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CONTENTS **Page**

Research Articles

Assessment of Different Growing Media on Cut Flower Performance of Two Gladiolus (*Gladiolus grandiflorus*) Cultivars 36-42
Ümmü Özgül KARAGÜZEL

The Effects of Different Growing Media and Humic Acid Applications on the Growth of Tomato Plants 43-48
Beyzanur YILDIZ Aslıhan ÇİLİNGİR TÜTÜNCÜ Salih DEMİRKAYA Harun ÖZER Adnan UĞUR

Evaluation of the Relations between Yield and Yield Components of Tomato (*Solanum lycopersicum* L.) Hybrids by Correlation and Path Analysis 49-54
İbrahim ÇELİK Serkan AYDIN Halim Can KAYIKÇİ Abdullah ÜNLÜ Emine GÜMRÜKÇÜ Nejla ÇELİK

Determination of *Phytophthora* Species Causing Root and Crown Rot on Tomatoes Grown in Antalya Province and Reactions of Some Tomato Genotypes against *Phytophthora nicotianae* 55-61
Emine GÜMRÜKCÜ Gürsel KARACA

The Effect of the Application of Mycorrhiza on Vegetative Growth, Mineral Element Intake, and Some Biochemical Characteristics of Strawberry Seedlings under Lime Stress 62-71
Gülden BALCI

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RESEARCH PAPER

Assessment of Different Growing Media on Cut Flower Performance of Two Gladiolus (*Gladiolus grandiflorus*) Cultivars

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Abstract

One of the most important problems encountered in the cultivation of cut flower gladiolus is soil-borne diseases and pests. This problem substantially reduces flower yield and quality. Soilless culture is very limited for gladiolus and it has not been studied extensively. The characteristics of the growing media used in soilless culture either directly or indirectly affect yield and quality. Therefore, it is quite essential to determine the appropriate growing media in cut flower cultivation. At the same time good flower production usually depends upon various factors including the type of growing media used. The present study was conducted to determine the effects of two different gladiolus varieties (*Gladiolus grandiflorus* L. cv. “Purple Flora” and “Ibadan”) and six different growing media (peat+pumice: 1:1, v/v; peat+perlite: 1:1, v/v; rice hull+pumice: 1:2, v/v; coarse sand+peat: 2:1, v/v; soil; and cocopeat) on the some quality parameters of gladiolus in in Batı Akdeniz Agricultural Research Institute (BATEM), Antalya, Türkiye. Quality parameters (stem length, flower stem diameter, stem weight, flowering time, number of florest) were significantly ($p < 0.01$) affected by the different growing media and cultivars. Among the growing media, the earliest flowering time (77.8 days) and the longest stem length (128.0 cm) were determined in peat+perlite, whereas the largest number of florets (15.0 florets spike^{-1}) were recorded in peat+pumice. Regarding the varieties, Purple Flora (84.7 days) flowered earlier than Ibadan (102.7 days), while Ibadan displayed more superior characteristics in terms of the other parameters.

1. Introduction

Gladiolus (*Gladiolus grandiflorus* L.) is an important ornamental plant with a bulbous corm which is among the international cut flower industry. The Netherlands, world leader in the flower bulb trade, exports annually more than 200 different gladiolus varieties to more than 88 countries (Anonymous, 2021). The Netherlands exported gladiolus to the world having worth of € 21.3 million following by the other EU countries (€0.2 million) in 2021 (Anoymous, 2022). Gladiolus is called the “queen of bulbous flowers” as it is a popular bulbous cut flower (Meena et al., 2018). It has a magnificent flowering feature with long and color variety in the flower vase (Mahadik and Neha, 2015). It is an indispensable flower especially in flower baskets, wreaths and car ornaments (Yalçıntaş, 2011). But gladiolus plants and bulbs are susceptible to fungal, bacterial and viral diseases. *Fusarium oxysporium* f. sp gladioli, a soil-borne disease, is one of the most important diseases of gladiolus (Lakshman et al., 2012). In the production of gladiolus and other bulbous cut flowers, especially in field production, soil-borne diseases can hinder production to a great extent. One of the most effective methods used to

eliminate soil-borne diseases is soilless culture (El Sharkawi et al., 2014). In addition, soilless culture is used to produce quality flowers for one year in ornamental plants (Ahmad et al., 2012 b). There are different types of soilless culture. For years, hydroponic culture has been used around the world to grow ornamental plants. Nowadays it has been replaced by solid media culture (aggregate systems). The properties of different materials used as growing media both directly and indirectly affect plant yield and physiology. Organic substrates includes sawdust, coco peat, peat moss, woodchips, fleece, marc, bark etc. whereas, inorganic substrate are perlite, vermiculite, zeolite, gravel, rockwool, sand, glass wool, pumice, sepiolite, expanded clay, volcanic tuff and synthetically produced substrates are hydrogel, foam mates (polyurethane), oasis (plastic foam) etc. (Hussain et al., 2014; Asaduzzaman et al., 2015). Thus, choosing the right material as the growing medium is crucial. Most growers use a peat based substrate for growing their crops; however there is a tendency using alternative growing media such as rice hulls and cocopeat in recent years (Ahmad et al., 2012a; El Hanafy et al., 2018).

