction between AzA and salt stress was insignificant (Table 1). The mean maximum fresh weight at 48 hours was 0.094 g from the 0.25 mM AzA dose, and the mean minimum fresh weight was 0.077 g from the control (0 mM AzA) group (Table 1; Figure 3a). When means were examined in terms of NaCl doses, it was observed that fresh weights decreased with increasing stress (Figure 3b). The highest mean fresh weight was 0.095 g in the control group treated with 0 mM NaCl, while the lowest mean fresh weight was

observed at the 80 mM NaCl dose (0.071 g), which was 25.26% lower (Table 1).

Analysis of variance showed that dry weights (DW) were significantly affected by AzA priming treatments. However, NaCl doses and AzA X NaCl interaction had no significant effect on dry weights. The means for AzA treatments showed that the highest dry weight was obtained from the 0.25 mM AzA dose (0.029 g), which was 10.34% higher than the control, while the lowest dry weights were obtained from the control (0 mM AzA) and 0.5 mM AzA doses, which shared the same significance group (Table 1).

The salt tolerance index (STI) was significantly affected by NaCl doses and AzA X NaCl interaction, while the effect of AzA doses on STI was insignificant (Table 1). Although AzA priming treatments did not show a statistically significant effect, they revealed noticeable differences in salt tolerance index means. The highest STI mean was observed in the 0.25 mM AzA priming treatment, which differed by 9.12% from the control, followed by the 0.5 mM AzA priming treatment, which differed by 8.64% from the control. The lowest STI mean was obtained from the control group without AzA treatment (64.51%) (Figure 4a). Increases in NaCl stress have reduced the mean salt tolerance indices (Figure 4b). The highest tolerance index was determined to be 93.27% at 20 mM salinity stress, while the lowest tolerance index was 75.05% at 80 mM NaCl stress level (Table 1). In the interactions between AzA and salinity stress, the highest salt tolerance index was observed at the 0.25 mM AzA × 40 mM NaCl interaction (99.79%), which provided 16.00% higher tolerance compared to the control group where azelaic acid was not applied and 40 mM salinity stress was applied. The lowest tolerance to salinity stress was observed at 69.49% in the 0 mM AzA and 80 mM NaCl interaction (Table 1; Figure 5).

Root lengths were significantly affected by AzA

and salt stress, while the effect of AzA and NaCl interaction on root lengths was found to be insignificant (Table 1). In terms of azelaic acid, the highest mean root length value was obtained from the 0.25 mM AzA priming treatment, which was 16.9% higher than the control. In contrast, the 0.5 mM AzA treatment shared the same significance group as the control (Figure 6a). When salt stress means were examined, the highest root length value of 9.63 cm was observed in the control group without salt treatment, while the lowest values were detected at 4.99 cm and 5.17 cm, respectively, in the 60 mM and 80 mM NaCl doses, which belonged to the same significance group (Table 1; Figure 6b).

According to the results of the variance analysis for shoot length (SL), no significant interaction was observed between AzA and AzA × NaCl (Table 1). However, the 0.25 mM AzA dose showed an 11.1% difference in shoot length compared to the control group without AzA treatment (Figure 7a). Shoot length means were significantly affected by salt stress conditions (Table 1). The highest mean shoot length was observed in the 0 mM NaCl (control) group, while the lowest mean shoot length was observed at 60 and 80 mM NaCl doses, which were approximately 50% lower than the control (Figure 7b).

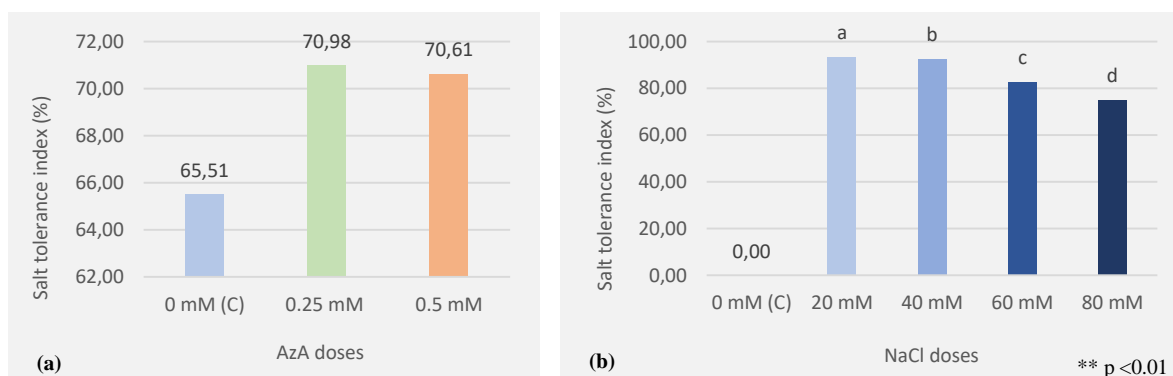


Figure 4. Effect of azelaic acid (a) and salinity stress (b) doses on the mean values of the salt tolerance index (STI)

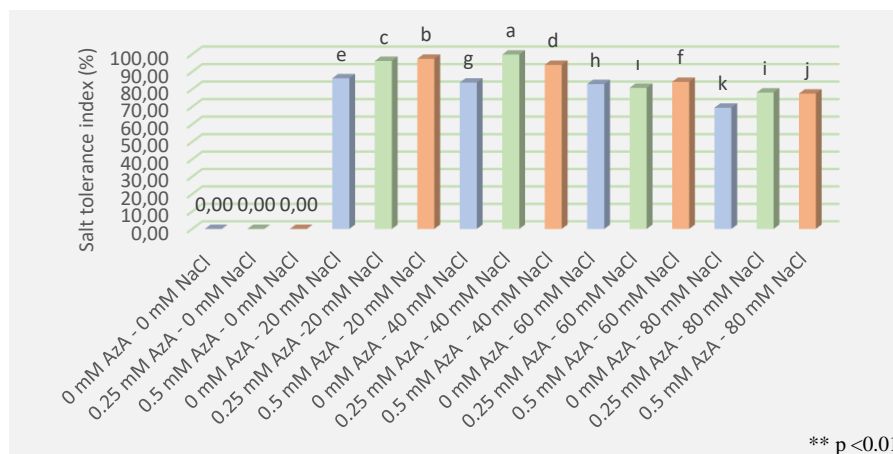


Figure 5. Effect of AzA priming treatments on salt tolerance index (STI) under salinity stress conditions

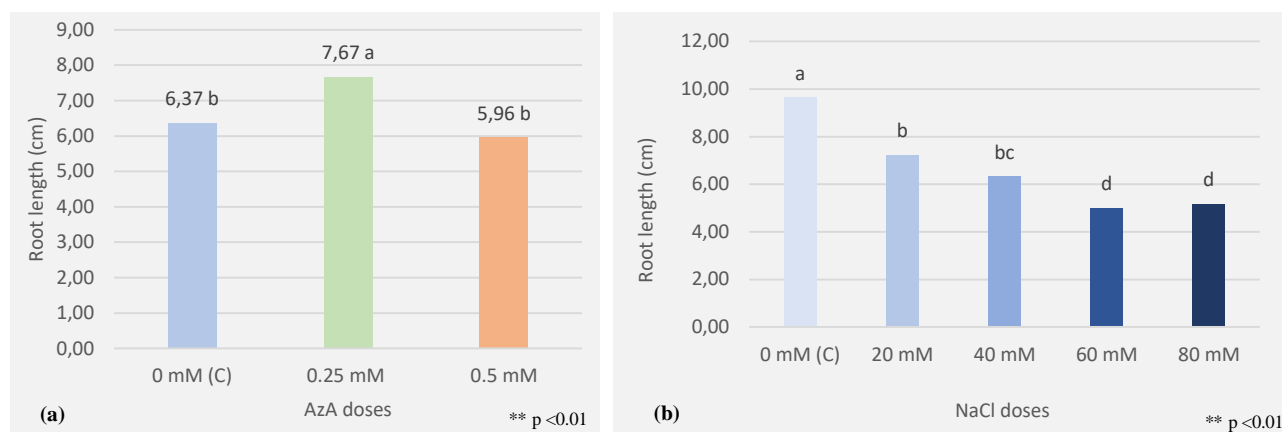


Figure 6. Effect of azelaic acid (a) and salinity stress (b) doses on the mean values of root length (RL)

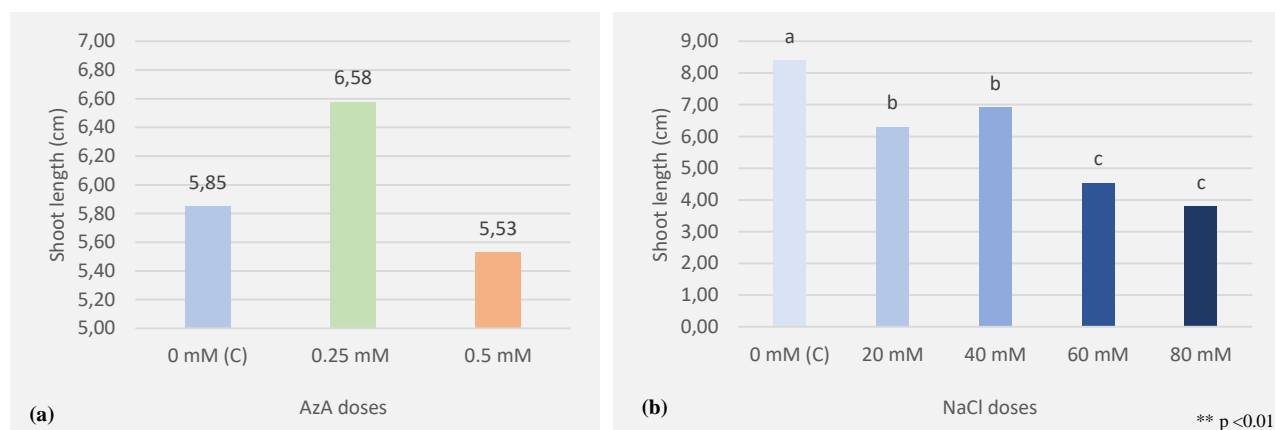


Figure 7. Effect of azelaic acid (a) and salinity stress (b) doses on the mean values of shoot length (SL)

Discussion

Germination rate is a fundamental measure of seed viability. As a general typical characteristic, cucumber seeds can show a germination rate of 95% within 24 hours under optimal conditions. In our study, the highest mean germination rate was obtained in the first 24-hour period following priming with azelaic acid (AzA), particularly with the treatment of 0.25 mM AzA. In a study, it was reported that seed germination increased in cucumber seeds that underwent priming compared to seeds that did not undergo priming (Pandey et al., 2017). Salt stress caused a decrease in germination rates. Our results are consistent with similar studies conducted on cucumbers (Mahdy et al., 2020; Rezvani et al., 2025) and beans (Özkorkmaz et al., 2020).

Radicle lengths were measured at 48 hours after germination. Our results, which are consistent with those of Li et al. (2023) in cucumbers, indicate that increasing NaCl doses suppressed radicle lengths (Figure 8). Under salt-free conditions, the highest radicle lengths were observed in the 0.25 mM and 0.50 mM AzA treatments, respectively (Figure 8-A).

Fresh weights measured at 48 hours showed an inverse trend with increasing salt levels. The lowest fresh weight values were observed under 60 and

80mM stress conditions. Salt stress did not affect dry weights. According to Anwar et al. (2020), priming agents that support cucumber germination and early seedling stages positively affected fresh and dry weights. In our study, the highest fresh and dry weight values were obtained from the 0.25 mM AzA priming group without salt stress.

The salt tolerance index is a useful indicator for identifying salt-tolerant genotypes under high NaCl concentrations (Masuda et al., 2021). The increase in stress levels caused a decrease in salt tolerance index values, similar to the study by Kusvuran et al. (2015). However, the interaction between salt stress and azelaic acid positively affected salt tolerance index values. The highest salt tolerance index was observed under the interaction of 0.25 mM azelaic acid priming and 40 mM salt stress. Accordingly, it can be stated that the treatment of 0.25 mM azelaic acid priming to seeds of the *Beith Alpha* cucumber variety improves salt tolerance under 40 mM salinity stress conditions.

According to measurements taken at the end of the 10th day, decreases in root length values were observed as NaCl doses increased. Salt stress has been reported in various studies to cause setbacks in root development, directly related to the plant's optimal water uptake (Topçu and Özkan, 2017; Yavuz et al., 2023; Güleç et al., 2025). In our study, while the mean

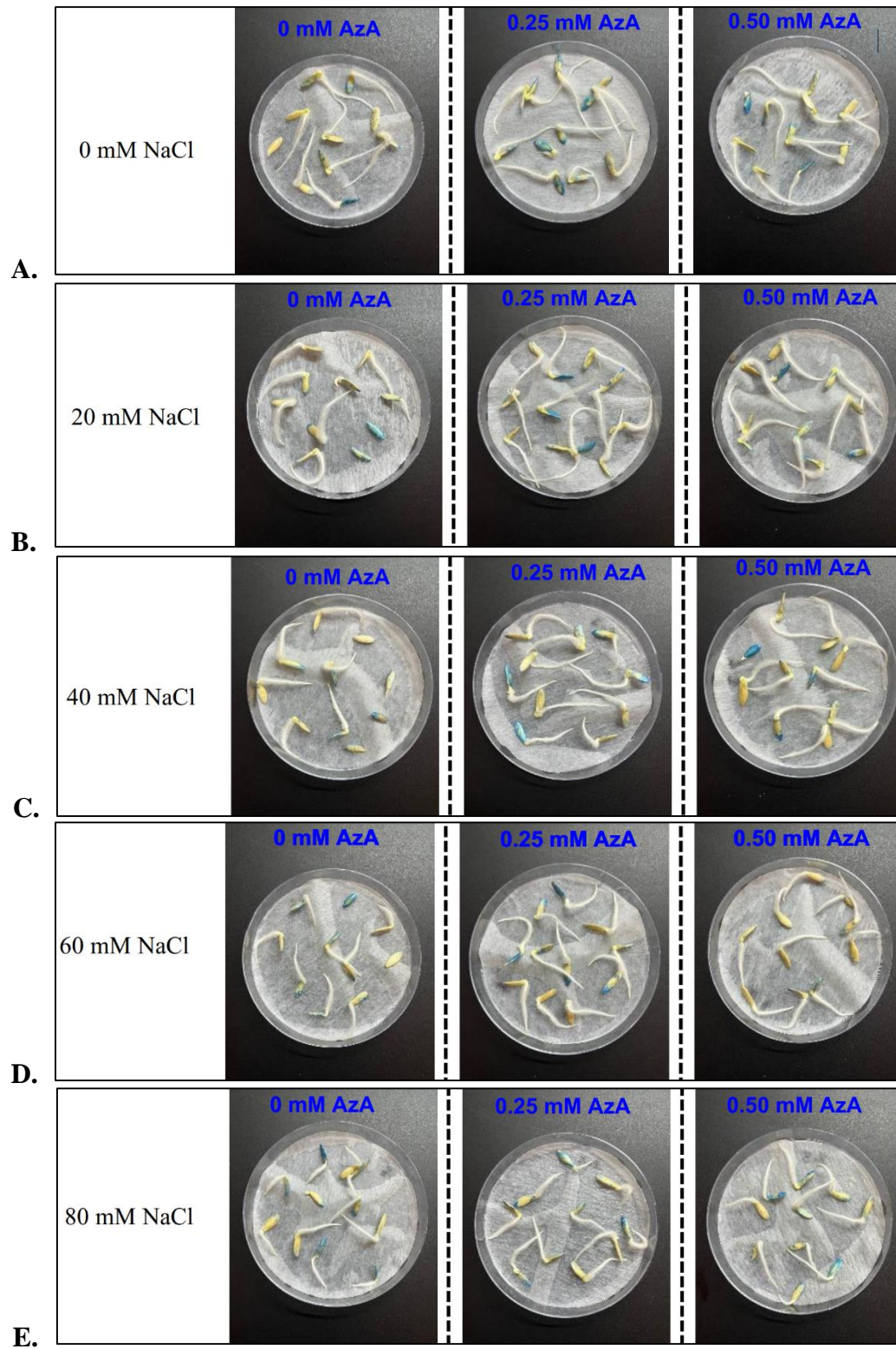


Figure 8. Cucumber seeds exposed to 0, 20, 40, 60, and 80 mM NaCl stress after 0, 0.25, and 0.50 mM AzA priming treatments at 48th hours.

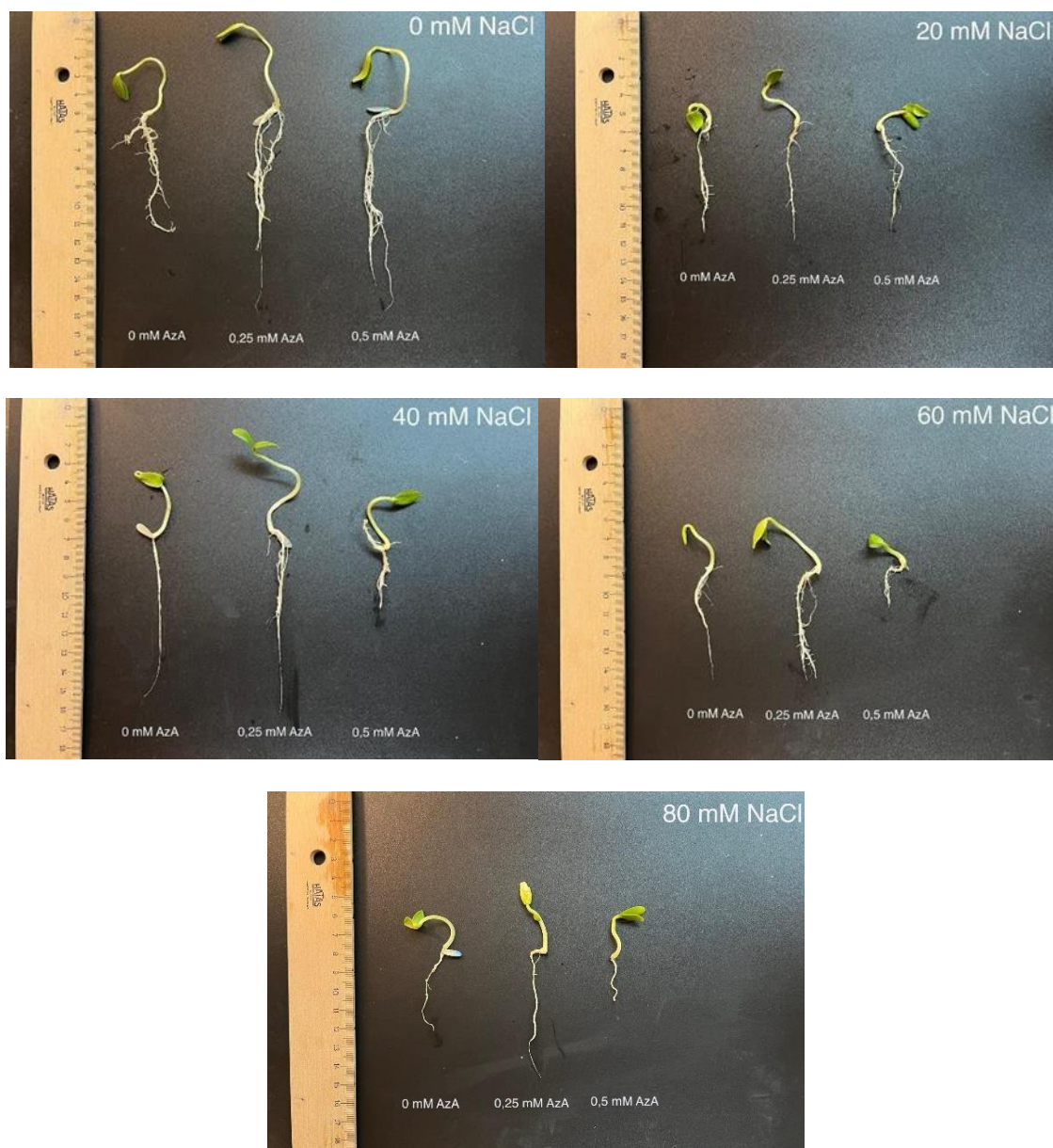


Figure 9. Early seedling stage appearance (10th day) of cucumber seeds exposed to 0, 20, 40, 60, and 80 mM NaCl stress after 0, 0.25, and 0.50 mM AzA priming treatments.

root lengths at 20 and 40 mM salinity levels were affected to the same degree of significance, the lowest mean root lengths were obtained at 60 and 80 mM salinity levels. The root length parameter was significantly affected in terms of azelaic acid means, with the highest mean value observed in the 0.25 mM priming treatment (Figure 9). Different studies have reported that root lengths increase or decrease depending on the dose of the priming agent ([Süheri et al., 2019](#); [Altuner et al., 2020](#)).

At the end of the 10th day, the measured shoot length values were negatively affected by salt stress conditions. The lowest mean shoot lengths were observed in 60 and 80 mM salinity stresses, which share the same significance group. Although azelaic acid means did not separate into any significance group, the highest mean shoot length was again

obtained from the 0.25 mM AzA dose, similar to root length means (Figure 9). [Haghighi and Sheibanirad \(2018\)](#) also reported that azelaic acid improved growth in tomatoes under salinity conditions.