In the literature, there are a lot of studies about the gladiolus: corm size (Laskar and Jana, 1994; Kazaz and Özzambak, 2002; Memon et al., 2009), plant growth regulators (Karagüzel et al., 1995), planting time (Kalasareddi et al., 1997; Özzambak and Kazaz, 2002; Ahmad et al., 2011), adaptation (Gürcan and Türkoğlu, 2000), irrigation (Baştuğ et al., 2006), plant nutrition (Halder et al., 2007), low temperature applications (Zalewska and Antkowiak, 2009), planting time and phenology (Schwab et al., 2015), heritability (Patra and Mohanty, 2015), planting spacing and depth (Tomiozzo et al., 2018), and hybridization (Hossain et al., 2012; Azimi, 2019) on yield, quality and earliness in gladiolus have been investigated by many researchers. However, the studies conducted on gladiolus in soilless culture are limited (Bazaraa et al., 2014; Saeed, 2018). For the above-mentioned reasons, the study aimed to determine the effects of different growing media and their mixtures on yield and some quality parameters of two gladiolus cultivars under soilless culture.

2. Material and Methods

2.1. Experimental site

The research was conducted in a naturally ventilated plastic greenhouse at the Batı Akdeniz Agricultural Research Institute (BATEM), Antalya, Türkiye between December 24, 2011 and March 13, 2012. The greenhouse is located at coordinates of 36°56'35˝ N and 30°53'39˝ E and is 12 m above sea level. The average temperature and relative humidity inside the greenhouse for the study period are presented in Figure 1. The temperature and relative humidity ranged from 7.6 to 23.3°C and 28.1 to 85.4%, respectively.

2.2. Plant material and planting

Corms with a circumference of 6-8 cm belonging to two different gladiolus varieties (Purple Flora and Ibadan) were used as plant material. Ibadan is an orange-colored variety with a flowering time of 95 to 105 days, whereas Purple Flora is a blue and purple- colored variety with a flowering time of 75 to 85 days (Anonoymous, 2009; Yalçıntaş, 2011). The corms were stored at 5ºC for 2.5 months and left in a solution containing 50% Benomyl as a protectant against fungal diseases for 30 minutes prior to planting. The corms were planted into the boxes (37.0 cm in length, 37.0 cm in width, and 22.5 cm in depth) in a depth of 5 cm with 15.0×15.0 cm spacing and in 2 rows on October 24, 2011.

2.3. Treatments

Five different growing media, i.e. peat+pumice (1:1, v/v), burnt rice hull+pumice (1:2, v/v), coarse

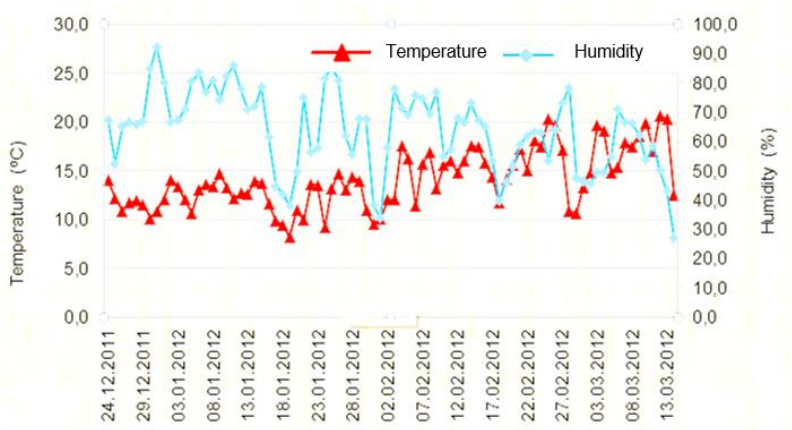

Figure 1. Average temperature and relative humidity measured inside greenhouse.

sand (>1 mm)+ peat (2:1, v/v), cocopeat, and peat+perlite (1:2, v/v), and soil alone as the control were used in the study. After preparation of the mixture, the pre-planting media was wetted until saturated. No fertilizer has been added to the media. The physical and chemical analyses of soil and different growing media were made and then they were placed into the boxes. The results of the analyses are presented in Tables 1 and 2.