Conclusions

The study investigated the effects of salinity stress on cucumber germination ability and aimed to determine whether AzA, used as a bio agent, could contribute to the tolerance mechanism. When the data obtained from the study were evaluated together, it was determined that salinity stress was statistically effective in all parameters (germination rate, radicle length, fresh weight, salt tolerance index, root length, and shoot length), except for dry weight values. Generally, as the severity of salt stress increases, the

obtained values tend to decrease. It can be said that the effect of salinity levels above 40 mM was particularly devastating for some parameters. AzA applications showed significant effects on germination rate, radicle length, fresh weight, dry weight, and root length. We can say that AzA, which is already active in plant metabolic processes, has positive effects on plant physiology when applied at appropriate doses. In this study, a dose of 0.25 mM AzA supported the germination ability of stressed cucumber seeds.

Based on the results obtained from the study, proper management of salinity is crucial for sustainable agriculture in arid and semi-arid regions. Given the current state of salinity worldwide, strategies such as improving drainage systems, regular leaching practices, selecting salt-tolerant varieties, and using bio agents effective in stress tolerance should be prioritized. Ultimately, it is believed that functionalizing the use of such substances naturally found in plants will contribute to agricultural production.

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Author Contribution

A.G.: Data curation, Investigation, Writing-Original draft. **N.Y.:** Methodology, Investigation, Formal analysis, Data curation, Validation. **D.Y.:** Writing-Original draft, Investigation.

Conflict of Interest

No potential conflict of interest was reported by the authors.

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The residual effects of vermicompost, leonardite and farmyard manure on soil properties

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Farmyard manure
Leonardite
Organic matter
Phosphorus
Nitrogen
Potassium

Abstract

The use of organic amendments on agricultural lands enhances soil nutrient concentrations and properties during mineralization processes. In this study, residual effects of vermicompost (VC), leonardite (L) and farmyard manure (FYM) and their doses (0, 20, 40, and 60 t ha⁻¹ in dry weight) on soil properties were investigated. For this purpose, different doses of three organic amendments were mixed into the soil and after 90 days, tomato seedlings were planted in pots. At the end of the 10-week plant growth period, the experiment was terminated and then soil samples were taken and analyzed to determine the residual effects of organic amendments and their increasing doses. The results showed that residual effects of organic amendments and their doses significantly increased soil organic matter (SOM), total nitrogen (N), available phosphorus (P), exchangeable potassium (K), available iron (Fe) and available zinc (Zn), and especially at 60 t ha⁻¹ application dose. Soil exchangeable calcium (Ca), available manganese (Mn) and available copper (Cu) were not changed by the applications. Soil reaction (pH) was decreased with residual effects of amendments and their doses compared to control, while soil electrical conductivity was increased due to mineralization. Residual effects of VC and FYM were more effective on soil properties than leonardite. Results showed that organic amendments could take advantage of the short-term benefits of nutrients supplied from manure application to improve soil quality and reduce fertilizer input cost.

Introduction

Soils of Türkiye have very low organic matter contents due to semi-arid and arid climatic conditions ([Demirtaş et al., 2013](#)). Soil organic matter influences a wide range of physical, chemical and biological attributes and processes, including the formation and stabilization of soil aggregates, nutrient cycling, water retention, pH buffering and cation exchange capacity ([Marschner, 2002](#)). In order to improve soil properties

and increase the amount of crop production, organic fertilizers should be applied to agricultural production areas. In addition, the increase in natural gas, electricity and oil prices as side effects of the recent economic crisis has led to an increase in the cost of inorganic fertilizers. The increase in chemical fertilizer prices encourages producers to use organic fertilizers ([Nazlı et al., 2016](#)).

Among organic fertilizers, farmyard manure is the best known in Türkiye. However, it is difficult to find farmyard manure in sufficient quantity and maturity at the required time due to the decline in animal husbandry for various reasons and the use of animal waste for energy production. This situation has led to the widespread use of compost, vermicompost, leonardite, liquid and solid humic acids, biochar and poultry manure ([Öktüren Asri et al., 2024](#)). Actually, about 62 million tons of crop plant residues and plant biomass is formed annually in Türkiye, nearly 90% of which is lignocellulosic waste and is rich in organic carbon and other plant nutrients ([Ünlü et al., 2023](#)). Vermicomposting is a very important method for the utilization of these wastes. Vermicompost is produced by the composting of various organic materials (stalk, straw, fruit and vegetable wastes, sawdust etc.) by specific species of earthworms and their associated microbiota during the decomposition of organic matter ([Demir, 2024](#)). Vermicompost was reported to improve soil fertility through improving soil organic matter content ([Öktüren Asri et al., 2024](#)), macro and micronutrient elements ([Zhang et al., 2020](#); [Demir, 2024](#)), porosity and structure ([Demir, 2024](#)).

Leonardite, one of the organic materials, is a product of atmospheric oxidation of lignite. It is rich in organic matter (50-75%) and humic acid (HA) content (30%-80%). HA contains active functional groups (e.g. quinonyl, carboxyls and phenolic hydroxyl), thereby, it has the capability to engage with metal ions, oxides, hydroxides and minerals ([Abdullah et al., 2024](#)). Due to humic acid (HA) content, positive effects of leonardite on aggregate stability, water holding capacity, organic matter content, nutrient elements concentration and enzyme activity of soil were reported by [Sesveren and Taş \(2022\)](#), [Alagöz et al. \(2006\)](#), and [Wang et al. \(2013\)](#).

The application of organic amendments to soil is known to provide macro and microelements to soil through mineralization. However, information on the time and speed of mineralization process is scarce. There are studies on the effects of organic matter added to the soil before crop cultivation on soil properties, but studies on the effects of organic materials after crop cultivation are very limited. Thus, the objective of this study was to evaluate the residual effects of different doses of farmyard manure, leonardite and vermicompost on soil properties after tomato harvest.

Materials and Methods

The study was carried out in a pot experiment in the greenhouse of Akdeniz University Faculty of Agriculture, Antalya-Türkiye. The pots were filled with a Alfisol soil. Some properties of soil and organic amendments were given in Table 1 and Table 2.

Table 1. Some properties of soil used in the study

Measured Parameters	Values
Total N, %	0.10
Available P, mg kg ⁻¹	39
Extractable K, mg kg ⁻¹	120
Extractable Ca, mg kg ⁻¹	6600
Extractable Mg, mg kg ⁻¹	658
DTPA- Extractable Fe, mg kg ⁻¹	6.7
DTPA- Extractable Cu, mg kg ⁻¹	2.2
DTPA- Extractable Mn, mg kg ⁻¹	11.7
DTPA- Extractable Zn, mg kg ⁻¹	0.90
pH (1:2.5 distilled water)	7.40
EC (1:2.5 distilled water), dS m ⁻¹	0.274
Lime (%)	26.4
Organic Matter (%)	1.25
Bulk density (g cm ⁻³)	1.18
Field capacity (%)	34.0
Wilting point (%)	22.0
Texture	Clay Loam (CL)

In the experiment performed in 3 L pots based on the randomized block experimental design as 4 replications. Leonardite (L), vermicompost (VC) and farmyard manure (FYM) were mixed in four different (0, 20, 40, and 60 tons ha⁻¹ in dry weight) doses with soil. After 90th day, tomato seedlings were planted to pots. No inorganic fertilizer was applied to the organic fertilizer plots throughout the life of the plant. At the end of the 10-week growing period, the experiment was ended and then soil samples were taken and analyzed for determining residual effects of organic amendments and their increasing doses.

Analytical methods

Soil analysis methods: Soil samples were taken and analyzed after air drying and passing through a 2 mm sieve. The pH and EC were measured in a 1:2.5 (w/v) soil to water ratio ([Jackson, 1967](#)). Total carbonates were determined using the Scheibler calcimeter ([Kacar, 2016](#)). Soil texture was determined based on the hydrometer method ([Bouyoucos, 1955](#)) and the organic matter was determined based on the modified Walkey-Black method ([Black, 1965](#)). Total nitrogen was determined based on the modified Kjeldahl method ([Kacar, 2016](#)). Available phosphorus was extracted with 0.5 M NaHCO₃ and determined based on the molybdate colorimetric method (Shimadzu UV 1800) ([Olsen and Sommers, 1982](#)). Extractable K, Ca, and Mg was extracted with 1 N ammonium acetate (NH₄OAc) and determined via ICP-OES (PerkinElmer Avio 2000) ([Kacar, 2016](#)). Available Fe, Zn, Mn, and Cu in the soil was extracted with Diethylene Triamine Pentaacetic Acid (DTPA) ([Lindsay and Norwell, 1978](#)), and determined with the ICP-OES device (PerkinElmer Avio 2000).

Table 2. Some properties of organic amendments used in the study

Measured Parameters	Leonardite (L)	Vermicompost (VC)	Farmyard Manure (FYM)
pH (1:5 distilled water)	6.20	7.46	6.50
EC (1:5 distilled water), dS m ⁻¹	5.20	6.53	6.70
Organic Matter, %	49.0	52	55.0
Organic C, %	27.0	28	27.5
Organic C/total N	9.64	10.76	9.48
Total N, %	2.80	2.60	2.90
Total P, mg kg ⁻¹	2500	3701	1300
Total K, mg kg ⁻¹	4100	5905	18800
Total Ca, mg kg ⁻¹	13270	5378	82300
Total Mg, mg kg ⁻¹	701	1193	2000
Total Fe, mg kg ⁻¹	69	34.8	6897
Total Zn, mg kg ⁻¹	1.41	7.57	114
Total Mn, mg kg ⁻¹	1.22	3.53	259
Total Cu, mg kg ⁻¹	0.19	2.20	31.9

Organic amendments analysis methods: The organic matter (OM) contents of leonardite, vermicompost and farmyard manure were determined in a combustion oven (550 °C) (Black, 1965). EC and pH were determined by using a portable EC and pH meter in 1:5 (w/v) organic material to water ratio (Jackson, 1967). Total N content was determined by Kjeldahl method. Total P, K, Ca, Mg, Fe, Zn, Mn, and Cu in the same solution were determined in the wet-digested samples via ICP-OES (Kacar and Inal, 2010).

Statistical methods

The statistical analysis was made according to the principles set by Yurtsever (1984). All data were analyzed using the JMP Statistical package program developed by SAS (SAS Institute, Cary, North Carolina, USA). Means were compared by analysis of variance (ANOVA) and the LSD test at the $p \leq .05$ significance level.

Results and Discussion

Residual effects of organic amendment applications, doses and interaction between organic amendments and their doses on soil reaction (pH) were found to be statistically significant ($p < 0.001$) (Table 3). Initial soil pH (1:2, 5) was 7.40, after the applications soil pH was changed between 7.31 and 7.46 (Figure 1). Organic amendments and their increasing doses caused to decrease soil pH compared to control. Ateş and Namli (2021) disclosed that decomposition process of organic materials changed to soil pH due to accelerating the release of organic acids and CO₂. Zhao et al. (2017) explained that the application of vermicompost resulted in decreased soil pH because the high ability of vermicompost to promote crop

system development and absorb more mineral ions while simultaneously producing hydrogen ions. Soil pH is an important soil property that can limit the effects of plant nutrition practices on crop yield and quality. Soils of Türkiye have high lime and pH level (92.64% of the Turkish soils reaction higher than 6.5) due to parent material and climatic conditions (Kacar and Inal, 2010). Thus, organic amendment applications and their effects of soil pH is important for Türkiye's soils.

Residual effects of organic amendments, doses and interaction between organic amendments and their doses on soil electrical conductivity (EC) were found to be statistically significant ($p < 0.001$) (Table 3). Initial soil EC was 0.27 dS m⁻¹. After applications, the lowest EC value (0.20 dS m⁻¹) was obtained from 0 t ha⁻¹ doses of control pots and the highest value (0.70 dS m⁻¹) was determined by 60 t ha⁻¹ of leonardite application (Figure 1). The residual effects of organic amendment doses increased to soil EC by 53.5%-64.9% (Table 2). Öktüren Asri et al. (2024) reported that soil EC increase with farmyard manure, vermicompost, spent mushroom compost and chicken manure by 12%-33%. Obour et al. (2017) explained that the relatively greater EC values with higher manure application rates may be attributed to higher residual K, Ca and inorganic N concentrations resulting from greater rates of manure application.

One important ecological service that organic matter management offers is the storage of carbon in soil organic matter. By raising the organic carbon content of soil and encouraging the synthesis of stable organic carbon molecules, organic sources aid in the sequestration of carbon (Boostani et al., 2020). Residual effects of organic amendment applications ($p < 0.01$) and their doses on soil organic matter (SOM) were found to be statistically significant ($p < 0.001$) (Table 3). The residual effects of organic amendment

doses increased to soil organic matter (SOM) by 19%-45% (Figure 1) compared to control. 60 t ha⁻¹ (1.61%) application dose was the most prominent application dose. Although the organic matter contents of organic materials were close (Table 2), leonardite (1.28%) and vermicompost (1.24%) were particularly effective and created the highest SOM, followed by farmyard manure (1.13%) (OA means in Figure 1). Öktüren Asri et al. (2024) reported that SOM increased by 13%–16% with organic amendments (vermicompost, farmyard manure, chicken manure, spent mushroom compost), these positive effective continued in the following year

and resulted in increases of 14%-24%, compared with the control. Dinakaran et al. (2024) found that soil organic matter was more accumulated with vermicompost compared to farmyard manure, and SOM increased by 23%, 19% and 25% ratio compared to control with 15, 30, and 60 t ha⁻¹ vermicompost doses.

Residual effects of organic amendments, application level and their interaction on soil total nitrogen (TN) concentration were found to be statistically significant ($p < 0.001$) (Table 3). The lowest total N (0.106%) was obtained from all of control pots

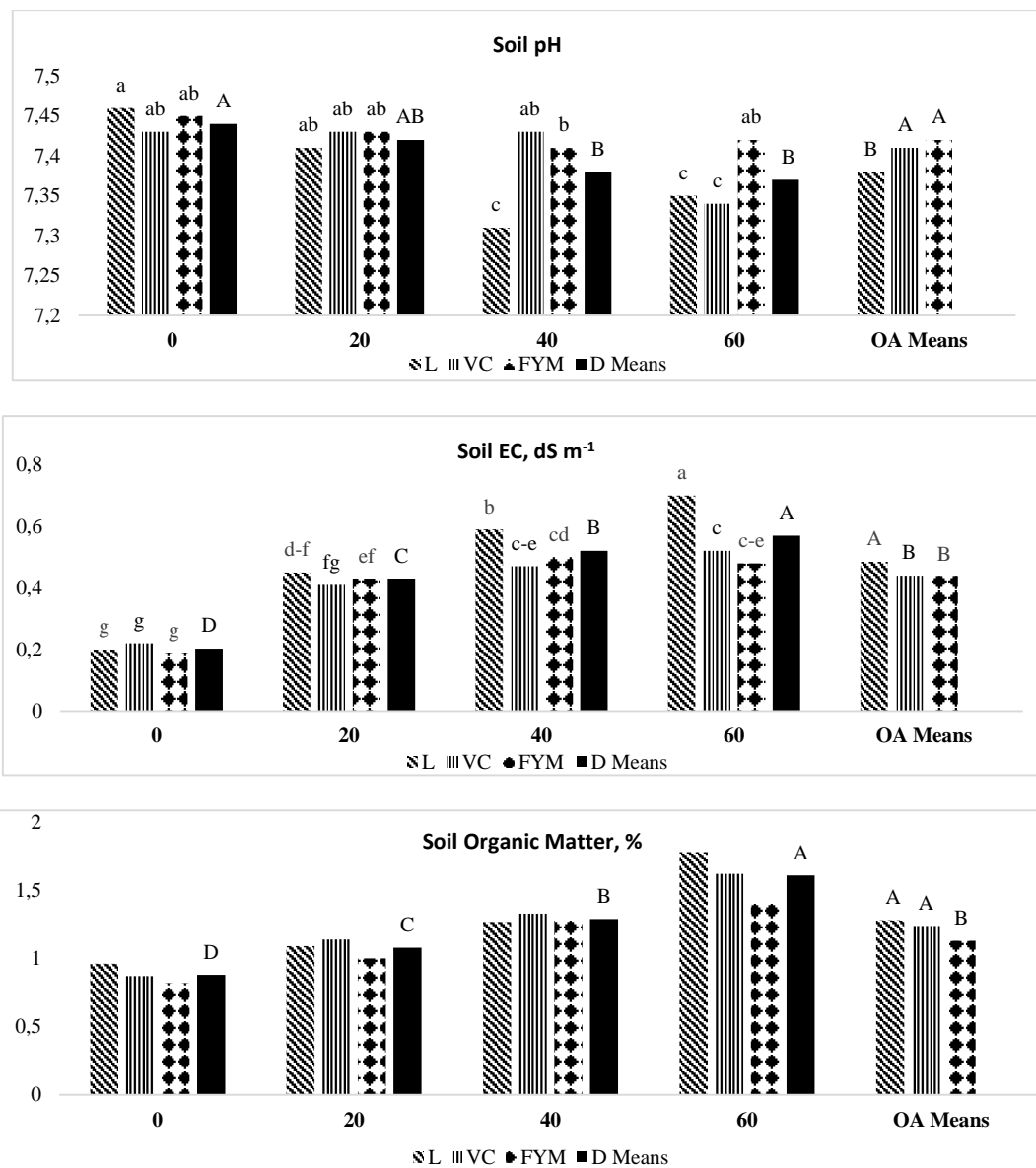


Figure 1. Residual effects of organic amendments on soil pH, EC and organic matter content

Lowercase letters indicate a significant difference in the interaction between organic amendments and their doses. Uppercase letters indicate a significant difference between the organic amendments (OA Means). Uppercase letters indicate a significant difference between the doses (D Means)

Table 3. Results of analysis of variance (*p* values) for soil properties treated with organic amendments

Soil Parameters	Organic Amendments (OA)	Dose of Organic Amendments (D)	OA*D
pH	0.0033**	<,0001***	<,0001***
EC	<,0001***	<,0001***	<,0001***
SOM	0.0292**	<,0001***	0.360ns
Total N	<,0001***	<,0001***	0.0004***
Available P	<,0001***	<,0001***	<,0001***
Exchangeable K	<,0001***	<,0001***	<,0001***
Exchangeable Ca	0.0037ns	0.3299ns	0.6825ns
Exchangeable Mg	0.3974ns	<,0001***	0.1634ns
Available Fe	<,0001***	<,0001***	<,0001***
Available Zn	<,0001***	<,0001***	<,0001***
Available Mn	0.9071ns	0.8214ns	0.6544ns
Available Cu	0.4102ns	0.5257ns	0.0584ns

* Significant at the $\alpha = 0.05$ probability level.

** Significant at the $\alpha = 0.01$ probability level.

*** Significant at the $\alpha = 0.001$ probability level.

ns = non-significant.

in 0 t ha⁻¹ dose, while the highest values was determined in 60 t ha⁻¹ VC (0.140%) and FYM (0.144%) (Figure 2). The organic amendment doses increased the soil TN by 10%-22%, compared with the control. High organic amendment application rates are expected to increase soil N mineralization and actively enhance organic N mineralization (i.e. protein and amino acids), which might be due to high organic N addition ([Ma et al., 2018](#)). Mineralization of organic N rates is influenced by a number of factors, especially the quantity and the microbial susceptibility of existing carbonaceous compounds which act as a source of energy ([Zhao et al., 2017](#)). Farmyard manure (0.126%) and vermicompost (0.122%) were found more effective on N compared to leonardite (0.117%) in this study (OA means in Figure 2). [Öktüren Asri et al. \(2024\)](#), reported that soil total nitrogen was increased by 7.7% - 20% with organic amendments at the end of the first growing season and this positive effect continued in the following year and caused increases of 12% - 25%. In this study, C/N rate of leonardite, vermicompost and farmyard manure were 9.64, 10.76 and 9.48, respectively (Table 2). According to [Chen et al. \(2018\)](#), a lower soil C/N ratio causes microorganisms more limited by C than by N, especially when this ratio is lower than 13-15. In this regard, N excess may easily occur with N deposition in ecosystems with low soil C/N, and C limitation accompanied by a C/N stoichiometric imbalance is also more likely to control the response of soil N mineralization to N deposition ([Song et al., 2022](#)).

Residual effects of organic amendments, application level and their interaction on soil available phosphorus (av-P) concentration were found to be statistically significant ($p < 0.001$) (Table 3). After the applications, the lowest av-P concentration (24.0 mg

kg⁻¹) was obtained from 0 t ha⁻¹ leonardite and the highest value (56.3 mg kg⁻¹) was determined by 40 t ha⁻¹ farmyard manure (Figure 2). While doses of organic amendments caused to enhance soil av-P concentration by 26.5%-39.9% compared to the control, residual effects of organic amendments increased av-P concentration by 36.4%-40.2% and the most effective material was FYM. [Sheoran et al. \(2024\)](#) reported that the application of 15 t of FYM ha⁻¹ resulted in a considerable increase av-P by 9.7%–12.1%.

Residual effects of organic amendments, application level and their interaction on soil exchangeable potassium (ex-K) concentration were found to be statistically significant ($p < 0.001$) (Table 3). Soil ex-K were increased by 27%-39% residual effects of organic amendments and 23%-48% their ascending doses (Figure 2). In this study soil had 34.88% clay, 35.28% silty and 29.84% sand, with clay loam texture. Soil clay content may be resulted in increasing soil ex-K concentration. Rai et al. (2014) explained that organic amendments increase soil ex-K by reducing K fixation and release due to the interaction of organic matter with clay, in addition to direct addition of K to the av-K pool of soils. [Yadav et al. \(2019\)](#) reported that FYM (15 t ha⁻¹) significantly increased soil ex-K concentration by 37% over inorganic fertilizer application. [Najafi Ghiri \(2014\)](#) reported that the mean increase in cumulative K release with vermicompost application was 88% compared to the control.

The residual effects of organic amendments were found to be not effective on soil exchangeable calcium (ex-Ca) concentration (Table 3). The residual effects of organic amendments were found to be not effective on soil exchangeable magnesium (Mg) concentration. But

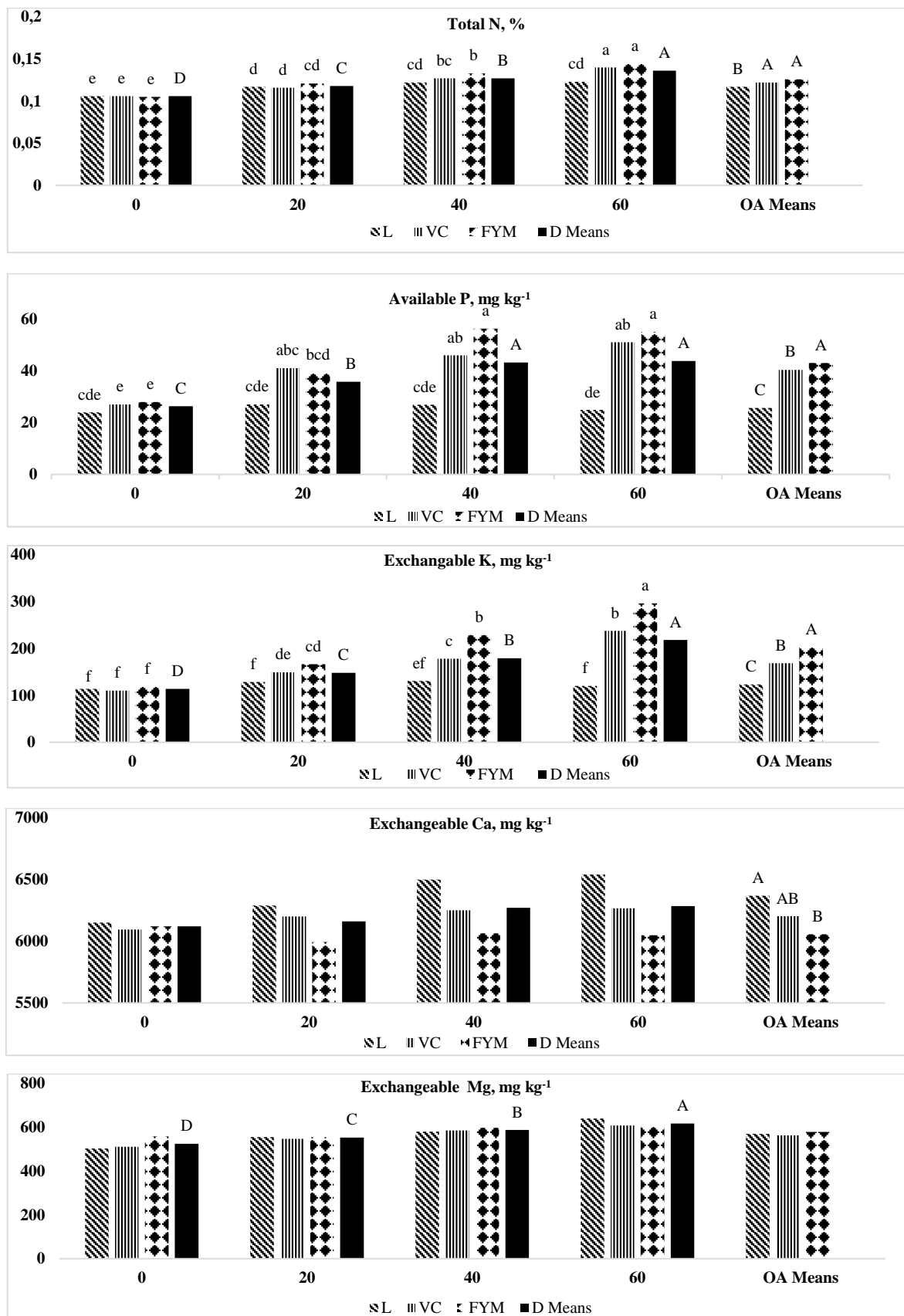


Figure 2. Residual effects of organic amendments on soil macro elements concentration

Lowercase letters indicate a significant difference in the interaction between organic amendments and their doses. Uppercase letters indicate a significant difference between the organic amendments (OA Means). Uppercase letters indicate a significant difference between the doses (D Means).

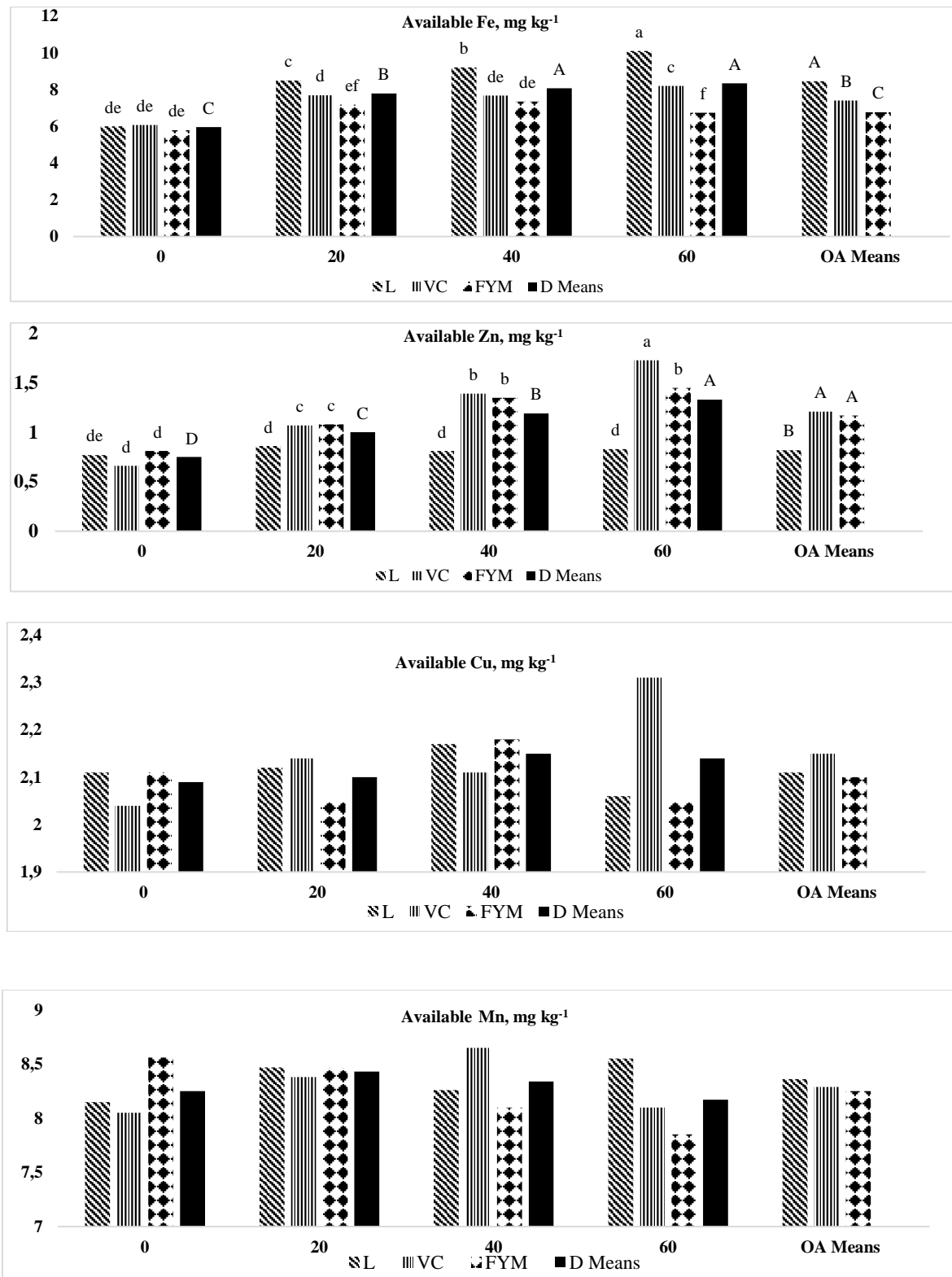


Figure 3. Residual effects of organic amendments on soil micro elements concentration

Lowercase letters indicate a significant difference in the interaction between organic amendments and their doses. Uppercase letters indicate a significant difference between the organic amendments (OA Means). Uppercase letters indicate a significant difference between the doses (D Means).

the increasing doses caused to soil ex-Mg concentration by 5%-15%. The highest ex-Mg concentration was obtained from 60 t ha⁻¹ dose of organic amendments (Figure 2). Demir (2019) found that vermicompost application increased soil ex-Ca and ex-Mg concentrations. Agbede et al. (2013) found that application of organic fertilizers tended to improve soil pH, organic C, total N, and exchangeable K, Ca and Mg more than chemical fertilizers.

Residual effects of organic amendments, application level and their interaction on soil available iron (av-Fe) concentration were found to be statistically significant ($p < 0.001$) (Table 3). The lowest av-Fe concentration (5.80 mg kg⁻¹) was obtained from 0 t ha⁻¹ farmyard manure, while the highest value (10.12 mg kg⁻¹) was attained by 60 t ha⁻¹ leonardite (Figure 3). Available Fe concentrations of soil were increased by 24%-29% residual effects of organic amendments and 9%-20% their ascending doses. Nuzzo et al. (2018) reported that more than 95% of the total plant-available Fe in the soil solution may be represented by organic Fe pool. An important Fe source in soil is represented by the insoluble Fe complexes with humic substances (HS). Iron complexation by humic substances is attributed to the oxygen-containing functional groups (carboxylic, phenolic and carbonyl). Thus, it is thought that humic substances and functional groups content of organic materials resulted in an increase in av-Fe concentration of soil in this study.

Conclusions

The aim of this study was to determine the residual effects of vermicompost, leonardite, farmyard manure and their increasing doses on soil properties. Residual effects of organic amendments were increased electrical conductivity, organic matter, total N, available P, exchangeable K, available Fe and Zn concentrations of the soil. Among application doses, 60 t ha⁻¹ dose was more effective for nutrient release. These results indicated that these treatments had compensated more than the nutrient removal of tomato seedlings for 10th weeks as evidenced from significant increase in available elements in soil. Based on the results, it is concluded that growers can take advantage of the long-term benefits of nutrients supplied from single or repeated organic amendments application to improve soil quality and reduce N, P and K fertilizer input cost.

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Author Contribution

S.S.: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Supervision, Resources, Validation, Writing – review & editing. **F.Ö.A.:** Formal analysis, Investigation, Statistical analysis, Visualization, Software, Writing-original draft, Writing –review & editing.

Conflict of Interest

No potential conflict of interest was reported by the authors.

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Effects of conditioner applications on organic matter and nitrogen content in sandy and clay loam textured soils

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Polyacrylamide

Humic acid

Abstract

This study aimed to investigate the effects of polyacrylamide (PAM) and humic acid (HA), wheat straw (WS) and hazelnut husk (HH) application on wheat yield, soil organic matter and nitrogen content of sandy and clay loam texture under greenhouse conditions. The study was conducted according to the experimental design of the randomized complete block experimental design. Wheat straw and hazelnut husk were applied at 0%, 2% and 4% doses, polyacrylamide at 0 ppm, 30 ppm, 60 ppm and 90 ppm, and humic acid at 0 ppm, 200 ppm and 1000 ppm. After five months of incubation period, wheat plants were grown in pots. The study was completed in a total of 312 days along with the incubation period. It was determined that the treatments affected the organic matter and total nitrogen content. As a result of the applications, it was determined that the organic matter content was more affected in clay loam textured soil, but the nitrogen content and wheat yield were more affected in sandy loam textured soil. It has been determined that the application of synthetic conditioners together with plant residues is the most effective application in increasing organic matter and nitrogen content.

Introduction

Soil carbon is important in preventing the effects of degradative processes (erosion, pollution, acidification, salinization) in the soil, maintaining its fertility and reducing carbon flow to the atmosphere. Most of the carbon, which can be found in different forms, is formed by the decomposition products of dead plant and animal tissues and microbial masses living in the soil ([Aşkın et al., 2014](#); [Jangir et al., 2019](#)). In today's conditions, agricultural practices to meet the needs of the increasing population and developing economies negatively affect soil quality, reduce productivity and lead to the need for more fertilization, irrigation and pesticides.

Soil organic carbon is significantly affected by land use and agricultural practices. [Dengiz et al. \(2015\)](#), who investigated the effects of land use and land cover on organic matter stocks in Madendere Basin, determined that soil organic carbon was significantly affected by land use and land cover, and the land use patterns in terms of density were forest (6.30 kg/m²), pasture (5.17 kg/m²), orchard (4.69 kg/m²) and cultivated land (3.85 kg/m²), respectively. [Yimer et al. \(2006\)](#) examined the changes in soil organic carbon and total nitrogen stocks depending on topography and vegetation in Bale Mountain in Ethiopia. They found that higher altitudes had more organic carbon and nitrogen than lower

altitudes. [Özdemir and Bülbül \(2024\)](#), in a study conducted on agricultural lands, found that soil organic matter content was significantly affected by basic soil properties and land use patterns. The researchers determined that eight different land use types on organic matter content were effective in the order of sugar beet < pasture < meadow < sunflower < orchard < wheat < alfalfa.

In recent years, synthetic soil conditioners have been widely used, along with organic-based fertilizers, to maintain and improve the functional properties of soil and increase the organic carbon content. For this purpose, barnyard manure ([Balik et al., 2023](#)), residential wastes ([Pedra et al., 2007](#); [Özdemir et al., 2014](#)), composted materials ([Tejada et al., 2003](#)), post-harvest crop residues ([De Neve et al., 2000](#)), organic by-products of agro-industrial enterprises ([Madejon et al., 2001](#); [Iqbal et al., 2019](#)) and the combined use of inorganic and organic fertilizers are emphasized. The physical and chemical properties of soils are positively affected by these materials' application; their erosion resistance increases, and soil organic carbon increases ([Zhang et al., 2019](#); [Liu et al., 2024](#)). [Leaungvutivirog et al. \(2002\)](#) examined the effects of chemical and organic fertilizers on the chemical and microbiological properties of soils and the yield of maize plants. [Tamer et al. \(2016\)](#) evaluated the effects of organic source fertilizer application on sunflower plants' soil properties and yield components. As a result of the study, they determined that 30 kg ha⁻¹ humic acid + chemical fertilizer application had the most effect on organic matter content. Moreover, the applications also affected pH, EC, lime, potassium (K) and phosphorus (P) contents.

In today's conditions, synthetic origin conditioners that provide faster results in protecting and improving soil properties, preventing erosion and improving plant production have begun to be emphasized. In this study; the effects of the application of wheat straw and hazelnut husk waste and humic acid and polyacrylamide separately and together with PAM on organic matter and total nitrogen content in the soil were examined.

Materials and Methods

The study was conducted on surface (0-20 cm) soil samples with two different textures (Sandy loam: SL, clay loam: CL) under greenhouse conditions. The samples were taken from two different locations (SL 41°55'-35°86'; CL 41° 61'-35° 90') where agricultural production is practiced in the Bafra region. Bafra Plain is located in the semi-humid humid moisture regime (Ustik) with an average annual temperature of 14.5 °C and an average annual precipitation of 716.6 mm ([Saygın et al., 2012](#)). Organic wastes, PAM and HA used in the study were obtained from different institutions.

The analysis results of the soils and crop residues used in the study are given in Table 1. As can be seen from the analysis of these data, the sandy loam (SL) soil taken from the Bafra region has a slightly alkaline character, saltless, low organic matter, and nitrogen content. Clay loam texture (CL) has a slightly alkaline character, is salt-free, has moderate organic matter and has high nitrogen content. The pH values of the soils are below 8.5, and there is no alkalinity problem ([Soil Survey Manual, 2017](#)).

Table 1. Analysis results of soils and organic residues

Soils	Sand, %	Silt, %	Clay, %	pH (1:2.5)	EC (1:1) (dS m ⁻¹)	OM (%)	CaCO ₃ (%)	N (%)	P (ppm)	Ca+Mg (me 100 g ⁻¹)
Soil (SL)	64.12	27.91	7.97	8.11	0.68	1.12	17.82	0.05	48.07	21.84
Soil (CL)	27.19	41.51	31.30	7.88	0.47	2.85	7.57	0.20	25.45	36.25
Organic wastes	pH (1:10)	EC (1:10) (µmhos cm ⁻¹)		OC (%)	Total N (%)	C/N	Ash (%)		P (ppm)	
Wheat Straw	5.69	2848.50		53.46	0.65	82.25	7.84		2055.00	
Hazelnut husk	6.16	2058.00		46.93	1.86	25.28	19.09		6291.52	

(SL: Sandy loam soil, CL: Clay loam soil)

Wheat straw and hazelnut husk used in the study had C/N ratio values of 82.25 and 25.28, respectively. The PAM used in the study has an anionic character $[-CH_2CHCONH_2-]_n$ and a molecular weight of approximately $10000 \text{ Mg.mol}^{-1}$ with a density value of 1.189 g/cm^3 . The experiment used commercially available liquid material containing 13.5% humic acid, 1.5% fulvic acid and 1.5% K_2O .

The factorial study used wheat (*Triticum aestivum*, *pandas variety*) as plant material. Soils were sieved through a 4 mm sieve and transferred to 5.5 kg pots; polymers and organic wastes were added at three different doses (WS 0, 2, 4%; HH 0, 2, 4%; HA 0, 200, 1000 ppm; PAM 0, 30, 60 ppm and 90 ppm) and PAM+conditioner doses. During the experiment, irrigation was applied again when half of the available moisture in the soil was depleted (Field capacity SL:16.84 and CL:6.78). Wheat plants were grown in the pots after five months of incubation. After the plant harvest, soil samples from the pots were analyzed and evaluated.

Soil texture; Bouyoucos hydrometer method (Demiralay, 1993), organic matter content (OM); Walkley-Black method (Kacar, 1995), lime ($CaCO_3$) content by volume; Scheibler calcimeter method (Kacar, 1995), calcium and magnesium; ammonium acetate extraction method (Kacar, 1995), total nitrogen; Kjeldahl method (Bremner, 1965), available phosphorus; Olsen according to pH (Sims, 2009), pH and electrical conductivity (EC) values; pH meter in saturation sludge (Hendershot et al., 1993) and EC meter (Hendershot et al., 1993). SPSS 22 package program and Duncan multiple comparison tests were used to statistically evaluate the data.

Results and Discussion

Organic matter

In this study in greenhouse conditions, synthetic conditioners of organic origin (polyacrylamide, humic acid) and organic wastes of plant origin (wheat straw and hazelnut husk) were applied separately and together to surface soil samples with two different textures (SL, CL) and left for incubation and wheat plants were grown. The organic matter contents results of the analysis of variance are given in Table 2, and the results of Duncan's multiple comparison tests are given in Table 3. There were differences in the post-harvest organic matter content of the soils according to the type and dose of application. Soil organic matter, composed of residues of plant and animal origin, is a dynamic component and is significantly affected by agricultural practices. The organic matter content of the SL in the experiment changed from 1.14% to 2.43% depending on the conditioner applications. When the conditioners were applied separately, it was determined that the organic matter content of the soils increased after the wheat straw and hazelnut husk applications compared to the control soil. At the same time, it decreased after the humic acid and PAM applications compared to the control soil. In terms of the said effect, when the separate application activities of the conditioners used were examined, it was seen that they were ranked as $HH > WS > PAM > HA$. On the other hand, when the effects of applying the conditioners together were examined, it was determined that they were ranked as $HH+PAM > WS+PAM > HA+PAM$ in terms of the increase in organic matter content.