2.4. Nutrient solution and irrigation

Irrigation water and nutrients were applied to the plants with drip irrigation method. The plants were provided with 100 cc of water per plant in each irrigation application. When the plants reached the three-leaf period, fertigation was started, and a nutrient solution was applied to the plants twice a week. The composition of nutrient solution (ppm) was as follows: N:250, P:35, K:350, Ca:180, Mg:50, Fe:3, Mn:1, Zn:0.06, Cu:0.1, B:0.1 and Mo:0.01. The pH was measured in every fertigation and was adjusted to values between 5.7-6.0, and the EC kept between 2.0-2.5 mS cm^{-1}.

2.5. Harvest and measurements

The harvest was carried out to leave two mutual leaves on the plant as of the soil level when the 2 to 3 florets at the very bottom of the spike colored. Five sample plants were counted randomly taken from each treatment and average was calculated. Stem length (cm $plant^{-1}$), stem diameter (mm $plant^{-1}$), stem weight (g $plant^{-1}$), flowering time (day) and the number of florets (number $spike^{-1}$) were determined according to Yalçıntaş (2011) in the study. Stem length (cm) is the distance from ground level to the last candle; the number of florets (number $spike^{-1}$) is the total number of flower candles on a spike; flowering time (days to 50% flowering) is the number of days for 50 per cent plants to flowering; stem diameter is calculated by digital caliper from 5 cm below of lower candle; stem weight is calculated using digital weighing with 0.01 g accuracy.

2.6. Experimental design and statistical analysis

The experiment was set up according to the split-plot experimental design in randomized blocks with 3 replications, and a total of 720 corms were used, with each replication containing 20 corms for each growing medium. The obtained data were subjected to an analysis of variance in Tarist statistical program, and the mean values were compared using Duncan's multiple range tests at the 0.05 level.

3. Results and Discussion

3.1. Stem length

The effects of growing media and cultivars on stem length were found statistically significant ($p < 0.01$), whereas the effect of the variety × growing media interaction on stem length was statistically insignificant (Table 3). Among the growing media, the longest stem length was recorded in peat+perlite (128.0 cm), followed by peat+pumice (126.0 cm); however, both media were in the same statistical group in terms of stem length. The stem

Table 1. Some physical and chemical characteristics of the growing media.

Characteristics	Peat	Cocopeat	Rice Hull	Pumice	Sand
pH	6.6	7.2	6.5	8.6	8.7
EC (µmhos cm^{-1})	159.0	193.0	890.0	44.0	68.0
Moist (%)	68.0	81.7	9.3	-	-
Dry matter (%)	32.0	18.3	90.7	-	-
Organic matter (%)	91.9	89.9	81.0	-	-
Ash (%)	8.1	10.1	19.0	-	-
Total N (%)	1.2	0.2	0.54	-	-
C (%)	53.3	52.1	47.0	-	-
C/N	43.3	358.5	87.5	-	-

Table 2. Physical and chemical characteristics of the studied soil.

Characteristics	Value
pH*	8.3
Lime (%-w w^{-1})	25.7
EC (µmhos cm^{-1})	183.0
Sand (%)	13.0
Clay (%)	31.0
Silt (%)	56.0
Organic matter (%)	1.6
P (ppm)	17.0
K (ppm)	259.0
Ca (ppm)	5502.0
Mg (ppm)	518.0

* measured on watery extract 1:2.5 (v/v).

Table 3. The effects of different growing media on stem length of gladiolus cultivars (cm).

Growing media (M)	Cultivars (C)		Mean for growing media
	Purple Flora	Ibadan	
Soil	102.0	116.3	109.1 b
Cocopeat	102.6	121.6	112.1 b
Pumice+Rice hull	103.3	121.3	112.3 b
Peat+Perlite	120.6	135.3	128.0 a
Sand+Peat	104.6	120.0	112.3 b
Peat+Pumice	116.0	136.0	126.0 a
Mean for cultivars	108.2 b	125.1 a	

Cultivars (C):** Growing media (M): ** C×M: ns
** Significant at the p < 0.01 level, ns: not significant.
Means followed by the same letter do not different significantly according to Duncan's test (p≤0.05).

Table 4. The effects of different growing media on stem diameter of gladiolus cultivars (mm).