Table 2. Analysis of variance results for organic matter values

Source of variation		Degrees of freedom	Sum of Squares	Mean Squares	F Value	Significance level
University	Variable (A)	6	7.668	1.278	77.641	.000
Soil (SL)	Treatment (B)	3	.280	.093	5.668	.002
	AxB	18	.714	.040	2.411	.006
	Error	56	.922	.016		
	Total	84	220.642			
Research	Variable (A)	6	117.413	19.569	613.304	.000
Soil (CL)	Treatment (B)	3	1.180	.393	12.332	.000
	AxB	18	6.751	.375	11.755	.000
	Error	56	1.787	.032		
	Total	84	1222.333			

$P < 0.05$ (A: Variable, B: Treatment, SL: Sandy loam soil, CL: Clay loam soil)

Table 3. Duncan test results of organic matter values

Treatments	Mean \pm Standard deviation			
	University area. (SL)		Research area (CL)	
Control	1.14	± 0.07 l	2.37	± 0.12 i
%2 WS	1.62	± 0.18 c-e	3.17	± 0.21 h
%4 WS	1.67	± 0.21 cd	5.58	± 0.30 b
%2 HH	1.60	± 0.17 c-f	4.21	± 0.23 f
%4 HH	2.25	± 0.12 a	6.24	± 0.29 a
HA 1.dose	1.17	± 0.01 kl	2.47	± 0.07 i
HA 2.dose	1.30	± 0.04 h-l	2.47	± 0.08 i
PAM 1.dose	1.26	± 0.04 j-l	2.42	± 0.03 i
PAM 2.dose	1.35	± 0.01 g-l	2.43	± 0.01 i
PAM 3.dose	1.36	± 0.02 f-l	2.47	± 0.03 i
%2WS+PAM 1.dose	1.27	± 0.14 i-l	3.16	± 0.10 h
%2WS+PAM 2.dose	1.48	± 0.12 d-j	3.23	± 0.29 h
%2WS+PAM 3.dose	1.51	± 0.08 c-i	3.65	± 0.24 g
%4WS+PAM 1.dose	1.63	± 0.08 c-e	4.62	± 0.21 de
%4WS+PAM 2.dose	1.65	± 0.08 c-e	4.35	± 0.32 ef
%4WS+PAM 3.dose	1.74	± 0.14 c	5.12	± 0.11 c
%2HH+PAM 1.dose	1.54	± 0.15 c-h	3.78	± 0.16 g
%2HH+PAM 2.dose	1.56	± 0.09 c-g	3.90	± 0.08 g
%2HH+PAM 3.dose	1.74	± 0.26 c	4.61	± 0.12 de
%4HH+PAM 1.dose	2.43	± 0.37 a	5.64	± 0.24 b
%4HH+PAM 2.dose	2.01	± 0.07 b	5.58	± 0.13 b
%4HH+PAM 3.dose	2.35	± 0.14 a	4.73	± 0.15 d
HA 1.dose+PAM 1.dose	1.40	± 0.00 e-k	2.54	± 0.24 i
HA 1.dose+PAM 2.dose	1.44	± 0.02 d-j	2.44	± 0.17 i
HA 1.dose+PAM 3.dose	1.54	± 0.01 c-h	2.46	± 0.08 i
HA 2.dose+PAM 1.dose	1.40	± 0.00 e-k	2.54	± 0.10 i
HA 2.dose+PAM 2.dose	1.44	± 0.02 d-j	2.45	± 0.13 i
HA 2.dose+PAM 3.dose	1.54	± 0.01 c-h	2.48	± 0.15 i

(PAM: polyacrylamide, HA: humic acid, WS: wheat straw, HH: hazelnut husk, SL: Sandy loam soil, CL: Clay loam soil)

The organic matter content of the CL in the experiment, which was 2.37%, changed to 6.01% depending on the conditioners applications. In terms of the said effect, when the separate application efficiency of the conditioners used was examined, it was seen that they were ranked as HH>WS>HA>PAM. On the other hand, when the conditioners were applied together, the ranking of their effect on organic

matter content was HH+PAM > WS+PAM > HA+PAM. When the variance analysis results of the effects of conditioner and dose applications on organic matter values in both soil groups were evaluated (Table 2), it was observed that the mean squares of conditioner and application levels were significant ($p < 0.05$). When the results of the Duncan multiple comparison test for the significant sources of variation were analyzed, it was

determined that the treatment averages generally differed (Table 3). When the variance analysis results of the effects of conditioner and dose applications on organic matter values in both soil groups were evaluated (Table 2), it was determined that the mean squares of the conditioner and application levels were significant ($p < 0.05$) and the means generally differed according to the Duncan multiple comparison test results (Table 3).

In the SL, the pots in which the second dose of hazelnut husk (%4HH) was applied alone and the pots in which the second dose of hazelnut husk was applied with the first and third doses of polyacrylamide (%4HH+30ppmPAM, %4HH+90ppmPAM) were similar.

When the proportional changes caused by the treatments according to the control organic matter values of the soils were examined, it was determined that the conditioners were more effective in the soil taken from the research land with CL texture than the soil taken from the university land with SL texture. This can probably be attributed to the ability of clay to slow down mineralization and retain organic matter. In many studies in this direction ([Parton et al., 1994](#); [Schimel et al., 1994](#); [Lantz et al., 2002](#)), it has been determined that texture, especially clay content, is significantly effective in reducing C binding and losses in soil. At the same time, an increase of 113.15% was realized in the soil of the university land as a result of the treatments compared to the experiment; an increase of 163.29% was realized in the soil of the research land. The most effective dose among the treatments was HH2+PAM1 (%4HH+30ppmPAM) in the soil belonging to the university land, while HH2 (%4HH) in the soil belonging to the research institute.

[Agglides and London \(2000\)](#) reported that compost applied to soils improved the chemical

properties of soils and increased soil organic matter. [Demir et al. \(2006\)](#) reported a significant increase in soil organic matter after tobacco waste and hazelnut waste application compared to the control soil. [Özdemir and Bülbül \(2024\)](#), in their study conducted in Turhal conditions, determined that soil organic matter content was significantly affected by basic soil properties and land use type and that eight different land use types were effective in terms of this effect in the form of sugar beet < pasture < meadow < sunflower < orchard < wheat < alfalfa.

Nitrogen

In this study in greenhouse conditions, synthetic organic-based conditioners (polyacrylamide, humic acid) and organic wastes of plant origin (wheat straw and hazelnut husk) were applied separately and together to soils with two different textures and left for incubation and then wheat plants were grown in pots. The nitrogen contents (average) results of the analysis of variance for these values are given in Table 4, and the results of Duncan's multiple comparison tests are given in Table 5. The conditioners applied separately and together in both soil groups showed increases in the total nitrogen contents of the soils after harvesting according to the application type and dose.

The nitrogen contents (average) results of the analysis of variance are given in Table 4, and the results of Duncan's multiple comparison tests are given in Table 5. The conditioners applied separately and together in both soils showed increases in the total nitrogen contents according to the application type and dose.

The nitrogen content of the SL increased from 0.06% to 0.16%, depending on the conditioner applications. When the conditioners were applied

Table 4. Analysis of variance results for total nitrogen values

Source of variation		Degrees of freedom	Sum of Squares	Mean Squares	F Value	Significance level
University Soil (SL)	Variable (A)	6	.041	.007	63.252	.000
	Treatment (B)	3	.001	.000	4.337	.008
	AxB	18	.004	.000	2.081	.019
	Error	56	.006	.000		
	Total	84	.902	.		
Research Soil (CL)	Variable (A)	6	.117	.019	106.316	.000
	Treatment (B)	3	.000	7.103E-005	.387	.762
	AxB	18	.007	.000	2.039	.022
	Error	56	.010	.000		
	Total	84				

P<0.05 (A: Variable, B: Treatment, SL: Sandy loam soil, CL: Clay loam soil)

separately, it was determined that the nitrogen content increased compared to the control soil, and this increase was directly proportional to the increase in the dose of the conditioners. In terms of the said effect, when the effectiveness of the conditioners applied separately was examined, it was seen that they were ranked as HH>WS>HA>PAM. On the other hand, when the effects of the application of the conditioners

together were analyzed, it was determined that there was an increase in nitrogen content compared to the control, and only in some doses of HA+PAM applications was there a decrease. Regarding the increase they caused, it was determined that the conditioners were ranked as HH+PAM>WS+PAM>HA+PAM.

Table 5. Duncan test results for total nitrogen values

Treatments	Mean \pm Standard deviation			
	University area. (SL)		Research area (CL)	
Control	0.08	± 0.01 j-l	0.15	± 0.00 i
%2 WS	0.11	± 0.01 e-h	0.16	± 0.01 g-i
%4 WS	0.11	± 0.02 e-g	0.22	± 0.01 c
%2HH	0.12	± 0.01 c-e	0.21	± 0.02 cd
%4 HH	0.14	± 0.01 ab	0.24	± 0.01 b
HA 1.dose	0.08	± 0.01 i-k	0.16	± 0.02 hi
HA 2.dose	0.11	± 0.01 e-h	0.16	± 0.01 g-i
PAM 1.dose	0.08	± 0.01 i-k	0.16	± 0.01 g-i
PAM 2.dose	0.08	± 0.01 i-k	0.16	± 0.01 g-i
PAM 3.dose	0.09	± 0.00 h-k	0.16	± 0.01 hi
%2WS+PAM 1.dose	0.09	± 0.01 g-j	0.18	± 0.01 f-i
%2WS+PAM 2.dose	0.09	± 0.00 h-k	0.19	± 0.01 d-g
%2WS+PAM 3.dose	0.10	± 0.01 f-i	0.18	± 0.01 f-i
%4WS+PAM 1.dose	0.11	± 0.01 d-f	0.19	± 0.02 d-f
%4WS+PAM 2.dose	0.11	± 0.01 d-f	0.18	± 0.01 e-h
%4WS+PAM 3.dose	0.11	± 0.01 d-f	0.19	± 0.01 c-f
%2HH+PAM 1.dose	0.11	± 0.01 d-f	0.20	± 0.00 c-f
%2HH+PAM 2.dose	0.12	± 0.02 c-e	0.20	± 0.01 c-e
%2HH+PAM 3.dose	0.11	± 0.01 e-h	0.21	± 0.03 cd
%4HH+PAM 1.dose	0.13	± 0.01 b-d	0.27	± 0.03 a
%4HH+PAM 2.dose	0.13	± 0.02 bc	0.28	± 0.00 a
%4HH+PAM 3.dose	0.16	± 0.01 a	0.28	± 0.02 a
HA 1.dose+PAM 1.dose	0.07	± 0.00 kl	0.16	± 0.02 hi
HA 1.dose+PAM 2.dose	0.07	± 0.00 kl	0.16	± 0.02 hi
HA 1.dose+PAM 3.dose	0.06	± 0.01 l	0.16	± 0.01 g-i
HA 2.dose+PAM 1.dose	0.07	± 0.00 kl	0.16	± 0.01 hi
HA 2.dose+PAM 2.dose	0.08	± 0.01 i-k	0.15	± 0.01 i
HA 2.dose+PAM 3.dose	0.08	± 0.01 j-l	0.15	± 0.00 i

(PAM: polyacrylamide, HA: humic acid, WS: wheat straw, HH: hazelnut husk, SL: Sandy loam soil, CL: Clay loam soil)

The nitrogen content in the soil of control agricultural research land, which was 0.151%, increased to 0.282% depending on the conditioner applications. In terms of the said effect, when the separate application activities of the conditioners used were examined, it was seen that they were ranked as HH>WS>PAM>HA. On the other hand, when the conditioners were applied together, it was determined that they were ranked as HH+PAM>WS+PAM>HA+PAM in terms of their effect on nitrogen content.

When the variance analysis results of the effects of conditioner and dose applications on total nitrogen values in both soil groups were evaluated (Table 4), it was observed that the mean squares of the conditioner and application levels were significant ($p<0.05$). When the results of the Duncan multiple comparison test for the significant sources of variation were analyzed, it was determined that the treatment averages generally differed (Table 5).

As a result of the applications in SL, there was a 110.66% increase compared to the control, while there was an 87.41% increase in CL. This was probably due to increased sand content, which increased mineralization. [Wang et al. \(2006\)](#), in a study investigating the effects of sand size distribution on nitrogen content in soils under different land use conditions, determined that increasing the amount of sand from 5.01 to 8.60% increased nitrogen content from 0.223% to 0.844%. Similarly, a 110.66% increase was observed in the university land soil compared to the control, while an 87.41% increase was observed in the research land soil. The most effective dose among the treatments was HH2+PAM3 (%4HH+90ppmPAM) in both the university and the research institute soil.

[Coşkun et al. \(2006\)](#) investigated the effects of tobacco waste (0.0, 2.0, 4.0 and 6.0%) and PAM (0.0, 15.0, 30.0 and 60.0 ppm) applied to soils with different

levels of erosion on the nitrogen and phosphorus content available to plants. At the end of the study, it was stated that tobacco waste and PAM applications increased the nitrogen content in the soil, and the increase in nitrogen content was greater in the application of tobacco waste.

Wheat yield

In greenhouse conditions, two different organic synthetics (polyacrylamide, humic acid) and plant-based organic wastes (wheat straw and hazelnut husk) were applied separately and together to incubate and then the yield of the wheat (average) is given in Table 7. According to the results, applications in both soil groups showed differences in yield according to the application and application dose. According to the yield results of wheat plants harvested after applications in sandy loam soil, it was determined that the yield of wheat plants increased after HA1, HA2, FZ1+PAM1, FZ1+PAM2, FZ1+PAM3, HA1+PAM2, HA2+PAM1, HA2+PAM2 and HA2+PAM3 applications. According to these results, it was determined that the individual application efficiencies of the regulators used were as HA>FZ>PAM>PAM. When the regulators were applied together, there was an increase in yield compared to the control, but it was determined that yield decreased in BS + PAM applications. This is probably related to the decomposition process of wheat straw. It was determined that their effects on wheat yield were ranked as FZ+PAM>HA+PAM>BS+PAM.

After the applications in clay loam soil, it was observed that the yield increased compared to the control in pots where only hazelnut husk was applied together with polyacrylamide, and the yield decreased in other applications. This is probably related to the slowing down of the mineralization process in fine-structured soil.

Table 6. Analysis of variance results for wheat yield values

Source of variation		Degrees of freedom	Sum of Squares	Mean Squares	F Value	Significance level
University Soil (SL)	Variable (A)	6	180153.619	30025.603	81.243	.000
	Treatment (B)	3	5702.086	1900.695	5.143	.003
	AxB	18	14061.476	781.193	2.114	.017
	Error	56	20696.333	369.577		
	Total	84	3478402.875			
Research Soil (CL)	Variable (A)	6	153424.509	25570.751		.000
	Treatment (B)	3	3185.048	1061.683		.003
	AxB	18	15887.223	882.624		.017
	Error	56	12158.208	217.111		
	Total	84	4598863.750			

P<0.05 (A: Variable, B: Treatment, SL: Sandy loam soil, CL: Clay loam soil)

Table 7. Duncan test results for wheat yield value (kg/da)

Treatments	Mean \pm Standard deviation	
	University area. (SL)	Research area (CL)
Control	224.33 \pm 23.82 b-d	265.17 \pm 16.23 a-d
%2 WS	161.17 \pm 2.27 g	204.92 \pm 9.64 hi
%4 WS	107.67 \pm 8.80 h	139.83 \pm 10.03 jk
%2HH	218.75 \pm 9.97 c-e	259.83 \pm 11.50 a-e
%4 HH	184.75 \pm 32.45 e-g	203.33 \pm 12.63 hi
HA 1.dose	232.42 \pm 15.35 a-d	235.92 \pm 8.18 e-g
HA 2.dose	237.5 \pm 12.42 a-d	246 \pm 24.49 d-g
PAM 1.dose	163.33 \pm 35.37 g	247.33 \pm 15.26 d-g
PAM 2.dose	206.33 \pm 18.57 d-f	243.58 \pm 11.67 d-g
PAM 3.dose	216.42 \pm 18.06 c-e	264.33 \pm 12.79 a-d
%2WS+PAM 1.dose	161.00 \pm 22.00 g	190.08 \pm 20.08 i
%2WS+PAM 2.dose	171.5 \pm 14.51 fg	182.83 \pm 15.93 i
%2WS+PAM 3.dose	173.25 \pm 12.01 fg	200.67 \pm 17.18 i
%4WS+PAM 1.dose	98.25 \pm 13.54 h	140.33 \pm 9.68 jk
%4WS+PAM 2.dose	107.42 \pm 5.03 h	156.75 \pm 10.18 j
%4WS+PAM 3.dose	86.5 \pm 12.67 h	128.17 \pm 7.56 k
%2HH+PAM 1.dose	247 \pm 33.00 a-c	278.25 \pm 27.15 a-c
%2HH+PAM 2.dose	243 \pm 16.20 a-d	281.92 \pm 0.80 a-c
%2HH+PAM 3.dose	265.25 \pm 10.43 a	286.75 \pm 1.95 a
%4HH+PAM 1.dose	208.25 \pm 20.81 de	284.17 \pm 18.57 ab
%4HH+PAM 2.dose	210.08 \pm 17.72 c-e	254.42 \pm 26.25 c-g
%4HH+PAM 3.dose	218.17 \pm 14.02 c-e	276.42 \pm 4.64 a-c
HA 1.dose+PAM 1.dose	184.83 \pm 27.19 e-g	246 \pm 13.31 d-g
HA 1.dose+PAM 2.dose	238.33 \pm 36.17 a-d	229.42 \pm 14.51 f-h
HA 1.dose+PAM 3.dose	224.33 \pm 5.03 b-d	254.42 \pm 4.25 c-g
HA 2.dose+PAM 1.dose	226.5 \pm 14.95 b-d	233.83 \pm 22.04 e-g
HA 2.dose+PAM 2.dose	257 \pm 11.69 ab	227.42 \pm 14.22 gh
HA 2.dose+PAM 3.dose	240.83 \pm 13.77 a-d	256.58 \pm 5.86 b-f

(PAM: polyacrylamide, HA: humic acid, WS: wheat straw, HH: hazelnut husk, SL: Sandy loam soil, CL: Clay loam soil)

When the changes caused by the applications compared to the control were examined proportionally, it was determined that the regulators were more effective in sandy loam soil than in clayey loam soil. As a result of the applications in sandy loam soil, there was an 18.28% increase compared to the control, while there was an 8.10% increase in clayey loam soil. [Thenmozhi et al. \(2004\)](#), In their study investigating the effect of humic acid on the quality characteristics of peanuts, they reported that the best

values would be obtained when 20 kg of humic acid per decare was applied together with chemical fertilizer. [Erdem et al. \(2020\)](#) applied humic acid, rhizobacteria and chemical fertilizers in their study investigating the effects of different fertilizer applications on wheat yield. According to the results obtained, they reported that humic acid application increased the number of ears per square meter, plant height, number of grains per ear and grain yield depending on the wheat variety.

As a result of the applications, it was determined that there was a positive relationship between nitrogen and organic matter content in the soil and yield (Table 8). However, this relationship was not found to be statistically significant (* $P < 0.05$ ** $P < 0.01$).

Table 8. Correlation between wheat yield, organic matter and nitrogen

n: 28	Soil (SL)		Soil (CL)	
	OM	N	OM	N
Wheat yield	0.103	0.157	0.266	0.108

(SL: Sandy loam soil, CL: Clay loam soil)

(* $P < 0.05$ ** $P < 0.01$)

Conclusions

When the effects of the treatments on soil organic matter and total nitrogen content were evaluated after harvesting the plant, it was determined that both total organic matter and total nitrogen contents were affected by the treatments. Regarding the changes in organic matter contents, it was determined that the treatments were more effective in the soil with CL texture than in the soil with SL texture. Indeed, while an increase of 113.15% was realized in the soil belonging to the university land (SL) due to the treatments compared the control, an increase of 163.29% was realized in the soil belonging to the research land (CL). The most effective dose among the treatments was HH2+PAM1 (%4HH+30 ppm PAM) in the soil belonging to the university land, while HH2 (%4HH) in the soil belonging to the research institute. On the other hand, when the changes in the total nitrogen content of the soils were analyzed proportionally, it was determined that the conditioners were more effective in the soil taken from the university land with SL texture than the soil taken from the research land with CL texture. The change occurred depending on the conditioner type and application dose. In the university soil, the highest increase was realized in all of the samples in which the and dose of hazelnut husk (HH2) was applied with polyacrylamide. In the research soil, the highest increase was again determined in the samples in which the 2nd dose of hazelnut husk (HH2) was applied with the 2nd and 3rd doses of polyacrylamide.

As a result, it was determined that separate and combined application of organic conditioners increased the organic matter and nitrogen contents of the soils, and the effects depended on the nature of the organic material and soil properties. It was determined that co-applicating synthetic conditioners with crop residues was the most effective application in increasing organic matter and nitrogen content. It is important to consider soil properties, conditioner properties, doses, and combinations to be applied in applications. The

findings were obtained under limited greenhouse conditions, and extending the study to field applications would be useful.

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Author Contributions

Ö.T.K.D.: Conceptualization, investigation, methodology, software, validation, formal analysis, investigation, resources, data curation, writingoriginal draft preparation, writing-review and editing, visualization, supervision, statistical analysis, project administration. **N.Ö.:** Conceptualization, investigation, methodology, software, validation, formal analysis, investigation, resources, data curation, writingoriginal draft preparation, writing-review and editing, visualization, supervision, statistical analysis, project administration.

Conflict of Interest

The authors declare that they have no known competing financial or non-financial, professional, or personal conflicts that might apper to influence the work reported in this paper.

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Impact of increasing vermicompost applications on the growth performance of radish (*Raphanus sativus*) in cadmium-contaminated soil

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Abstract

Soil contamination by heavy metals, including cadmium (Cd), adversely impacts plant growth and affects agricultural productivity. Organic improvements, especially vermicompost (VC), may reduce metal toxicity. This study investigated the impact of increasing VC dosages on the growth of radish (*Raphanus sativus*) under Cd stress. A greenhouse experiment was performed using a randomized plot design with four replications. In this experiment, four different vermicompost (VC) rates (0%, 1%, 2%, and 4%) and three Cd concentrations (0, 5, and 10 mg kg⁻¹) were applied. The Cherry Belle cultivar was cultivated and harvested 45 days post-sowing. The shoot's length and weight (fresh and dry), the tuber's diameter, and the tuber's weight (fresh and dry) were all measured. The vermicompost treatment significantly ($p \leq 0.01$) enhanced all parameters. The lowest values have been determined without vermicompost at 10 mg Cd kg⁻¹. The maximum shoot length and tuber diameter were achieved with 4% vermicompost and no cadmium, whereas the highest shoot and tuber weights were obtained with 4% vermicompost at 10 mg Cd kg⁻¹. In summary, vermicompost reduced Cd toxicity and improved radish growth and yield, suggesting its potential as a sustainable solution for soils contaminated with heavy metals.