Growing media (M)	Cultivars (C)		Mean for growing media
	Purple Flora	Ibadan	
Soil	9.3 b	8.7 c	9.0 c
Cocopeat	10.0 ab	10.8 a	10.4 ab
Pumice+Rice hull	10.0 ab	10.8 a	10.4 ab
Peat+Perlite	10.6 a	11.2 a	10.9 a
Sand+Peat	10.0 ab	9.7 b	9.8 b
Peat+Pumice	10.3 ab	10.4 a	10.3 ab
Mean for cultivars	10.1 b	10.3 a	

Cultivars (C):** Growing media (M): ** C×M: ns
** Significant at the p < 0.01 level, ns: not significant.
Means followed by the same letter do not different significantly according to Duncans test (p≤0.05).

length ranged from 109.1 to 112.3 cm among the other growing media and the differences among them were statistically insignificant. Regarding the varieties, Ibadan (125.1 cm) had a longer stem length than Purple Flora (108.2 cm). Tehranifar et al. (2011) investigated the effect of three soilless media on growth and development of two types of Lilium, and they were used 100% coco peat, 50% gravel + 50% sand and 40% peat + 60% perlite as growing media. They noted the lowest flower stem length with 31.7 cm was obtained from 50% gravel + 50% sand, while the highest were obtained from 40% peat + 60% perlite substrates. El Hanafy et al. (2018) tested the effect of using substrate soilless culture technique on the plant growth, flowering and corms production of Gladiolus *cv*. "Chinon". Three types of soilless culture substrates were put under investigation as follow; mixture of sand and rice husk (1:1 v/v), mixture of peat moss: rice husk (1:1 v/v) and perlite 100%. They reported the longest stem length recorded in peat moss and rice husk (1:1 v/v) (87.5 cm) followed by the sand and rice husk (1:1 v/v) (81.3 cm) in first year in gladiolus. In the second year, the highest stem length was obtained from peat: rice husk (91.3 cm) followed by perlite (82.0 cm). Bhandari et al. (2017) investigated four different growing media (soil, coconut peat, coconut peat+sand, coconut peat+soil) in *Lilium longiflorum* and they found that the maximum stem length was from coconut peat media with 99.8 cm.

3.2. Stem diameter

As a result of the evaluation of the data about stem diameter, the varieties and the growing media were found to have statistically significant (p < 0.01) effects individually (Table 4). Flowers with the thickest stem (10.9 mm) were obtained from the plants grown in peat+perlite. On the other hand, the flowers with the smallest stem diameter (9.0 mm) were in the plants grown in soil. Results of stem diameter confirmed the data given by Tehranifar et al. (2011) who observed the highest root diameter in the lily when grown in peat + perlite substrates. El Hanafy et al. (2018) reported that the highest value for the spike base diameter was recorded in peat: rice husk substrate with 11.2 mm and 12.1 mm while the smallest value was obtained in sand: rice husk substrate 6.0 mm and 7.0 mm in 2016 and 2017 respectively. In our study, the thinnest stem diameter was obtained from the mixture of sand + pumice after soil.

3.3. Stem weight

The effects of the growing media and varieties on stem weight were statistically significant (p < 0.01), while the effect of the growing media × variety interaction was statistically insignificant (Table 5). Among the growing media, the greatest stem weight was obtained from the mixture of peat and pumice (103.6 g plant^{-1}), whereas the lowest stem weight was recorded in pumice+rice hull (76.5 g plant^{-1}). When the stem weights of the varieties were examined, Ibadan was found to have a stem weight of 113.9 g plant^{-1} and Purple Flora was detected to have a stem weight of 65.8 g plant^{-1}. Stem weight results are in alliance with the observations of Yalçıntaş (2011) who claimed in gladiolus when grown in traditional media. Yalçıntaş (2011), determined the stem weight as 114.2 g plant^{-1} in Ibadan and as 56.0 g plant^{-1} in Purple flora.

Table 5. The effects of different growing media on stem weight of gladiolus cultivars (g plant^{-1}).

Growing media (M)	Cultivars (C)		Mean for growing media
	Purple Flora	Ibadan	
Soil	63.8 a	97.3 b	80.5 bc
Cocopeat	66.2 a	120.4 a	93.3 ab
Pumice+Rice hull	61.3 a	91.6 b	76.5 c
Peat+Perlite	61.8 a	122.8 a	92.3 ab
Sand+Peat	64.9 a	121.4 ab	88.1 abc
Peat+Pumice	76.7 a	130.3 a	103.6 a
Mean for cultivars	65.8	113.9	

Cultivars (C):** Growing media (M): ** C×M: ns
** Significant at the p < 0.01 level, ns: not significant.
Means followed by the same letter do not different significantly according to Duncan's test (p≤0.05).

Table 6. The effects of different growing media on flowering time of gladiolus cultivars (day).

Growing media (M)	Cultivars (C)		Mean for growing media
	Purple Flora	Ibadan	
Soil	92.6 a	113.3 a	103.0 a
Cocopeat	86.0 bc	102.0 b	94.0 b
Pumice+Rice hull	89.6 ab	117.3 a	103.5 a
Peat+Perlite	77.0 d	78.6 c	77.8 c
Sand+Peat	80.0 cd	101.3 b	90.6 b
Peat+Pumice	83.0 cd	104.0 b	93.5 b
Mean for cultivars	84.7 b	102.7 a	

Cultivars (C):** Growing media (M): ** C×M: **
** Significant at the p < 0.01 level.
Means followed by the same letter do not different significantly according to Duncans test (p≤0.05).