Introduction

Heavy metal contamination in soil has become a global concern due to the increasing use of chemical fertilizers, soil conditioners, pesticides, sewage sludge, and wastewater in agricultural practices (Khan et al., 2007). These metals accumulate in soils through agricultural activities, mining, industrial processes, vehicular emissions, and natural events such as volcanic eruptions (Rodríguez-Hernández et al., 2022; Yang et al., 2022). Among these metals, Cd is considered one of the most hazardous due to its high

toxicity, mobility, and persistence in the environment. Cd has no known biological function in plants, animals, or humans, and poses serious risks to both plant growth and human health (Marschner, 2008). It enters agricultural soils through natural sources, such as parent materials rich in heavy metals, as well as anthropogenic inputs, including atmospheric deposition, phosphate fertilizers, and sewage sludge application. Cd contamination reduces plant productivity by disrupting key physiological and

biochemical processes such as photosynthesis, water balance, and nutrient uptake ([Rizwan et al., 2016](#)). In agricultural soils, Cd concentrations typically range from 0.01 to 2.7 mg kg⁻¹ under uncontaminated conditions ([Kubier et al., 2019](#); [Huang et al., 2022](#)), while contaminated soils may contain more than 3 mg kg⁻¹, depending on soil type and pollution source ([Kubier et al., 2019](#)). Cd toxicity symptoms in plants generally occur when Cd concentrations in plant tissues exceed 3-30 mg kg⁻¹ ([Ismael et al., 2018](#); [Haider et al., 2021](#)). It also competes with essential nutrients such as zinc (Zn), iron (Fe), and magnesium (Mg), thereby reducing their uptake and translocation within the plant ([Khaliq et al., 2019](#)). Moreover, Cd can move through soil, water, and air, eventually entering the food chain and accumulating in edible plant parts such as grains ([Robson et al., 2014](#)). To counter these effects, several studies have shown that applying mineral fertilizers containing micronutrients such as Zn, Fe, Mn, silicon, and selenium can reduce Cd uptake by plants through competitive absorption mechanisms ([Hussain et al., 2021](#)). In recent years, increasing attention has been directed toward organic fertilizers due to their environmental advantages over chemical alternatives. Organic fertilizers support soil health, reduce pollution, and enhance plant growth ([Khaitov et al., 2019](#); [Alam et al., 2020](#)). Among these, VC, a stabilized organic amendment produced through the decomposition of organic waste by earthworms particularly valued in organic farming. It improves soil structure by increasing porosity and aeration, enhancing water retention, and fostering high levels of microbial activity ([Garg and Gupta, 2009](#); [Demir et al., 2010](#); [Boran, 2015](#)). In arid and semi-arid regions, such as Türkiye, the scarcity of soil organic matter poses a major challenge to sustainable agriculture. As emphasized by [Yıldız \(2012\)](#), increasing organic matter levels in such soils is critical for long-term productivity. VC has been shown in numerous studies to improve crop growth and quality. For example, [Adiloğlu et al. \(2015\)](#) reported that higher VC doses significantly increased lettuce yield, plant diameter, leaf size, and fresh weight, while nutrient content remained largely unchanged, except for increases in Fe and Mn. Similarly, [Büyüklilil and Adiloğlu \(2016\)](#) found that VC reduced Fe, Zn, and B concentrations in sunflower but enhanced the uptake of N, P, K, Mg, Ca, Cu, and Mn. In another study, [Adiloğlu et al. \(2017\)](#) observed that increasing VC doses significantly decreased the concentrations of heavy metals such as Cr, Co, Cd, Ni, and Pb in cucumber plants. These findings collectively suggest that VC not only enhances plant quality but also mitigates heavy metal toxicity. Other organic amendments, such as biochar, have also been shown to significantly influence plant responses under heavy metal stress. For instance, under Cd stress conditions, [Demirbaş and Coşkan \(2019\)](#) reported that biochar application increased maize yield. Likewise, [Özkan et al. \(2016\)](#) demonstrated that VC improved several soil

properties, including pH and available phosphorus. The mobility and bioavailability of heavy metals in soil can be influenced by various factors associated with organic fertilizers, such as their solubility, salt content, pH, and the redox potential of the soil ([Walker et al., 2004](#); [Angelova et al., 2010](#)). Considering these findings, the present study aims to investigate the effects of increasing VC dosages on the growth and developmental parameters of radish (*Raphanus sativus*) cultivated in soil artificially contaminated with Cd.

Materials and Methods

The experiment was conducted in the greenhouse facilities of the Department of Soil Science and Plant Nutrition, Faculty of Agriculture, Selçuk University, using 3-kg capacity plastic pots arranged in a completely randomized design (CRD) with four replications. The physical and chemical properties of the soil are provided in Table 1, while the chemical characteristics of the VC are presented in Table 2. A total of 48 pots were used in a factorial design consisting of three Cd levels (0, 5, and 10 mg kg⁻¹) and four VC application rates (0%, 1%, 2%, and 4%). The factorial arrangement and allocation of treatments are illustrated in Figure 1. The VC application rates were expressed as percentages of the dry weight of the potting soil (w/w). For a pot containing 3,000 g of dry soil, the corresponding amounts of vermicompost added were 0 g (0%), 30 g (1%), 60 g (2%), and 120 g (4%). Vermicompost was thoroughly mixed with the soil before filling the pots to ensure uniform distribution. These application rates were selected based on previous greenhouse experiments and agronomic recommendations, which reported beneficial effects of vermicompost at 1-5% (w/w) on soil physicochemical properties and plant growth ([Arancon et al., 2019](#); [Juleel et al., 2023](#); [Demir and Kiran, 2019](#); [Kiran, 2019](#)). The common nutrient content in vermicompost is organic carbon 27.20%, N 1.83%, K 0.8%, Ca 5.70%, and other micronutrients in small amounts. Cadmium was applied in dry form as CdCl₂ and thoroughly mixed into the soil. The radish cultivar 'Cherry Belle' was used as the test plant. To meet the plants' nutritional requirements, a uniform application of base fertilizers was incorporated into the soil before sowing. Nitrogen was added as urea (46% N) at a rate of 100 mg kg⁻¹ (300 mg pot⁻¹); phosphorus was supplied using triple superphosphate (43% P₂O₅) at 50 mg kg⁻¹ (150 mg pot⁻¹); potassium was added as potassium sulfate (51% K₂O) at 100 mg kg⁻¹ (300 mg pot⁻¹); and magnesium was provided as magnesium sulfate (16% MgO) at the same rate. Micronutrients were also added as follows: boron at 2 mg kg⁻¹ (6 mg pot⁻¹) from Etidot-67 (20.8% B), zinc at 2 mg kg⁻¹ (6 mg pot⁻¹) from zinc sulfate (23% Zn), and iron at 5 mg kg⁻¹ (15 mg pot⁻¹) from iron sulfate (19% Fe).

Table 1. Some physical and chemical analysis results of the soil sample were used in the experiment

Parameters	Results	Method
Texture	Clay loam	Bouyoucos 1951
pH [1:2.5 soil: distilled water]	7.75	Richards 1954
EC [1:5 soil: distilled water, $\mu\text{S cm}^{-1}$]	130	U.S. Salinity Lab. Staff 1954
Lime (CaC	30	Hızalan and Ünal 1966
Organic matter	1.45	Smith and Weldon, 1941
N ($\text{NH}_4\text{-N}+\text{NO}_3\text{-N}$)	17	Bremner 1965
P (Available)	9.6	Olsen et al. 1954
K (Exchangeable)	236	FAO, 1990
Ca (Exchangeable)	7400	
Mg (Exchangeable)	168	
Fe (Available)	1.78	
Cu (Available)	0.38	Lindsay and Norvell 1978
Mn (Available)	6.6	
Zn (Available)	0.16	
Cd (Available)	0.03	
B (Available)	1.3	Richards 1954

Table 2. Some chemical properties of the vermicompost used in the experiment

Characteristics	Value	Characteristics	Value
K (Exchangeable)	0.8	pH	7.92
Ca (Exchangeable)	5.7	EC (1:5) dS/m	2.95
Mg (Exchangeable)	0.7	Organic Matter	44.62
Fe (Total)	0.84	C (Total organic carbon)	27.2
Zn (Total)	84	C/N	14.86
Mn (Total)	430	N (Total)	1.83

Radish plants were harvested 45 days after sowing, or upon reaching full maturity, and key growth parameters were measured immediately after harvest. These parameters included shoot length, shoot fresh and dry weights, tuber diameter, and tuber fresh and dry weights. During hand harvesting, each plant was carefully uprooted to minimize damage to the shoots and tubers. Following harvest, plants were gently washed with distilled water to remove adhering soil

particles. Shoot length was measured using a standard ruler for morphological evaluation. Fresh weights of shoots and tubers were determined using a precision digital scale. To assess dry biomass, the samples were oven-dried at 65 °C until a constant weight was achieved. Tuber diameter was measured using a digital caliper for accuracy.

Statistical analyses were conducted using Minitab 19 software. Analysis of variance (ANOVA) was performed to evaluate the effects of treatments, and

Control	Control	Control	Control
Cd-0+VC-1	Cd-0+VC-1	Cd-0+VC-1	Cd-0+VC-1
Cd-5+VC-1	Cd-5+VC-1	Cd-5+VC-1	Cd-5+VC-1
Cd-10+VC-1	Cd-10+VC-1	Cd-10+VC-1	Cd-10+VC-1
Cd-0+VC-2	Cd-0+VC-2	Cd-0+VC-2	Cd-0+VC-2
Cd-5+VC-2	Cd-5+VC-2	Cd-5+VC-2	Cd-5+VC-2
Cd-10+VC-2	Cd-10+VC-2	Cd-10+VC-2	Cd-10+VC-2
Cd-0+VC-4	Cd-0+VC-4	Cd-0+VC-4	Cd-0+VC-4
Cd-5+VC-4	Cd-5+VC-4	Cd-5+VC-4	Cd-5+VC-4
Cd-10+VC-4	Cd-10+VC-4	Cd-10+VC-4	Cd-10+VC-4
Cd-5+VC-0	Cd-5+VC-0	Cd-5+VC-0	Cd-5+VC-0
Cd-10+VC-0	Cd-10+VC-0	Cd-10+VC-0	Cd-10+VC-0

Figure 1. Layout of the experimental design, showing the factorial arrangement of three Cd levels (Cd-0, Cd-5, and Cd-10 mg kg⁻¹) and four vermicompost (VC) application rates (VC-0%, VC-1%, VC-2%, and VC-4%) across 48 pots. The control treatment is Cd-0 + VC-0%.

mean comparisons were carried out using Tukey's test at a significance level of $p \leq 0.01$ when significant differences were detected.

Results and Discussion

The results demonstrated that increasing VC doses had a statistically significant effect ($p \leq 0.01$) on specific growth parameters of radish plants cultivated in Cd-contaminated soil. Notably, the interaction between VC and Cd treatments was significant for shoot length. The greatest shoot length (25.50 cm) was observed in plants treated with 2% and 4% VC without Cd contamination (Figure 2), whereas the shortest shoot length (19.25 cm) occurred in plants exposed to 10 mg Cd kg⁻¹ without any VC application. These findings are consistent with previous research. For instance, [Abdoosi \(2019\)](#) reported that the VC application mitigated the negative impact of Cd on spinach growth, particularly plant height. Similarly, [Syed et al. \(2022\)](#) observed that VC significantly enhanced plant height when applied in combination

with NPK fertilizers and heavy metals. In their study, the tallest spinach plants (23.10 cm) were recorded under the treatment of 5 t ha⁻¹ vermicompost + N25P8K10 + Pb4, while the shortest (4.10 cm) was observed under Cd treatment alone. Both Cd and Pb independently suppressed plant growth to comparable extents. Furthermore, [Chen and Lai \(2025\)](#) showed that soil amendments such as sulfur-enriched VC improved organic matter content and reduced the bioavailability of Cd. The application of such amendments shifted Cd from highly mobile forms (acid-soluble and exchangeable fractions) to less mobile, oxidizable fractions, thereby limiting its mobility and accumulation. This transformation supported enhanced plant growth, as demonstrated in lettuce. The application of 4% VC combined with 10 mg Cd kg⁻¹ soil resulted in the highest shoot fresh weight (33.03 g), indicating it as the most effective treatment. Similarly, 4% VC with 5 mg Cd kg⁻¹ also significantly increased shoot fresh weight (Figure 3). Organic amendments like VC reduced Cd mobility within the cytoplasm and organelles, while enhancing its sequestration in the cell

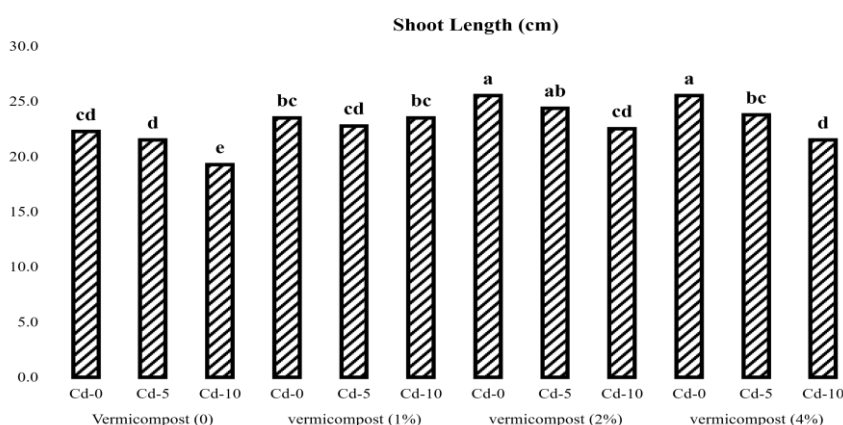


Figure 2. Effects of increasing doses of VC on the shoot length of radish plants grown in Cd-contaminated soil. Different letters above the bars indicate statistically significant differences between treatments according to Tukey's test at $p \leq 0.01$. Values sharing the same letter are not significantly different

wall and vacuoles. This effect was particularly noted by [Chen and Lai \(2025\)](#), who reported that VC combined with organic fertilizers from chicken manure increased sulfur and thiol compound concentrations in lettuce.

The lowest shoot fresh weight (19.83 g) was observed in radish plants grown in soils treated with 5 and 10 mg Cd kg⁻¹ without any VC application (Figure 3), indicating that the absence of VC under Cd stress negatively impacted plant growth. Conversely, increasing doses of VC had a statistically significant positive effect ($p \leq 0.01$) on shoot biomass in radish plants cultivated in Cd-contaminated soil. [Mojdehi et al. \(2022\)](#) reported that elevated heavy metal concentrations in the growth medium increased superoxide dismutase (SOD) activity by 30–50% in ornamental sunflowers, indicating oxidative stress.

However, the application of VC effectively reduced metal accumulation in soil, tubers, and stems, thereby mitigating stress-related effects. These findings suggest that VC can alleviate heavy metal toxicity, and ornamental sunflowers may also serve as potential phytoremediators in Pb- and Cd-contaminated soils.

Regarding shoot dry weight, the highest values were recorded in plants treated with 4% VC + 5 mg Cd kg⁻¹ (2.31 g), 2% VC without Cd (2.41 g), and 4% VC + 10 mg Cd kg⁻¹ (2.42 g). The lowest shoot dry weight (1.82 g) was observed in plants grown without any VC application (Figure 4). Similarly, increasing doses of VC significantly influenced radish tuber diameter in Cd-contaminated soils. The largest average tuber diameter (26.64 mm) was obtained from plants treated with the highest VC dose (4%) across different Cd levels. In

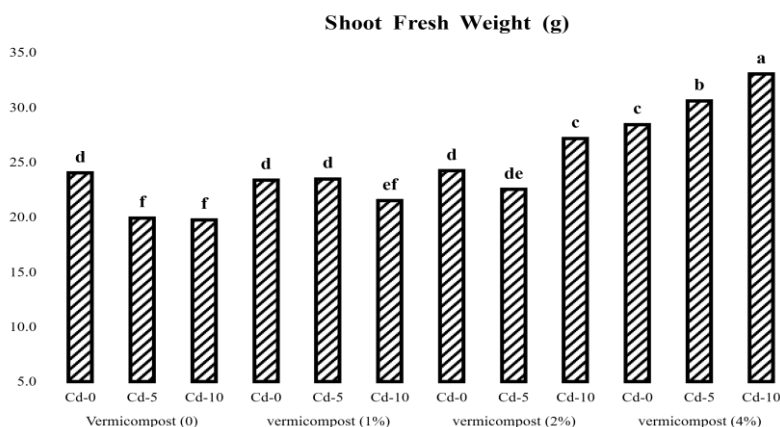


Figure 3. Effects of increasing doses of VC applications on shoot fresh weight of radish plants grown in Cd-contaminated soil. Different letters above the bars indicate statistically significant differences between treatments according to Tukey's test at $p \leq 0.01$. Values sharing the same letter are not significantly different

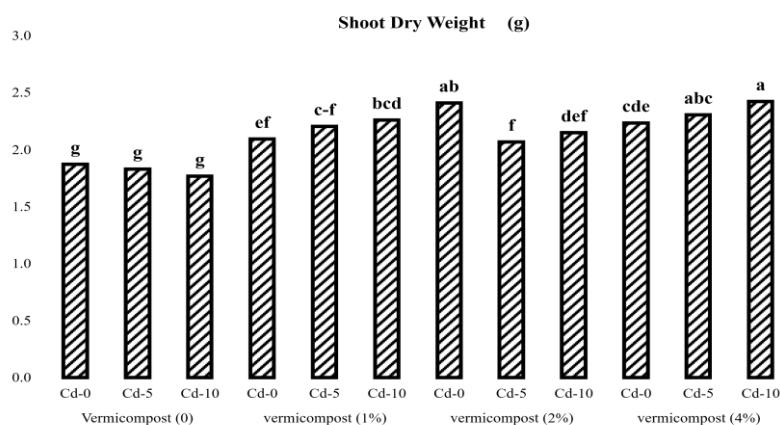


Figure 4. Effects of increasing VC doses on shoot dry weight of radish plants grown in Cd-contaminated soil. Different letters above the bars indicate statistically significant differences between treatments according to Tukey's test at $p \leq 0.01$. Values sharing the same letter are not significantly different

contrast, the smallest diameter (16.40 mm) was recorded in plants exposed to 10 mg Cd kg⁻¹ without VC supplementation (Figure 5). Supporting these findings, [Al Mamun et al. \(2021\)](#) conducted pot experiments on Cd-contaminated soil (0.8 mg kg⁻¹ Cd) using varying doses of VC and biochar. Their study showed that the combined application of 5 t ha⁻¹ VC and biochar reduced Cd concentration in red amaranth (*Amaranthus cruentus*) by 72% and significantly enhanced biomass production.

Similarly, [Yen et al. \(2021\)](#) reported significant increases in the fresh weight of lettuce and pak choi (ranging from 3.9 to 14.4 g per plant) following VC application, compared to controls (0.4-2.5 g per plant). This improvement was attributed to enhanced organic matter content and increased availability of phosphorus, as well as exchangeable magnesium and potassium. Moreover, VC treatment reduced Cd accumulation in plant tissues by 60-75%. In another study, [Abdoosi \(2019\)](#) observed that Cd exposure significantly decreased the dry weight of spinach, whereas VC application notably improved it, with a 49.4% increase recorded upon the incorporation of

10% VC into the soil relative to the control.

In the current study, increasing VC doses significantly influenced the tuber fresh weight of radish plants grown in Cd-contaminated soils. The highest tuber fresh weights-29.35 g and 30.03 g-were recorded in plants treated with 4% VC combined with 5 and 10 mg Cd kg⁻¹, respectively. In contrast, the lowest tuber fresh weight (16.90 g) was observed in control plants that received neither VC nor Cd treatment (Figure 6). These findings suggest that VC applications can enhance tuber biomass even under heavy metal stress, likely due to improved soil structure and nutrient availability. The increased availability of essential minerals-particularly phosphorus, which supports root development and nutrient uptake-is considered a key factor in the observed root weight improvement ([Ebrahimi et al., 2021](#); [Khosropour et al., 2022](#)).