Table 7. The effects of different growing media on number of florets in gladiolus cultivars (number spike^{-1}).

Growing media (M)	Cultivars (C)		Mean for growing media
	Purple Flora	Ibadan	
Soil	10.3	12.3	11.3 c
Cocopeat	12.6	13.6	13.1 b
Pumice+Rice hull	11.6	15.0	13.3 b
Peat+Perlite	12.6	14.0	13.3 b
Sand+Peat	12.3	14.0	13.1 b
Peat+Pumice	13.6	16.3	15.0 a
Mean for cultivars	12.22 b	14.22 a	

Cultivars (C):** Growing Media (M): ** C×M: ns
** Significant at the p < 0.01 level, ns: not significant.
Means followed by the same letter do not different significantly according to Duncan's test (p≤0.05).

3.4. Flowering time

Cultivars, growing media and the cultivars × growing media interaction significantly (p < 0.01) affected flowering time (Table 6). In the cultivars × growing media interaction, the earliest flowering time was in peat+perlite in both varieties (Purple Flora: 77.0 days; Ibadan: 78.6 days), whereas the latest flowering time was in pumice+rice hull in Ibadan (117.3 days). Among the growing media, the earliest flowering time was determined in peat+perlite (77.8 days), later flowering time was found sand + peat (90.6 days). The latest flowering time was obtained in soil (103.0 days).

Regarding the varieties, Purple Flora (84.7 days) flowered earlier than Ibadan (102.7 days). Tehranifar et al. (2011), reported that the time of bud emergence (29.6 day) and flower harvest (53.5 day) were days earliest when used peat+perlite as media in lilium. El Hanafy et al. (2018) determined the earliest flowering time with sand: rice husk substrate 87.3 and 86.6 days after planting while the latest flowering time was obtained with perlite substrate 106.0 and 101.0 days after planting in 2016 and 2017, respectively.

3.5. Number of florets

The effects of the cultivars and the growing media on the number of florets were statistically significant (p < 0.01), whereas the effect of the cultivars × growing media interaction on the number of florets was statistically insignificant (Table 7). The growing media were in 3 different groups in terms of the number of florets. The mixture of peat and pumice with the largest number of florets (15.0 florets plant^{-1}) was in the first group; the 2nd group was comprised of pumice+rice hull (13.3 florets plant^{-1}), peat+perlite (13.3 florets plant^{-1}); and soil with the smallest number of florets (11.3 florets plant^{-1}) was in the last group and it followed sand+ peat and cocopeat media (13.1 florets plant^{-1}).

Potassium has an important role in the production of proteins and hydrocarbons. Pumice is

alkaline and rich in potassium. In potassium deficiency, growing stops and this stress may limit flowering because the plant cells can not divide to allow the growth and because of potassium deficiency reduce the quality of flowers (Anonymous, 2019a; Khalaj et al., 2019). Therefore, the presence of pumice may have caused a high number of flowers and a short duration of flowering due to the potassium content.

When evaluated all of results among the growing media, the best results were obtained from peat+perlite in terms of plant height, stem diameter and flowering time, in peat+pumice in terms of the number of florets and plant weight. In terms of plant height, one of the most important criteria for quality in gladiolus, the plants in peat+perlite and peat+pumice were approximately 18.8 to 16.8 cm longer than the plants grown in soil. In terms of the number of florets, the plants grown in the growing media concerned had 2.0 to 3.6 more florets than the plants grown in soil. Lopez et al. (2008) reported that different growing media significantly affected growth and flowering in gladiolus (*Gladiolus tristis* subsp. Concolor) and that the stem length, the spike length and the number of florets in the plants grown singly in peat were greater than those of the plants grown singly in perlite. According to Issa et al. (1999), using peat moss based substrates gave more yield with high quality of flower comparing with single substrate.

To determine the rate of plants growth it would be important their physical-chemical properties, water holding capacity and aeration capacity of growing media. Soils are generally inadequate for the production of plants because soils do not provide the aeration, drainage and water holding capacity required. Soil compound can limit plant growth during greenhouse production. Even when the top soil is worked, plants may suffer when roots reach the compacted subsoil. (Anonymous, 2019b). To reduce this disadvantage in pure substrates, various materials with large particle are added (Sambo et al., 2008) the most commonly used being perlite. The addition of perlite to peat improves aeration (Londra et al., 2012).

When the varieties were evaluated in terms of the parameters under examination in the study, it was concluded that Purple Flora flowered 18 days earlier than Ibadan, while better results in terms of the other parameters were obtained in Ibadan in comparison with Purple Flora.