The results of the study indicate that increasing VC doses significantly affected the tuber dry weight of radish plants cultivated in Cd-contaminated soils. The highest average tuber dry weight (1.99 g) was recorded in plants treated with 4% VC, followed closely by those grown in soil without VC but exposed to 10 mg Cd kg⁻¹

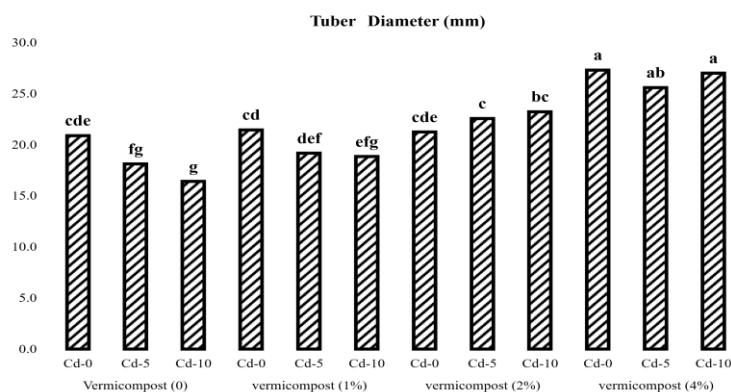


Figure 5. Effects of increasing doses of VC applications on the tuber diameter of radish plants grown in Cd-contaminated soil. Different letters above the bars indicate statistically significant differences between treatments according to Tukey's test at $p \leq 0.01$. Values sharing the same letter are not significantly different

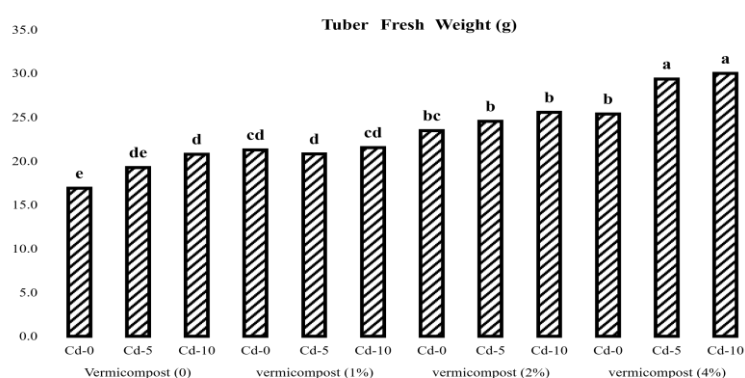


Figure 6. Effects of increasing doses of VC applications on the tuber fresh weight of radish plants grown in Cd-contaminated soil. Different letters above the bars indicate statistically significant differences between treatments according to Tukey's test at $p \leq 0.01$. Values sharing the same letter are not significantly different

(1.91 g). In contrast, the lowest tuber dry weight (1.26 g) was observed in plants receiving 1% VC in combination with 5 mg Cd kg⁻¹, with similarly low values detected in treatments lacking both VC and Cd. These findings highlight the importance of sufficient VC application in enhancing tuber development and mitigating the adverse effects of Cd stress (Figure 7).

Based on Table 3 and the analysis of production components, the results revealed several positive correlations among the measured parameters. Plant height was positively correlated with shoot dry weight and tuber diameter. Fresh plant weight showed a strong positive relationship with shoot dry weight, tuber diameter, and tuber fresh weight. Additionally, shoot dry weight was positively associated with both tuber diameter and tuber fresh weight. Tuber diameter also correlated positively with tuber fresh and dry weights, and a similar positive correlation was observed between tuber fresh and dry weights.

The application of VC to Cd-contaminated soils mitigates the adverse effects of Cd by reducing its toxicity and enhancing soil health. This positive impact is largely due to the chemical and biological composition of VC, which improves soil structure,

fertility, and microbial activity. Numerous studies have confirmed that VC enhances the biological, chemical, and physical properties of soil, thereby supporting the growth of healthy, high-yielding plants (Arancon et al., 2003; Jat and Ahlawat, 2006; Alam et al., 2007; Ali et al., 2007; Singh et al., 2008; Rangarajan et al., 2008). For instance, Ethur et al. (2021) observed that VC application improved shoot development and fruit yield in Cd-contaminated pepper plants, with optimal results at a 50% VC rate. Similarly, Wang et al. (2021) reported increased lettuce biomass and reduced Cd accumulation following the use of a VC-based composite amendment (95% VC + 5% modified shell powder). Iqbal et al. (2024) found that VC application enhanced the grain yield of fragrant rice under Cd stress, improved soil fertility, and enriched fungal diversity, while also reducing Cd uptake in grains, shoots, and tubers. In another study, Wu et al. (2025) demonstrated that VC improved celery biomass in Cd-contaminated field conditions by lowering soil Cd availability and promoting bacterial communities capable of immobilizing Cd. Overall, VC contributes to sustainable agriculture by enhancing soil fertility, retaining moisture, supplying essential nutrients, and

Table 3. Correlation matrix of growth parameters in radish

	Shoot Length	Shoot Weight	Fresh Shoot Weight	Dry Tuber Diameter	Tuber weight	fresh
Shoot Fresh Weight	0.23					
Shoot Dry Weight	0.58**	0.66**				
Tuber Diameter	0.46**	0.89**	0.61**			
Tuber fresh weight	0.24	0.80**	0.67**	0.77**		
Tuber dry weight	-0.08	0.34*	0.10	0.38**	0.48**	

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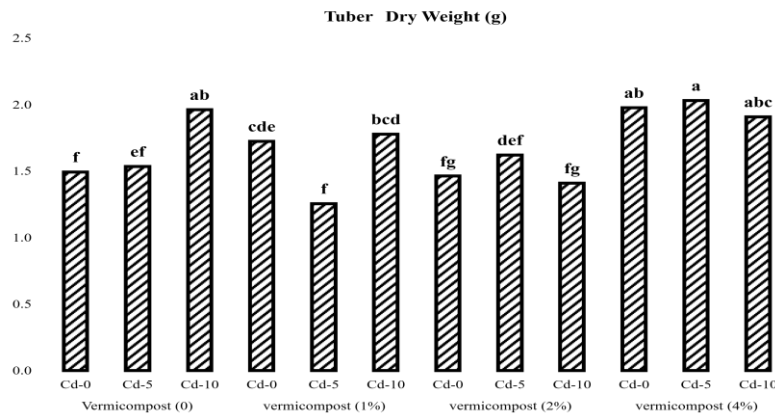


Figure 7. Effects of increasing doses of VC applications on the dry weight of radish tuber grown in Cd-contaminated soil. Different letters above the bars indicate statistically significant differences between treatments according to Tukey's test at $p \leq 0.01$. Values sharing the same letter are not significantly different

reducing environmental pollution. It has been shown to stimulate growth, flowering, and productivity in a wide range of vegetable crops (Oyege and Balaji Bhaskar, 2023; Mohite, 2024; Muslim, 2025; Sonone, 2025).

Conclusions

The application of VC significantly mitigated the adverse effects of Cd contamination on the growth of radish (*Raphanus sativus*). Under non-contaminated conditions, the highest VC dose (4%) resulted in substantial improvements: shoot fresh weight increased by 44.2%, shoot dry weight by 22.2%, tuber diameter by 66.5%, and tuber fresh weight by 22.1% compared to the most stressed treatment (0% VC + 10 mg Cd kg⁻¹). These enhancements are attributed to improved soil biological activity, nutrient availability, and physical structure induced by VC application. However, under high Cd stress (10 mg Cd kg⁻¹), the 4% VC treatment led to a slight decrease (5.0%) in tuber dry weight, suggesting that the effectiveness of VC may

vary depending on the growth parameter and severity of contamination. Overall, the results highlight VC as a promising and environmentally sustainable soil amendment for improving plant growth in Cd-contaminated soils. Further studies across different environmental conditions and crop species are recommended to validate and extend these findings.

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Author Contributions

F.U., M.N., and U.Ç.K.: conceptualized and designed the study. F.U., M.N., Ö.F.Ö., O.A.H.C.H., İ.G., and S.U.: conducted the experiments and performed data analysis. F.U., M.N., and U.Ç.K.: drafted the manuscript, carried out the statistical analysis, and finalized the manuscript for submission.

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relationships that could have influenced the work reported in this paper.



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Detailed soil mapping and classification in semi-humid regions: A focus on Kahramanmaraş-Çağlayancerit

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Soil series

Abstract

Soil classification studies are an integral part of soil survey and mapping activities. This study examines into soil classification within the context of soil survey and mapping efforts, providing crucial insights into the physical and chemical characteristics of soils in the area of Boylu village of Çağlayancerit district in Kahramanmaraş. The research area covers approximately 761 ha. The research identified eleven distinct soil series in the region. Based on soil taxonomy, five series are classified as Entisols, three as Vertisols, two as Inceptisols, and one as Alfisols. In addition, according to the World Reference Base for Soil Resources (WRB) classification system, the series comprises three Vertisols, two Calcisols, two Regosols, and two Cambisols, along with one Luvisol and one Fluvisol. The most extensive soil series in the area was Çamlık, encompassing 14.37% of the total study area. Following this series were the Bölükkamalak (10.25%), Merkyazısı (10.22%), and Körkuyu (9.96%) series. The smallest represented series was also Boylu, accounting for 2.12% of the area.

Introduction

Soil is a precious natural resource that takes thousands of years to form. Its conservation is crucial for future generations because it serves as the foundation that provides essential sustenance and habitat for living organisms (Soil Survey Staff, 2022). Recent scientific research indicates that soil faces threats from pollution (Varol, 2020; Akbay et al., 2023; Yılmaz, 2023), erosion (Aytop and Pinar, 2024), and overuse (Bhattacharyya et al., 2023) particularly when land is exploited beyond its ecological capacity. For instance, transforming productive agricultural land into industrial or residential areas results in a significant loss of high-quality soil. To address these issues, it is

important to understand land characteristics, by implementing classification systems based on detailed soil surveys and mapping studies, which are vital for sustainable land management (Dengiz, 2011; Şenol et al., 2015). Soil survey and mapping are the methods used to classify soils and determine their characteristics. As a result of these studies, detailed soil maps are produced (Soil Survey Staff, 2022).

Soil maps are generated through comprehensive soil survey and mapping research studies, forming an essential foundation for scientific analyses in soil science. Soil maps offer vital insights into land characteristics such as soil depth, stoniness, slope,

salinity, and texture. With advancements in modern soil science, the data depicted in these maps has become increasingly detailed. Throughout history and today, soil maps have played a crucial role across various sectors, including agriculture ([Aytop and Şenol, 2022](#); [Saygın and Dengiz, 2023](#); [Gozukara et al., 2024](#)), industry ([Chumaidiyah et al., 2023](#)), environmental regulation ([AbdelRahman et al., 2022](#)), taxation ([Weiers and Reid, 1974](#)), and military planning ([Rose and Clatworthy, 2024](#)).

Soil maps serve as essential tools in agricultural sciences, offering crucial data that sustain land evaluation studies. The outcomes of such evaluations play a significant role in informing the development of agricultural land use planning, thereby mitigating the risk of overexploiting land resources and ensuring sustainable management ([Aytop and Şenol, 2022](#)).

The soil survey and mapping project encompasses various phases, including office works, field investigations, laboratory analyses, and soil classification processes ([Şenol et al., 2015](#)). Soil classification entails a comprehensive assessment of the physical, chemical, biological, and morphological properties of soils to systematically categorize them. These classification frameworks are instrumental in

producing detailed soil maps ([Soil Survey Staff, 2022](#)). Numerous nations have developed their own soil classification systems; notably, the Soil Taxonomy developed by the United States Department of Agriculture ([Soil Survey Staff, 2022](#)) and the World Reference Base (WRB) established by the Food and Agriculture Organization of the United Nations (FAO) are among the most widely adopted systems.

During the soil survey and mapping process, the most useful tool is Geographic Information System (GIS). GIS are extensively utilized in soil survey and mapping endeavors within the academic sphere ([Saleh et al., 2023](#)). These systems markedly enhance the digitization of various cartographic elements, including slope, elevation, and topography, thereby supporting rigorous scientific analysis ([Saygın and Dengiz, 2023](#)). Furthermore, GIS technologies are instrumental in precisely delineating the boundaries of soil series and mapping units, contributing to the advancement of soil science research ([Aytop and Şenol, 2022](#)).

The current study aims to characterize the fundamental physical, chemical, and morphological properties of soils within an approximately 761-hectare agricultural area situated in Boylu village, Çağlayancerit district, Kahramanmaraş Province, located at semi-

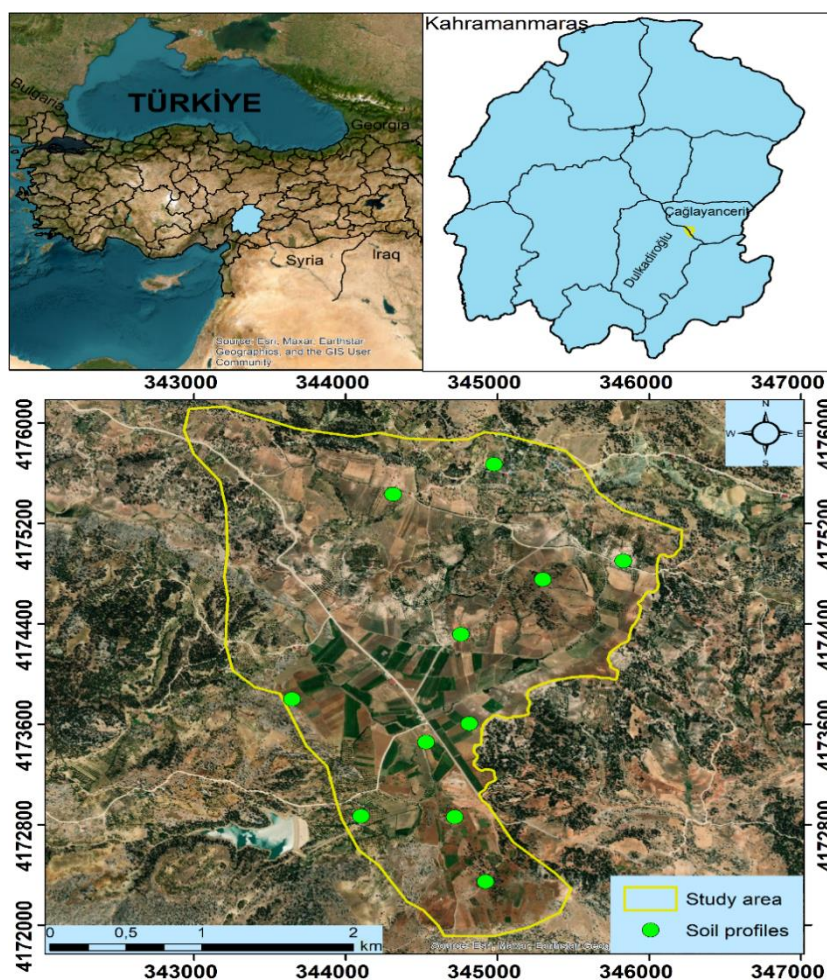


Figure 1. Spatial distribution map of the study area showing its geographic location and the defined soil profile sites

humid environmental region, and to classify these soils utilizing GIS.

Materials and Methods

Materials

The study area is situated between 37°41' and

37°43' North latitudes and 37°12' and 37°16' East longitudes, within the district of Çağlayancerit in Kahramanmaraş Province. The southwestern part of the study area is bordered by Dulkadiroğlu District. The area of the study area is 760.57 ha.

The Çağlayancerit district exhibits a climate characterized by high temperatures and aridity during the summer months, accompanied by severe cold and

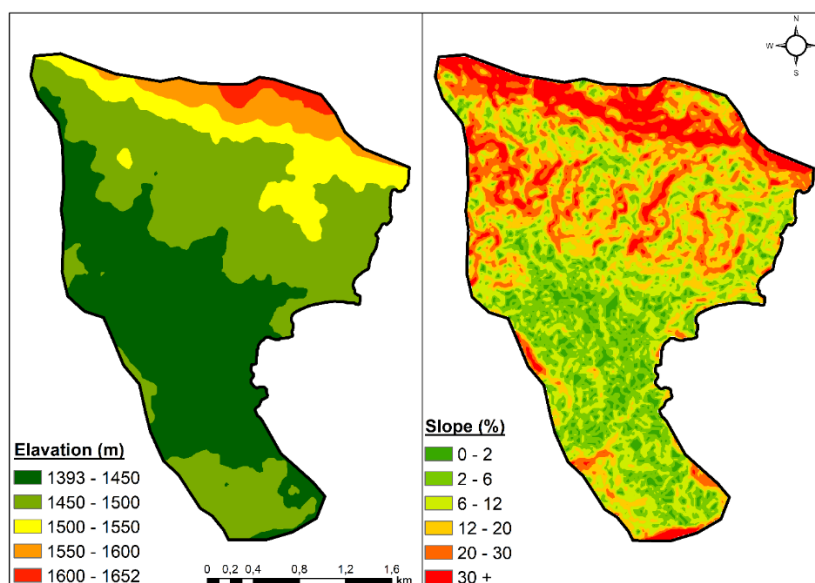


Figure 2. Slope and elevation map of the study area

substantial rainfall in winter. The region's mean annual rainfall is recorded at 744.1 mm, with an average annual temperature of 13.6 °C. In the summer, temperatures can surpass 40°C (Anonymous, 2019). the soil temperature and moisture regimes were classified as mesic and xeric. The research site's elevation varies from 1393 to 1652 metres above sea level (Figure 2). The Boylu neighborhood, located in the northern part of the region, is situated at a higher elevation. Conversely, the southern areas have an average elevation of approximately 1400 m. Consistent with the elevation data, the slope map indicates that northern areas are characterized by steeper slopes, with a gradual decrease in slope steepness toward the south (Figure 2). Annual crops are cultivated in the central zones where the terrain is flat, whereas walnut trees are grown on steeper slopes at higher elevations. Additionally, there are forested areas in the southern part of the study area (Figure 1).

Methods

First, preliminary information about the study area was collected, including the acquisition of a DEM map (Anonymous, 2025), a geological map, Google Earth imagery, and topographic maps. Temporary soil boundaries within the study area were delineated using Google Earth imagery from various years and supplementary sources. Subsequently, location points for profile pits were identified and examined in the field, with soil samples collected from the profiles based on horizon principles. The profile pits were excavated using an excavator. The morphological properties of soils were identified based on Soil Survey Staff (2022). A 10% HCl solution, a tape measure, and the Munsell colour chart were employed to assess the morphological characteristics of the soils (Dinc and Senol, 2001). Following laboratory analyses, the second field survey was conducted, and the soil boundaries were finalized. While establishing soil boundaries, terrain features—including stoniness, color, texture, lime content, and slope—were carefully considered.

Table 1. Results of morphological, physical analyses and chemical of the soil series

Horizon	Depth (cm)	Soil Colour (Dry-moist)	CaCO ₃ (%)	Texture			Class
				Clay (%)	Silt (%)	Sand (%)	
Beşenli							
Ap	0-19	5YR 6/6 5YR 4/6	28.33	23.38	52.75	23.87	SiL
A2	19-50	5YR 5/6 5YR 4/6	27.74	29.71	37.98	32.31	CL
Ck1	50-71	5YR 6/4 5YR 4/4	32.51	20.09	51.99	27.92	SiL
Ck2	71-100	5YR 7/4 5YR 4/4	36.76	19.16	54.86	25.98	SiL
Boylu							
Ap	0-30	10YR 5/4 10YR 4/4	0.82	46.59	27.43	25.98	C
ACss	30-60	10YR 5/6 10YR 4/6	0.86	55.03	23.21	21.76	C
C1ss	60-80	10YR 6/6 10YR 4/6	0.15	50.81	23.21	25.98	C
C2	80-102	7.5 YR 7/4 7.5YR 6/4	1.28	40.30	10.93	48.77	SC
2C	102-112	7.5YR 6/4 7.5YR 5/6	3.50	23.38	32.71	43.91	L
Bölükkamalak							
Ap	0-19	10YR 7/2 10YR 5/3	24.24	23.38	37.98	38.64	L
A2	19-32	10YR 7/2 10YR 4/3	31.26	25.49	43.26	31.25	L
C	32-74	10YR 7/1 10YR 7/2	36.34	19.16	44.31	36.53	L
Crk	74-150	10YR 7/1 10YR 7/2	38.73	14.94	37.98	47.08	L
Elmacıkderesi							
Ap	0-30	10YR 5/4 10YR 3/4	7.46	27.60	37.98	34.42	CL
A2	30-50	10YR 5/4 10YR 4/4	10.27	30.64	35.11	34.25	CL
2A	50-90	10YR 5/3 10YR 3/2	7.70	36.97	33.00	30.03	CL
2C	90-135	10YR 5/4 10YR 4/4	19.73	33.93	39.04	27.03	CL
Epcingüney							
Ap	0-34	2.5Y 6/3 2.5Y 5/6	33.85	44.48	31.65	23.87	C
A2	34-54	2.5Y 6/2 2.5Y 3/2	31.71	49.46	27.73	22.81	C
C	54-100	2.5Y 7/3 2.5Y 6/6	37.20	47.35	30.89	21.76	C
Hacıağalar							
Ap	0-30	7.5YR 4/4 7.5YR 3/4	0.75	51.57	26.67	21.76	C
A2ss	30-67	7.5YR 5/6 7.5YR 4/6	0.97	68.45	18.23	13.32	C
A3ss	67-93	7.5YR 6/4 7.5YR 5/6	0.97	68.45	16.12	15.43	C
C1	93-114	7.5YR 6/5 7.5YR 6/8	0.82	51.57	18.23	30.20	C
C2	114-169	7.5YR 6/4 7.5YR 6/8	0.86	55.79	16.12	28.09	C

İnala							
A	0-19	7.5YR 4/4	0.89	32,58	33,00	34,42	CL
		7.5YR 3/4					
Bt1	19-45	5YR 4/6	0.89	68,45	18,23	13,32	C
		2.5YR 3/6					
Bt2	45-78	5YR 4/6	1.04	70,56	18,23	11,20	C
		2.5YR 3/6					
BC	78-103	5YR 4/6	0.98	68,45	16,12	15,43	C
		2.5YR 4/6					
C	103-135	7.5 YR 5/6 (moist)	1.34	72,67	7,68	19,65	C
Körkuyu							
Ap	0-25	10YR 4/6	2.74	36.80	26.67	36.53	CL
		10YR 3/6					
BA	25-42	10YR 5/4	1.78	47.52	26.67	25.81	C
		10YR 3/3					
Bw	42-70/85	10YR 3/4 (moist)	2.23	50.69	25.62	23.70	C
Küllucular							
Ap	0-32	7.5YR 6/6	17.66	26.42	26.67	46.91	SCL
		7.5YR 4/4					
Bw	32-67	7.5 YR 6/4	4.29	47.52	20.34	32.14	C
		7.5YR 3/4					
BC	67-90	7.5 YR 6/3	20.82	49.63	22.45	27.92	C
		7.5YR 4/4					
Ck	90-125	7.5YR 7/3	36.66	39.08	29.84	31.08	CL
		7.5YR 6/6					
Merkyazısı							
Ap	0-22	10YR 4/4	0.89	43.30	24.56	32.14	C
		10YR 3/4					
Ad	22-45	10YR 5/3	0.74	45.41	21.40	33.19	C
		10YR 3/4					
Bss1	45-75	10YR 5/3	0.82	47.52	22.45	30.03	C
		10YR 3/4					
Bss2	75-117	10YR 5/3	0.74	49.63	23.51	26.86	C
		10YR 3/4					
BC	117-137	10 YR 7/4	18.22	32.75	29.84	37.41	CL
		10YR 5/4					
Ck	137-170	7.5YR 7/3	36.58	39.08	21.40	39.52	CL
		7.5YR 6/6					
Çamlık							
A	0-22	10YR 4/2	1.81	20.93	38.74	40.32	SCL
		10YR 2/2					
AC	22-49	10YR 5/4	1.98	18.82	45.07	36.10	SCL
		10YR 4/3					
C1	49-90	10YR 6/4	1.82	25.53	29.92	44.55	SL
		10YR 5/4					
C2	90-116	5YR 5/6	1.20	8.65	21.48	69.87	SL
		(moist)					
2C1	116-141	5YR 5/4	1.82	10.38	17.64	71.98	L
		(moist)					
2C2	141-166	5YR 5/4	1.51	27.26	19.75	52.99	L
		(moist)					
2C3	166-189	5YR 5/8	2.61	21.31	25.70	52.99	L
		(moist)					

Additionally, horizon sequences were examined with a soil auger to verify soil layers, and the precise locations of soil boundaries were delineated. Additionally, the soils were classified according to both Soil Taxonomy and World Reference Base (WRB). The ArcGIS 10.7.1 (GIS) software was employed in the creation and digitization of the maps.