Although the results obtained between the varieties in the study are generally in agreement with the findings by Yalçıntaş (2011), their lower and upper limits vary. This might result from different ecological and growing conditions. The results obtained in the study in terms of plant height, stem diameter and the number of florets resemble the findings by a large number of researchers (Gürcan and Türkoğlu, 2000; Özzambak and Kazaz, 2002; Akpınar and Bulut, 2006; Anonymous, 2009; Saraç et al., 2010).

4. Conclusion

In the study conducted to determine the quality characteristics of two different gladiolus varieties grown in different covered growing media in Antalya, peat+perlite and peat+pumice showed quite good results in terms of the parameters under examination as compared with the other media. Moreover, regarding the varieties, Ibadan showed more superior characteristics than Purple Flora. The plants grown in pumice+rice hull and soil flowered rather later in comparison with the plants that were grown in the other media, and the lowest values in terms of the other parameters were obtained from soil, pumice+rice hull, and sand+peat. In the light of the data obtained in the study, peat+perlite and peat+pumice might be successfully used in the cultivation of gladiolus.

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RESEARCH PAPER

HORTICULTURAL STUDIES

The Effects of Different Growing Media and Humic Acid Applications on the Growth of Tomato Plants

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Abstract

This study was carried out to determine the effects of different growing media (peat + perlite, cocopeat, hazelnut husk, rock wool) and different humic acid doses (2, 4, and 8 ml L^{-1}) on tomato plant growth. The performance of the seedlings from planting to fruit set was evaluated to determine the growth of the tomato plant. For this purpose; stem diameter (mm), plant height (cm), leaf number, first flowering, first fruit set, total plant dry weight (g), root volume (m^3), and relative growth rate (g cm^{-2} $days^{-1}$) parameters were examined. According to the results, the highest stem diameter of 15 mm was obtained in the hazelnut husk media at a dose of 4 ml L^{-1} of humic acid. As a result, while the best root growth was obtained at 2 ml L^{-1} humic acid doses in peat-perlite media, the fastest growth and N (3.33%), P (0.58%) and K (3.91%) content were obtained at 8 ml L^{-1} humic acid doses in cocopeat media. The best leafing, flowering and fruit set were obtained in hazelnut husk media with 2 ml L^{-1} humic acid doses. As a result, the highest relative growth rate (0.064 g cm^{-2} $days^{-1}$) was determined at 8 ml L^{-1} doses of humic acid applied in the cocopeat media.

1. Introduction

While agricultural chemicals are used unconsciously to meet the nutritional needs of the global population and increase yield and resource efficiency, it causes adverse effects on human health, environmental pollution and deterioration of soil structure. On the other hand, there are product returns due to pesticides, especially from foreign markets, which harms the country's economy. Over time, polluting agricultural lands with chemical fertilizers will decrease the productivity of the soils and cause an increase in social problems and the need for food. Due to these concerns, organic agriculture and organic fertilization have become widespread in recent years to reduce synthetic agricultural chemicals such as pesticides and fertilizers and increase the sustainability of agricultural production systems. Improving organic matter in the soil applying organic fertilization; it has been revealed by the researchers that it improves the physical and chemical properties of the soil, increases the amount of microbial biomass, and increases the plant yield and quality by increasing the nitrogen fixation events (Fraser et al., 1988; Tüzel et al., 2011; Özer and Uzun, 2013; Özdemir and Özer, 2016).

Humic acid is an organic soil conditioner with a very high organic matter content, whose colors can change from yellow to black, produced in the form of pellets or as the liquid in concentrated form by processing lignite coal or solid leonardite. At the same time, humic acids have an international certificate of conformity to organic farming (Akıncı, 2011; Kacar, 2013; Sayarer, 2020). Humic acid, an organic soil conditioner, increases the soil water holding capacity, removes the salt in the soil from the root zone, and increases the plant's nutrient uptake by regulating the soil pH. The positive effects of humic acid on plants growth and development are

due to increased availability of nutrients with water, plant root zone development, and increased chlorophyll content. Sabzevari et al. (2010) found that humic acid application had a positive effect on germination rate, seedling growth and emergence rate; Demirtas et al. (2014) revealed in their study that humic acid increases N, P, K, Fe and Cu plant nutrients in tomato plants and significantly affects fruit quality.

Different media are used in tomato production, especially seedling and plant growing. In today's seedling cultivation, organic (peat, bark, hazelnut husk, sawdust, straw-straw and cocopeat) and inorganic (sand, gravel, perlite, vermiculite, pumice, rock wool and zeolite) media are used as growing media (Taşdelen et al., 2021). In many studies, it has been revealed that the effects of different growing environments on different plants may be different (Polat et al., 2017; Yıldırım and Hatipoğlu, 2020; Taşdelen et al., 2021). However, the interaction of these media with different growth regulators and their effects on growth rate after planting have not been studied. In this study, it was aimed to determine the effects of organic cultivation of tomato seedlings produced in different growing media (peat+perlite, rock wool, cocopeat, hazelnut husk) and different humic acid doses (2, 4 and 8 ml L^{-1}) until the first fruit set period after planting.