In disturbed soil samples, various analyses were conducted to determine key soil properties. These included texture assessment following [Bouyoucos \(1951\)](#), organic matter content as per [Jackson \(1979\)](#), CaCO_3 content measured with the Scheibler calcimeter according to [Soil Survey Laboratory Staff \(1992\)](#), exchangeable cations and cation exchange capacity (CEC) following [Rhoades \(1982\)](#), as well as soil pH and electrical conductivity (EC) measurements performed on saturation extracts.

Results and Discussion

Eleven soil series have been identified in the study area. These include two marine, four alluvial, two colluvial, one Palaeozoic sediment, one formed on crystalline limestone, and one resulting from mudflow. The Beşenli and Bölükkamalak series are formed on marine parent material. Marine fossils have been found in patches within these soils, suggesting that these areas may have once been shallow sea beds. These soils contain diagnostic horizons with high CaCO_3 content (Ck), as shown in Table 1. In two profiles, the soils are deep, non-saline, and exhibit pH levels exceeding 7.50. The Ap, Ck1, and Ck2 horizon order of the Beşenli series are classified within the silty loam (SiL) texture class, whereas the A2 horizon belongs to the clay loam (CL) class. All horizons of the Bölükkamalak series are categorized under the loamy (L) texture class. The soils of the Beşenli series display a 5YR colour according to the Munsell colour scale, while the horizons of the Bölükkamalak series exhibit a 10YR spectral color (Hue). The base saturation levels of both series exceed 80%. The amount of organic matter in all horizons of these two series is less than 2% and contains low organic matter (Table 2).

The Boylu, Elmacıkderesi, Çamlık and Hacıağalar series were developed on alluvial parent material. The slopes of areas formed on alluvial parent material are flat or nearly flat. The Boylu series includes low calcium carbonate content, with the exception of the 2C horizon, which exceeds 2%. In contrast, the Hacıağalar series is entirely free of CaCO_3 throughout its profile (less than 1%). The Elmacıkderesi series exhibits moderately calcareous horizons with calcium carbonate content below 11%, except for the 2C horizon. Texture analysis indicates that all horizons of the Hacıağalar series are clayey, while those of the Elmacıkderesi series are clay loam. In the Boylu series, textures vary: Ap, ACss, and C1ss horizons are clayey; C2 is sandy clayey; and the 2C horizon is loamy. Soil

colour readings show a consistent 10YR for the Boylu and Elmacıkderesi series, whereas the Hacıağalar series horizons are uniformly 7.5YR (refer to Table 1). The pH levels range from 5.05 to 6.65 in the Hacıağalar series, 6.32 to 7.42 in the Boylu series, and 7.64 to 7.76 in the Elmacıkderesi series, with higher CaCO_3 content likely contributing to the slightly alkaline pH of the latter due to calcium's role in increasing soil pH. Notably, the organic matter in the Ap horizons exceeds two across all series, whereas it is lower in other horizons. Base saturation values are 78-83% for Boylu, 90-95% for Elmacıkderesi, and 58-90% for Hacıağalar. The Hacıağalar series, characterized by consistently clay-textured horizons, exhibits higher CEC values than the other two series. All three soil series are non-saline (see Table 2). The Çamlık series features various textured horizons: the A and AC horizons are sandy clay loam, the C1 and C2 horizons are sandy loam, and the 2C1, 2C2, and 2C3 horizons are loam. Notably, the CaCO_3 content is generally below 2%, except for the 2C3 horizon, which contains 2.61% CaCO_3 . The A horizon of the Çamlık series, located in a forested area, exhibited a notably higher organic matter content at 13.30%, with its sub-horizon, AC, containing 5.11%. Organic matter content tends to decline in sub-horizons. The Çamlık series has demonstrated neutral to slightly acidic pH levels and is non-saline. The base saturation values for horizons within the Çamlık series ranged from 79% to 91% (refer to Table 1;2).

In this research, the parent material of two soil series was classified as colluvial. Colluvial parent material refers to material that has been transported downslope from sloping terrain predominantly through the action of water or wind ([Leopold and Völkel 2007; Kühn, 2025](#)). Epcingüney and Küllucular are soil series formed on colluvial parent material. The Epcingüney series is characterized by uniformly clayey horizons throughout, with a high CaCO_3 content ranging from 31.71% to 37.20%. In contrast, the Küllucular series exhibits variable CaCO_3 content between 4.29% and 36.66% and includes a Ck horizon. The organic matter content in the Küllucular series exceeded 1% in the upper two horizons but fell below 1% in the BC and Ck horizons. In the Epcingüney series, the Ap and A2 horizons contained 2.70% and 2.18% organic matter, respectively, while the C horizon had 0.78%.

The pH levels across all horizons in the Küllucular and Epcingüney series were slightly alkaline. Both series are characterized by non-saline soils. In the Epcingüney series, the Ap, A2, and C horizons had base saturations of 95%, 94%, and 96%, respectively. For the Küllucular series, the Ap horizon's base saturation was 92%, with the Bw at 88%, BC at 90%, and Ck at 97% (Table 2).

The parent material of the Merkyazısı series is mudflow. It has the Ap, Ad, Bss₁, Bss₂, AC, and Ck horizons. The lower two horizons have a silty loam texture, whereas the other horizons are clay-textured.

Table 2. Results of some chemical analyses of the series

Horizon	Organic Matter (%)	pH	EC (dS/m)	Base Saturation (%)	CEC (me/100g)	Exchangeable cations (me/100g)			
						Ca ⁺⁺	Mg ⁺⁺	K ⁺	Na ⁺
Beşenli									
Ap	1.18	7.50	0.68	94	31.18	17.05	7.47	4.01	0.92
A2	1.01	7.66	0.69	99	31.24	19.60	6.06	4.64	0.79
Ck1	0.29	7.69	0.62	96	29.10	17.85	5.30	3.91	0.92
Ck2	0.45	7.83	0.46	99	26.79	15.91	5.13	4.40	1.20
Boylu									
Ap	2.49	6.93	0.75	81	29.67	13.61	4.42	4.47	1.52
ACss	1.03	6.56	0.68	78	33.85	14.32	5.66	4.74	1.70
C1ss	0.85	6.32	0.70	78	34.07	15.30	6.97	3.34	1.13
C2	0.54	6.92	0.73	82	32.17	13.79	6.06	4.79	1.72
2C	0.51	7.42	1.00	83	39.09	19.19	5.76	6.15	1.46
Bölükkamalak									
Ap	1.82	7.53	0.45	93	30.30	15.73	6.01	5.42	1.13
A2	1.79	7.65	0.52	88	32.66	16.77	6.46	4.65	0.87
C	1.06	7.90	0.47	83	29.33	15.96	4.44	2.89	0.93
Crk	0.39	8.01	0.44	98	26.93	17.05	5.66	3.00	0.80
Elmacıkderesi									
Ap	2.32	7.70	0.72	90	22.95	11.06	5.15	3.94	0.56
A2	1.26	7.72	0.67	91	33.56	14.84	8.66	6.22	0.98
2A	1.99	7.64	0.60	93	30.74	14.93	6.85	5.62	1.22
2C	1.15	7.76	0.58	95	38.10	17.41	9.84	7.67	1.13
Epcingüney									
Ap	2.70	7.59	0.71	95	34.02	16.92	8.59	5.49	1.26
A2	2.18	7.63	0.85	94	33.00	17.58	7.47	4.65	1.32
C	0.78	7.86	0.46	96	46.44	27.39	10.64	4.89	1.51
Hacıağalar									
Ap	2.46	6.65	0.60	60	43.48	14.55	5.05	5.39	1.00
A2ss	0.79	5.70	0.41	58	58.74	19.60	7.27	5.74	1.41
A3ss	0.66	5.14	0.59	77	49.07	18.59	9.49	7.52	2.05
C1	0.32	5.05	0.47	90	38.67	17.58	7.27	7.67	2.18
C2	0.75	5.69	0.40	83	33.75	12.73	7.27	5.98	1.97
İnala									
A	2.56	6.56	0.41	74	32.06	12.32	6.06	4.52	0.84
Bt1	1.57	6.35	0.30	46	46.68	8.89	6.26	5.48	0.87
Bt2	1.33	5.81	0.21	42	43.55	8.89	4.24	4.13	0.83
BC	1.19	5.57	0.17	58	38.43	8.48	7.27	5.37	1.01
C	0.42	5.20	0.19	84	28.17	10.71	7.07	4.86	1.09
Körkuyu									
Ap	1.36	7.70	0.78	90	37.77	18.99	8.48	5.30	1.07
BA	1.15	7.61	0.76	85	37.33	16.26	8.48	5.88	1.23
Bw	0.98	7.58	0.84	98	36.04	16.57	13.54	4.17	1.07
Küllucular									
Ap	1.50	7.67	0.74	92	43.02	20.70	10.32	7.23	1.14
Bw	1.40	7.55	0.85	88	45.00	20.18	11.10	7.44	0.96
BC	0.52	7.60	0.82	90	38.94	19.09	9.55	5.30	0.93
Ck	0.42	7.73	0.48	97	39.31	20.45	10.45	6.67	0.71
Merkyazısı									
Ap	1.53	6.73	1.31	66	33.92	11.11	5.86	4.18	1.28
Ad	1.61	6.67	0.72	71	33.16	12.32	6.87	3.43	1.05
Bss1	1.03	6.77	0.73	64	40.18	12.73	8.28	3.24	1.32
Bss2	0.96	7.54	0.81	82	41.58	14.89	11.10	5.84	2.23
BC	0.18	7.52	0.73	94	32.50	14.39	8.84	5.07	2.19
Ck	0.21	7.75	0.61	99	33.38	13.64	10.61	6.29	2.67

Çamlık									
A	13.30	6.60	0.75	91	34.29	13.26	10.61	6.45	0.96
AC	5.11	6.56	0.76	83	29.75	9.85	8.79	5.29	0.82
C1	2.60	6.18	0.47	79	38.36	17.55	7.68	2.90	2.19
C2	0.48	6.42	0.63	85	23.12	8.08	6.06	4.16	1.38
2C1	0.35	6.27	0.72	89	27.43	12.37	6.94	3.48	1.50
2C2	0.09	6.28	0.70	87	24.79	11.74	6.57	1.75	1.43
2C3	0.24	7.23	1.00	84	26.35	10.98	6.57	3.18	1.29

The bottom horizon displays a 7.5YR hue, while the others show 10YR. The CaCO_3 content of the upper four horizons is below 1%, with the AC horizon containing 18.22% and the Ck horizon 36.58%. The pH values of the Ap, Ad, and A3ss horizons are 6.73, 6.67, and 6.77, respectively. The pH values of the remaining horizons range from 7.52 to 7.75. Base saturation varies between 64% and 99%.

The Korkuyu series is developed on a parent material of hard crystalline limestone. Although the Ap and Bw horizons exhibit slight calcareous properties,

the BA horizon lacks calcium carbonate entirely, suggesting leaching of calcium carbonate in these soils. The upper horizon possesses a clay loam texture, whereas the underlying horizons are characterized by a clay texture. Soil color is classified as 10YR according to Table 1. Organic matter content across the series is low to very low. The pH levels are slightly alkaline in all horizons, and the soils are non-saline. Base saturation ranges from 85% to 98%. The CEC values for the series range between 36.04 and 37.77 me/100g, as detailed in Table 2.

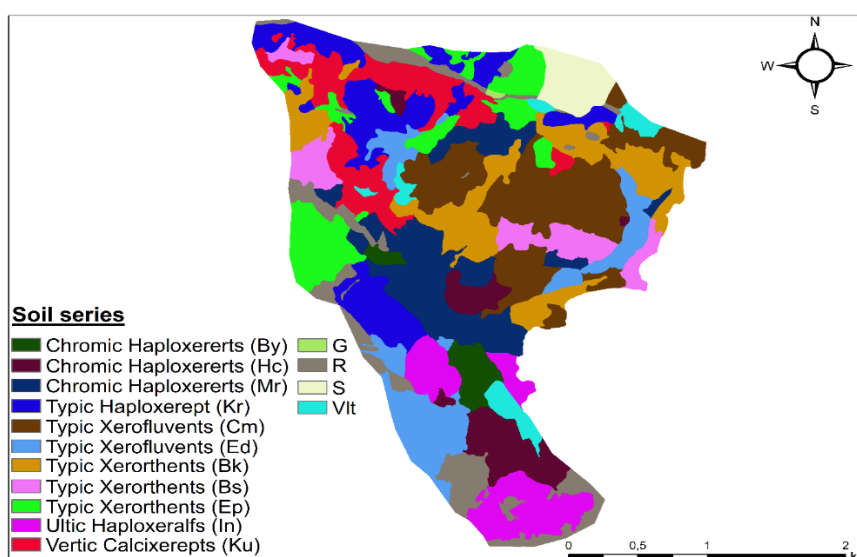


Figure 3. Spatial distribution map of soil series and other land use types

Table 3. Spatial distribution ratios of soil series and other land use types

Soil series and other land use types	Area (ha)	Ratio (%)
Bölükkamalak (Bk)	77.93	10.25
Beşenli (Bs)	36.51	4.80
Boylu (By)	16.13	2.12
Çamlık (Cm)	10.32	14.37
Elmacıkderesi (Ed)	60.68	7.98
Epcingüney (Ep)	58.62	7.71
Hacıağalar (Hc)	40.17	5.28
İnala (In)	50.54	6.64
Korkuyu (Kr)	75.73	9.96
Küllucular (Ku)	61.54	8.09
Merkyazısı (Mr)	77.70	10.22
Rocky (R)	56.68	7.45
Various land types (Vlt)	17.81	2.34
Settlement (S)	20.62	2.71
Graveyard (G)	0.60	0.08
Total	760.57	100.00

The İnala series comprises deep soils developed on Paleozoic sedimentary parent material. These soils possess Bt horizons with clay illuviation. While the Ap horizon has a CL texture, the other horizons have a clay texture. The profile is predominantly characterized by a red hue, and the soils are non-calcareous (see Table 1). The organic matter content is 2.56% in the surface horizon, decreasing progressively toward the lower horizons. The soil series displays a pH range from neutral to strongly acidic, with no evidence of salinity. Base saturation varies between 42% and 84% throughout the soil profile. Notably, the highest cation exchange capacity (CEC) is found in the Bt1 and Bt2 horizons, where clay illuviation is present (refer to Table 2).

Spatial distribution of soil series in the study area

The distribution of the soil series within the study area is shown in Figure 3, and their coverage within the total area is presented in Table 3. The predominant soil series within the study area is Çamlık, encompassing 109.32 hectares. Following in order of area coverage are Bölükkamalak (77.93 ha), Merkyazısı (77.70 ha), Körkuyu (9.96 ha), Küllucular (8.09 ha), Elmacıkderesi (7.98 ha), Epcingüney (7.71 ha), İnala (6.64 ha), Hacıağalar (5.28 ha), Beşenli (4.80 ha), and Boylu (2.12 ha). Additionally, rocky terrains constitute 7.45% of the total landscape. Land types such as built-up areas, settlements, and cemeteries account for 2.34%, 2.71%, and 0.08% of the total area, respectively (Table 3).

Classification of the study area soils

The soils within the study area have been classified following the standards of Soil Taxonomy and

the World Reference Base for Soil Resources (WRB, 2015). Specifically, the Beşenli, Bölükkamalak, Elmacıkderesi, Epcingüney, and Çamlık soil series are categorized under the Entisols in Soil Taxonomy, reflecting their young geological age and absence of B horizons. As a suborder, the Elmacıkderesi and Çamlık series have been classified as Fluvent due to their alluvial parent material, while the other series have been classified as Orthents. The classification is further refined based on soil moisture regime: given the xeric conditions of the study area, the Elmacıkderesi and Çamlık series are classified as Xerofluvents within the great group, while the other series are designated as Xerorthents. The sub-groups of these series were determined as Typic Xerorthents and Typic Xerorthents (Table 4). According to WRB standards, the Beşenli and Bölükkamalak series are classified as Haplic Calcisol, the Elmacıkderesi and Çamlık series as Eutric Fluvisol, and the Epcingüney series as Eutric Regosol (Table 4).

The Körkuyu and Küllucular soil series are categorized as Inceptisols due to the presence of a Bw horizon and observable profile development, distinguishing them from series classified within the Entisols. Additionally, these series are designated as Xerepts as a suborder, reflecting their soil moisture regime. The Küllücüler series has been classified as Calcixerepts because of the presence of the Ck horizon, while the Körkuyu series is classified as Typical Haploxerepts. The appearance of weak, shiny slip surfaces in the subhorizons of the Küllucular series leads to its classification as Vertic Calcixerepts within this subgroup. Conversely, the Körkuyu series remains classified as Typic Haploxerepts. According to WRB, the Körkuyu and Küllucular series were classified as Eutric Cambisol due to the presence of a Cambic horizon (Table 4).

Table 4. Classification of soils in the study area

Soil Series	Soil Taxonomy (2022)				WRB (2015)
	Orders	Suborders	Great Group	Subgroups	
Beşenli	Entisol	Orthent	Xerorthents	Typic Xerorthents	Haplic Calcisol
Bölükkamalak					Haplic Calcisol
Epcingüney					Eutric Regosol
Elmacıkderesi	Entisol	Fluvents	Xerofluvents	Typic Xerofluvents	Eutric Fluvisol
Çamlık					
Körkuyu					
Küllucular	Inceptisol	Xerepts	Haploxerepts	Typic Haploxerepts	Eutric Cambisol
			Calcixerepts	Vertic Calcixerepts	
Boylu					
Hacıağalar	Vertisol	Xererts	Haploxererts	Chromic Haploxererts	Chromic Vertisol
					Chromic Vertisol
Merkyazısı					Calcic Vertisol
İnala	Alfisol	Xeralfs	Haploxeralfs	Ultic Haploxeralfs	Chromic Luvisols

The Boylu, Hacıağalar, and Merkyazısı series are classified within the Vertisols in the taxonomy owing to their high clay content and prominent slickensides. According to the classification system, these series belong to the same class, suborder (Xererts), great group (Haploxererts), and subgroup (Chromic Haploxererts). Under the WRB system, they are classified as Chromic Vertisols, characterized by their vertical structure and reddish colouration (Table 4).

The Inala series is classified within the Alfisols, characterized by the presence of an argillic horizon and a base saturation exceeding 35%. As a suborder, it is designated as Xeraf, reflecting its xeric soil regime. At the great group level, it is classified as Haploxeraf. Specifically, the Inala series is further classified as Ultic Haploxeraf, a subgroup distinguished by a base saturation of less than 75% within the upper 75 cm of the soil profile. This series was classified as Chromic Luvisols according to the WRB (2015) (Table 4).