2. Material and Method

The research was carried out in the glass greenhouse and open field of Ondokuz Mayıs University, Faculty of Agriculture, Department of Horticulture, between April-July 2022. The seeds of H-2274 tomato cultivar were used as plant material in the study. Four different seed sowing media (peat-perlite (65% + 35%), cocopeat, hazelnut husk and rock wool) were used in this study. Peat, perlit, cocopeat and rock wool are commercial growing media. However, hazelnut husk media was prepared. The hazelnut husk waste, which have completed their natural drying processes, were the grinding process with an 8 mm sieve diameter. Before the seed sowing process, the pH values of the environments were determined with the pH meter (Adwa waterproof), and the EC values were determined with the EC meter (Adwa waterproof). A suspension was prepared to measure pH and EC values by taking 1:10 media/pure water from the media into a beaker. Prepared suspensions were shaken on a magnetic stirrer for 1 hour, and then pH and EC values were measured (Table 1).

The tomato seeds used in the study were sown in 210 well viols with 2.6 × 2.6 cm diameter cells containing four different media. In the study, a total of 360 seeds, 30 in each replication, were sown with three replications. The viols, in which the seeds were sown were placed on the growing benches in the glass greenhouse with heating control, and five minutes of irrigation were applied three times a day (at 10.00, 14.00 and 16.00) throughout the growing period. Greenhouse temperature (°C) and relative humidity (%) (KT100, Kimo, France) values were measured during the seedling growing period (Table 2).

In the trial, three different humic acid doses (2, 4 and 8 ml L^{-1}) were applied to the seeds planted in cocopeat, rock wool, hazelnut husk, and peat-perlite. To obtain the humic acid used in the application, after heating 310 L of water at approximately 80°C, the mixture obtained by adding 13 kg of potassium hydroxide (-OH) was completed to 400 liters by adding 80 kg of leonardite, salicylic acid (250 ppm) and IBA (500 ppm). Humic acid was applied every 15 days after germination of the seeds until the 4 leaf seedling stage. Humic acid was sprayed to the leaves in such a way that all the leaves of the seedlings were wet. Seedlings that reached the four leaf stage were planted in the open field, and their growth performances until fruit sets were examined. In the study, planting sites were prepared in areas where tomatoes were not grown in previous years. Soil samples were taken from 0-30 cm depth from different points to represent the trial area before planting. In the texture analysis of soil samples belonging to the experimental area; clay (36.5%), sand (27.3%), silt (36.2%), pH (7.92), EC (0.24 dS m^{-1}), organic matter (6.02%), total nitrogen (0.24%), plant available phosphorus (11.4 ppm) and exchangeable potassium (0.72 cmol kg^{-1}) values were determined according to Kacar (2009). The raised bed was prepared with

Table 1. pH and EC values of seed sowing media.

Growing media	pH	EC (dS cm^{-1})
Peat + Perlite	5.49	0.55
Cocopeat	5.73	0.51
Hazelnut husk	6.45	0.81
Rock wool	8.03	0.21

Table 2. Temperature and relative humidity values in the greenhouse during the seedling and growing period.

	Greenhouse		Field	
	Temperature (°C)	Humidity (%)	Temperature (°C)	Humidity (%)
Lowest	16.1	40.4	12.6	27.2
Highest	41.6	91.9	39.1	81.4
Average	29.2	66.5	25.7	66.6

a height of 20 cm and a width of 1 m as the planting site. Drip irrigation pipes with a dripper spacing of 25 cm were placed in the prepared planting places (raised beds) in a way suitable for double-row planting. Irrigation was carried out with a system that can irrigate according to soil moisture throughout the growing period. Then, ground mulch was applied over the planting areas. Tomato seedlings were planted on the prepared tubes in accordance with the randomized blocks trial design, with a row spacing of 50 × 50 cm. No additional fertilization was made since planting.

In order to determine the quality of the seedlings, the seedlings were uprooted at the first fruit set; they were divided into roots, stems and leaves. Plant height was measured in meters from root collar to growth tip. Stem diameter was measured in mm from the root collar with the help of a digital caliper. The number of leaves was determined as pieces by counting the number of leaves manually since planting. After planting, the number of days until the first flowering and first fruit set was calculated and determined as flowering and fruit set. Leaves, roots and stems separated from seedlings and plants were placed separately in small paper bags in an oven at 65°C. The drying process was carried out for at least 48 hours. It was decided whether the drying process was completed by applying the weight change method on the samples that did not complete their drying during this period. When it was understood that the samples were completely dry, the dry weights of the leaves, root and stem were weighed with a balance sensitive to 0.01 g. After weighing 0.5 g of the ground samples, dry digested at 550°C for 4-8 hours in the ash furnace, the ash was dissolved in hydrochloric acid. The total nitrogen (N) content of the plant samples was determined according to the Kjeldahl method, the phosphorus (P) contents were determined spectrophotometrically according to the vanado-molybdophosphoric acid method in the obtained solutions, and the potassium (K) content was determined using atomic absorption spectrophotometer in the obtained solutions (Jones, 2001).