Conclusions

This study provides an in-depth assessment and classification of soils within the 761-hectare research region, identifying eleven distinct soil series developed on various parent materials, including marine, alluvial, colluvial, limestone, Paleozoic sediments, and mudflow deposits. The findings clearly indicate that parent material and landscape position significantly influence soil morphology, texture, carbonate accumulation, organic matter content, and pH levels. Marine-derived soils exhibit high CaCO_3 contents and low organic matter, whereas alluvial soils display a wide range of textures and higher organic matter concentrations in surface horizons. Colluvial soils are characterized by elevated clay content and increased carbonate levels, with processes of carbonate accumulation and leaching affecting soil fertility and stability. Soils originating from Paleozoic and limestone parent materials show more advanced profile development, including clay illuviation and evidence of carbonate leaching, which are essential for understanding soil resilience and land use suitability. Spatial analysis indicates that the Çamlık series is the most prevalent in the area, while the Boylu series is the least extensive, highlighting the heterogeneous distribution of soil-forming environments. Based on classification, five soil series are categorized as Entisols, three as Vertisols, two as Inceptisols, and one as Alfisols under Soil Taxonomy. Additionally, the World Reference Base (WRB) identifies Calcisols, Regosols, Cambisols, Fluvisols, Vertisols, and a Chromic Luvisol. These results underscore the complexity of pedogenic processes operating under the region's xeric moisture regime, emphasizing the coexistence of both weakly and strongly developed soils within a relatively small spatial extent.

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Author Contributions

H.A.: Soil survey studies, soil sampling, data analysis, interpretation, conceptualisation, investigation, writing, reviewing and editing. **C.H.Y.:** Soil survey studies, soil sampling and writing. **Y.K.K.:** Soil survey studies, soil sampling, writing, reviewing and editing. **R.S.:** Soil survey studies, soil sampling and writing. **S.Ş.:** Soil survey studies, soil sampling, writing, reviewing and editing. **O.D.:** Writing, reviewing and editing.

Conflict of Interest

The authors declare that they have no known conflicts of interest, whether financial, non-financial, professional, or personal, that could influence the work reported in this article.

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Estimation of enteric and manure-derived methane emissions in sheep and goats in Ankara Province

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Abstract

Since livestock account for most of Türkiye's agricultural greenhouse gas emissions, strategies for estimating livestock emissions must be developed. Methane emissions per animal category were estimated in this study using the Intergovernmental Panel on Climate Change (IPCC) Tier 2 method for enteric and manure methane (CH₄) emissions in two sheep breeds (Akkaraman and Anatolian Merino) and three goat breeds (Hairy, Angora, Saanen) raised in Ankara province, Türkiye. Average enteric CH₄ emission factor (EF) was 9.61 kg CH₄ head⁻¹ yr⁻¹, with the highest value of 10.86 kg CH₄ head⁻¹ yr⁻¹ in Hairy goats, and lowest 8.55 kg CH₄ head⁻¹ yr⁻¹ in Anatolian Merino. Estimated gross energy (GE) was highest in Hairy goats (30.10 MJ head⁻¹ day⁻¹), and lowest in Anatolian Merino (21.73 MJ head⁻¹ day⁻¹). Average dry matter intake (DMI) was 1.40 kg DM head⁻¹ day⁻¹. The total average volatile solids (VS) excretion was estimated at 0.95 kg DM head⁻¹ yr⁻¹, and the average methane emission factor (EF) for animal manure was 0.46 kg CH₄ head⁻¹ yr⁻¹. Total mean VS estimate was twice as much as the IPCC default values, and the CH₄ EF for animal manure was higher than the IPCC default values. The current study's estimates will be helpful in the creation of the national methane inventory. In case more detailed data are collected to improve population data, subcategories may be expanded, and subgroups may vary by region.

Introduction

The average temperature on Earth's surface has increased by 0.6 °C since the beginning of the industrial era, and this increase is projected to reach an additional 1.4-5.8 °C by the year 2100 (FAO, 2006). The Kyoto Protocol has set a limit for countries worldwide to reduce their emissions by 8-10% of the 1990

emissions as a milestone (FAO, 2006). One powerful greenhouse gas (GHG) that causes global warming is methane (CH₄). The main causes of CH₄ emissions from animals are enteric fermentation and manure management. Approximately 18% of greenhouse gas emissions in the world are attributed to the livestock

sector ([Steinfeld et al., 2006](#); [EPA, 2023](#)). An estimated 2.2 billion tons of CO₂ equivalent are released by livestock each year, making up over 80% of agricultural CH₄ emissions and 35% of all anthropogenic CH₄ emissions ([Soren et al., 2017](#)). In Türkiye, inventories of methane emissions from enteric fermentation and manure management have been reported by [TÜİK, \(2018\)](#) following the methodology of Intergovernmental Panel on Climate Change (IPCC, 2006). In total, 37% of methane emissions originate from livestock farming ([FAO, 2006](#)). In Türkiye's agricultural sector, methane (CH₄) emissions originating from enteric fermentation account for approximately 48%, while those from manure management contribute 11.2% ([TÜİK, 2018](#)). Hydrogen generation, a crucial step before methane is formed in the rumen, is enhanced by high concentrations of structural carbohydrates. High acetic and butyric acid production increases hydrogen generation, whereas non-structural carbohydrates, such as starch and sugars, reduce hydrogen formation ([Aguilera and Molina-Alcaide, 2021](#)). Including alternative roughages (Blepharis scindica herbage) in the diet of ewes could be a promising approach to reduce enteric methane emissions up to 49.3%, from 14.9 to 6.9 kg CH₄ head⁻¹ yr⁻¹ in Malpura ewes ([Bhatt et al., 2021](#)). The addition of olive cake ([Aguilera and Molina-Alcaide, 2021](#)), soybean oil ([Lima et al., 2019](#)), selected tropical tree leaves supplementation ([Malik et al., 2017](#)) reduces enteric CH₄ emissions in sheep.

Most of the nutrients required by sheep are obtained from grazing resources, as they are primarily raised on rangelands. Improving the nutritive value of feeds may significantly improve flock productivity and feed efficiency, and decrease enteric methane emissions ([Hristov et al., 2013](#); [Johnson and Johnson, 1995](#)). Dry matter intake (DMI) is the major determinant of enteric CH₄ production and, consequently, the most important variable for predicting enteric CH₄ production in sheep ([Aguilera and Molina-Alcaide, 2021](#)). The methane production potential (B₀), also referred to as the maximum methane production capacity, of manure varies depending on the animal species and diet (IPCC, 2019 ref). The methane conversion factor (MCF) defines the proportion of B₀ obtained. Theoretically, the MCF varies between 0-100%. Temperature and retention time are important in the calculation of MCF. Manure in liquid form, in hot conditions and kept for a long time, produces more methane. In such manure management systems, the MCF varies between 65-80%. Dry manure has an MCF of about 1% under cold conditions. In the Tier 2 system, the average MCF should be calculated according to the manure management system for each climate zone. The MCF is then multiplied by VS and B₀. The assumed MCFs for each manure management system were developed

from previous studies; the inclusion of additional studies on methane production under various management scenarios may increase the accuracy of these factors ([IPCC, 2019](#)).

The selection of greenhouse gas estimation method depends on factors such as emission sources, research objectives, desired accuracy, and availability of financial resources ([Bhatta et al., 2007](#)). Direct measurements are not always possible, especially when estimations apply to large areas such as regions, countries, or even continents. Indirect methods are preferred for large areas. Inventories, equations, and mathematical models allow the estimation of greenhouse gas emissions when considering large numbers of animals and farms ([Storm et al., 2012](#)). IPCC guidelines ([IPCC, 2006](#)) are accepted as standard methods for emission estimation for each production sector. The IPCC method offers three levels of analysis, called Tier 1, Tier 2 and Tier 3. The choice of Tier depends on the availability of the information requested for the calculations and the size of the system considered. Estimations referring to large areas such as continents and nations are usually carried out by applying Tier 1 and Tier 2. Tier 3 is usually applied to limited areas or even to single entities such as industries or farms. Given the high variability in ruminant production systems (due to breeds, production direction, geographical and environmental conditions, available feeds, management, etc.), a simplified approach should be sufficient for initial estimates in most countries ([Zervas and Tsiplakou, 2012](#)).

In this study, the Tier 2 method recommended by the [IPCC, \(2019\)](#) was applied to estimate enteric and manure-derived CH₄ emissions in two sheep breeds (Akkaraman and Anatolian Merino) and three goat breeds (Hairy, Mohair, and Saanen) commonly raised in Ankara Province in Türkiye.

Materials and Methods

Field study

The enterprises were determined according to the density, diversity, and production systems of livestock operations in the districts of Ankara Province. A total of 25 sheep and goat enterprises were visited across the province of Ankara. Average values were obtained from data collected through surveys and on-site observations, scientific publications, and information provided by the Ministry and breeder associations.

Methodology

The method for estimating gross energy (GE) consumption for animal categories is given below.

$$GE = [(NEm + NEa + NEl + NEp) / REM] + (NEg + NEw) / REG] / DE$$

Explanation of the terms in the model;

GE: Gross energy, (MJ head⁻¹ day⁻¹),
 NEm: net energy for maintenance, (MJ head⁻¹ day⁻¹),
 NEa: net energy for activity, (MJ head⁻¹ day⁻¹),
 NEl: net energy for lactation, (MJ head⁻¹ day⁻¹),
 NEp: net energy for pregnancy, (MJ head⁻¹ day⁻¹),
 NEg: net energy for growth, (MJ head⁻¹ day⁻¹),
 NEw: net energy for wool/mohair, (MJ head⁻¹ day⁻¹),
 DE: digestible energy fraction of GE (DE/GE %)

The method for estimating emission factors (EF) for animal categories is given below.

$$EF = [GE * (Ym/100) * 365] / 55.65 \text{ (kg CH}_4 \text{ head}^{-1} \text{ yr}^{-1})$$

Explanation of the terms in the model;

EF: Emission factor (kg CH₄ head⁻¹ yr⁻¹),
 Ym = methane conversion factor (%)

Estimating dry matter intake (DMI)

Dry matter intake (DMI) was estimated by dividing the gross energy (GE) requirement of each animal subcategory by the assumed dietary energy density (18.45 MJ kg⁻¹ DM), following [IPCC \(2019\)](#).

Manure characteristics

The two main factors—volatile solids (VS) and the maximum methane production capacity (B₀)—have a significant effect on methane emissions. Reference B₀ values of 0.19 for sheep and 0.18 for goats were obtained from the [IPCC \(2019\)](#) guidelines. In this study, IPCC default values were used for methane conversion factor (MCF), in which MCF values for dry storage and pasture systems ranged from 0.47% to 4.0%, depending on the climate zone.

The method to estimate manure-derived emission factors (EF) for each animal category is given below.

$$EF(T) = (VS(T) \times 365) \times [Bo(T) \times 0.67 \text{ kg m}^{-3} \times \sum S, k \text{ MCF}_{S, k} / 100 \times MS(T, S, k)]$$

Explanation of the terms in the model;

EF_(T) = emission factor of animal manure for animal category (T), (kg CH₄ head⁻¹ yr⁻¹),
 VS_(T) = volatile solid excretion of animal category (T) (kg CH₄ head⁻¹ day⁻¹),
 B₀ (T) = maximum CH₄ producing capacity for each category,
 0.67 = conversion factor from m³ CH₄ to kg CH₄,
 MCF_{S, k} = methane conversion factor for manure management system (S), %,
 MS (T, S, k) = animal waste management system.

Climate class

Ankara has a Mediterranean climate according to the Köppen–Geiger climate classification, with hot, dry summers and cold winters. Recent observations indicate that the annual average precipitation is 387 mm, indicating a semi-arid climate over the long term ([Danandeh et al., 2020](#)). The average number of rainy days is 104, with an mean maximum temperature of 17.8 °C, an mean minimum temperature of 6.3 °C, and a mean annual temperature of 11.9 °C ([Arslan and Bağdatlı, 2021](#)).

Results and Discussion

The activity data for sheep and goats, along with the estimated GE, DMI, and enteric EF values, are presented in Table 1. The average live weight (LW) was 49.7 kg head⁻¹ for the breeds studied. Average lactation milk yield (LMY) was 133.4 kg per lactation, and fiber (wool/mohair) production averaged 2.73 kg head⁻¹ yr⁻¹. The mean enteric EF was 9.61 kg CH₄ head⁻¹ yr⁻¹, with

Table 1. Activity data and estimated GE, DMI, and enteric CH₄ emission factors for sheep and goat breeds

Breed	LW	LMY	Wool/mohair	Litter size	GE	DMI	EF
Akkaraman	51.08	78.77	2.25	1.18	23.68	1.28	9.32
Anatolian Merino	52.08	76.00	3.25	1.40	21.73	1.18	8.55
Angora goat	43.30	73.25	2.67	1.13	25.06	1.36	9.04
Hairy goat	50.65	115.50	-	1.15	30.10	1.63	10.86
Saanen goat	51.38	323.67	-	1.60	28.55	1.55	10.30
Mean	49.7	133.4	2.73	1.29	25.82	1.40	9.61

LW: live weight (kg head⁻¹), LMY: lactation milk yield, GE: gross energy (MJ head⁻¹ day⁻¹), DMI: dry matter intake (kg DM head⁻¹ day⁻¹), EF: enteric emission factor (kg CH₄ head⁻¹ yr⁻¹)

the highest value of 10.86 kg CH₄ head⁻¹ yr⁻¹ in Hairy goats and the lowest value of 8.55 kg CH₄ head⁻¹ yr⁻¹ in Anatolian Merino sheep. Estimated GE was highest in Hairy goats (30.10 MJ head⁻¹ day⁻¹) and lowest in Anatolian Merino sheep (21.73 MJ head⁻¹ day⁻¹).

Average DMI was 1.40 kg DM head⁻¹ day⁻¹.

Table 2 shows the average VS excretion and the CH₄ emission factor (EF) for animal manure in comparison with the IPCC default values. The mean VS estimate was twice as much as the IPCC default values,

Table 2. Estimated values and IPCC default factors for volatile solids (VS) and CH₄ emission factors from animal manure

Breed	VS	EF	PCC	
			VS	EF
Akkaraman	0,83	0,45	0,42	0,39
Anatolian Merino	0,82	0,36	0,43	0,34
Angora goat	0,88	0,32	0,39	0,22
Hairy goat	1,06	0,38	0,46	0,35
Saanen goat	1,15	0,77	0,46	0,46
Mean	0,95	0,46	0,43	0,35

VS: volatile solids (kg DM head⁻¹ day⁻¹), EF: emission factor (kg CH₄ head⁻¹ yr⁻¹)

and the CH₄ EF for animal manure was also higher than the IPCC default value. The total average VS excretion was estimated at 0.95 kg DM head⁻¹ day⁻¹, and the average CH₄ EF for animal manure was 0.46 kg CH₄ head⁻¹ yr⁻¹.

The activity data of the studied breeds were selected from peer-reviewed journals. Therefore, breed-specific phenotypes were collected through a review of published articles. Published data were used for Akkaraman sheep (Dellal et al., 2002; Dağ et al., 2000; Şireli, 1996; Gürsoy, 2006; Altın, 2001; Esen and Özbey, 2002; Kahraman and Yüceer, 2020; Aşkan and Aygün, 2020; Çolakoglu and Özbeyaz, 1999; Özmen et al., 2015), for Anatolian Merino (Boztepe, 2013; Tuncer and Cengiz, 2018; Aktaş et al., 2016), for Angora goat (Vatansever and Akçapınar, 2006; Erol, 2012, 2014, 2017; Yertürk and Odabaşoglu, 2007), for Hairy goat (Koyuncu, 1990; Şam et al., 2024; Erten and Yılmaz, 2013; Erişir and Gürdoğan, 2004), and for Saanen goat (Bolacali and Kucuk, 2012; Tölü et al., 2009, 2010; Ceyhan and Karadağ, 2009; Peşmen, 2005; Aktaş et al., 2012). After the review, the GE, DMI and EF for enteric fermentation were estimated. For example, the IPCC (2019) default live-weight values for sheep and goats are 40 and 36 kg, respectively. In this study, LW values were determined based on values reported in the literature (Table 1). The LW values of the sheep and goat breeds were higher than the IPCC (2019) default values.

According to the IPCC (2019), Tier 1 enteric CH₄ EF values were reported as the IPCC (2019) as 9 and 5 kg CH₄ head⁻¹ yr⁻¹ for high (40 kg LW) and low (31 kg LW) production systems, respectively. Methane EF values were reported as 12.5 and 0.22 kg CH₄ head⁻¹ yr⁻¹ for enteric and manure emissions, respectively, in grazing

sheep in Australia (Bell et al., 2012). The enteric CH₄ EF was reported as 6.68 kg CH₄ head⁻¹ yr⁻¹ for sheep with an LW of 48.7 kg (Patra et al., 2016). Patra and Lalhriatpuii (2016), who aimed to explain methane emissions based on diet dynamics through 42 scientific studies conducted on goats, reported enteric methane emissions of 14.3 g head⁻¹ day⁻¹ (equivalent to 5.2 kg CH₄ head⁻¹ yr⁻¹). Zhou et al. (2007) reported average enteric methane emissions for sheep and goats as 5.34 and 4.62 kg CH₄ head⁻¹ yr⁻¹ under Chinese national conditions. In this study, the mean GE, DMI, and enteric EF were 25.82 MJ head⁻¹ day⁻¹, 1.40 kg DM head⁻¹ day⁻¹, and 9.61 kg CH₄ head⁻¹ yr⁻¹, respectively.

Two experiments with twelve rams of the autochthonous Segureña breed (initial average body weight, BW) 40.2 ± 0.75 kg were conducted. GE was 18.8 MJ kg⁻¹ DM, and GE intake was 12.09 MJ day⁻¹ (Aguilera and Molina-Alcaide, 2021). GE intake ranged from 20 to 23 MJ day⁻¹ with 74% GE digestibility under Australian pasture conditions (Bell et al., 2012). GE intake was estimated at 16.3 MJ day⁻¹ (Patra et al., 2016). Dry matter intake was 1.06 kg DM head⁻¹ day⁻¹, and DM digestibility ranged between 65% and 72% in Malpura ewes (LW: 35 kg) (Bhatt et al., 2021). DMI ranged from 1.22 to 1.28 kg DM head⁻¹ day⁻¹ in Santa Inês crossbred intact male sheep in Brazil (Lima et al., 2019). In this study, the lowest GE estimate was 21.73 MJ head⁻¹ day⁻¹.

Moeletsi and Tongwane (2015) reported VS excretion under South African conditions as 0.40 and 0.30 kg VS head⁻¹ day⁻¹ for sheep and goats, respectively. Zhou et al. (2007) reported average manure methane emissions of 0.10 and 0.13 kg CH₄ head⁻¹ yr⁻¹ for sheep and goats, respectively. In this study, the mean VS estimate was twice as much as the

IPCC default values, and the manure CH₄ EF was also higher than the IPCC default values (Table 2). However, most of the EF values from animal manure were only slightly higher than the IPCC defaults. The [IPCC \(2019\)](#) report presented regional averages for animal waste management systems (AWMS) of 54% and 46% for sheep and goats, respectively, and 9% and 91% for landfills and pastures, respectively. In this study, solid storage and pasture AWMS differed among the studied breeds. The combined results of the experimental data and this study indicated higher VS output and higher EF values under the studied manure management systems.

There is a need to develop methods that are globally recognized and applicable in Türkiye to reduce greenhouse gas emissions. For example, the use of specific feed additives in animal diets can reduce emissions originating from enteric fermentation. More importantly, high-quality feeds can substantially decrease emissions. The use of high-yielding breeds or intensive livestock production systems results in lower greenhouse gas emissions compared with extensive or pasture-based systems. Breeds with high productivity and feed efficiency produce greater output per animal, which in turn reduces natural resource use and CH₄ emissions. In Türkiye, raising high-yielding breeds and their crossbreeds under appropriate feeding conditions is considered one of the most important strategies for mitigating greenhouse gas emissions.

The choice of a greenhouse gas estimation method depends on several factors. Indirect methods, such as inventories, equations, and mathematical models, enable the estimation of greenhouse gas emissions when large numbers of animals and farms are considered ([Storm et al., 2012](#)). The IPCC offers three analytical levels—Tier 1, Tier 2, and Tier 3—as standard emission estimation approaches for each production system or animal category. The choice of Tier depends on the availability of the data required for calculations and the scale of the system under consideration. Given the high variability present in ruminant production systems, a simplified approach may be sufficient for initial estimates in Türkiye.

Conclusions

The current study's estimates will be useful in developing the national methane inventory. If more detailed data are collected to improve population statistics, additional subcategories may be added, or existing subgroups may differ among regions. There may be uncertainties or assumptions with limited empirical support in the existing population and performance data. Therefore, the inventory framework used in this study can serve as a tool for continuous estimation and for updating data management systems. Performance data directly influence the

estimation of gross energy (GE), dry matter intake (DMI), and volatile solids (VS), serving as fundamental parameters in livestock subcategory assessments. Consequently, the estimation of national enteric methane emissions by livestock subcategory should be conducted in accordance with the Tier 2 methodology.

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Author Contribution

M.Y.: Project Administration, Conceptualization, Investigation, Methodology, Software, Resources, Data Curation, Writing-Original Draft Preparation, Visualization, Supervision. **A.E.:** Investigation, Methodology, Software, Writing – Review & Editing. **M.F.Y. and V.D.:** Investigation, Formal Analysis. **E.K. and S.B.:** Investigation, Methodology, Formal Analysis. **E.Ü.:** Investigation, Methodology, Software. **M.İ.C. and R.S.:** Investigation, Methodology.

Conflict of Interest

The authors declare that they have no known competing financial or non-financial, professional, or personal conflicts that could have appeared to influence the work reported in this paper.

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