WinRhizo root analysis program (Regent Instrument Inc. Canada) was used to examine tomato plants root anatomy and determine the rooting levels in detail. Plant roots taken after planting were carefully washed and dried with a paper towel. The root part was placed on the scanner part of the device and transferred to the computer environment in three dimensions. Root volume (cm^3), which reveal root architectures in detail, were examined as a result of root scanning with the WinRhizo program. Root, stem and leaf dry weights and their ratios, leaf area (using the previously created models), net assimilation rate and relative growth rate analyzes were made according to Uzun (1996). The study was conducted in a Randomized Complete Block Design in split-plot arrangements with three replications. SPSS 17.0 statistical software was used to analyze experimental results using two-way ANOVA (growing media × humic acid doses). The differences between treatment was determined by the Duncan's multiple comparison tests at the $p<0.05$ significance level.

3. Results and Discussion

Peat + perlite and coconut fiber media came to the fore in seed germination rate in the study. Considering the pH (5.49-5.73) and EC (0.55-0.51 dS cm^{-1}) values of these media, it is seen that they were in the appropriate range for plant growth and development (Table 1). Thus, it is known that peat is a suitable environment for seedling cultivation due to its low volume weight and high water holding capacity (Munsuz et al., 1982, Demiral, 2016). It is thought that the hazelnut husk media has a higher EC (0.81 dS cm^{-1}) value compared to other environments that harm seed germination. It is known that salinity inhibits seed germination, reduces nutrient use in the seed and goes into dormancy (Ahmad et al., 1992; Yıldız et al., 2007). The lowest seed germination determined in the hazelnut husk media with the highest electrical conductivity.

The effects of different growing media (peat-perlite, cocopeat, hazelnut husk and rock wool) and humic acid doses (2, 4 and 8 ml L^{-1}) on stem diameter, plant height, number of leaves, flowering and fruit set times were investigated. According to the findings obtained, the stem diameter, the plant height, the number of leaves, the flowering and fruit set time were found to be statistically significant (**$p < 0.01$; *$p < 0.05$) (Table 3).

The maximum stem diameter (15 mm) was obtained from the 4 ml L^{-1} dose of hazelnut husk media, followed by 4, 2 and 8 ml L^{-1} doses of peat+perlite media, respectively. Considering the tomato plant height values, the highest plant height was obtained from the 2 ml L^{-1} doses of rock wool (45.7 cm) and hazelnut husk (45.7 cm) media. These values were followed by the 4 ml L^{-1} dose of rock wool (45 cm) media. Considering the leaf number values, it was found statistically significant at the doses of 2 ml L^{-1} for hazelnut husk (9 leaves) and 2-4 ml L^{-1} for rock wool (9 leaves). While there were similarities between the flowering times and fruit set times of the grown tomato plants, the latest flowering and fruit set was achieved at 2 ml L^{-1} of hazelnut husk media. Similar results were obtained in the study in which the effects of seedling quality on flowering and fruit set in tomatoes were examined, while the earliest flowering date was 27 days. The earliest fruit set date was 30 days (Özer and Kandemir, 2016).

The highest root volume (5967 cm^3) was found to be statistically significant in the peat + perlite media at a dose of 2 ml L^{-1}. The humic acid application doses of peat + perlite media were

Table 3. The effects of different growing media (peat-perlite, cocopeat, hazelnut husk and rock wool) and humic acid doses (2, 4 and 8 ml L^{-1}) on stem diameter, plant height, leaf number, days from planting to first flowering and fruit set.

Growing media	Doses (ml L^{-1})	Stem diameter (mm)	Plant height (cm)	Leaf number	Flowering	Fruit set
Peat-perlite (65% + 35%)	2	12.4 c	31.0 f	7.0 b	33.0 ab	36.0 b
	4	13.0 b	30.7 f	7.0 b	38.0 ab	43.0 ab
	8	12.2 c	34.7 de	8.0 ab	30.0 b	34.0 b
Cocopeat	2	7.0 ı	31.7 f	7.0 c	33.0 ab	38.0 ab
	4	8.7 h	35.3 d	7.0 c	38.0 ab	42.0 ab
	8	8.1 g	38.7 c	8.0 ab	33.0 ab	37.0 ab
Hazelnut husk	2	11.4 d	45.7 a	9.0 a**	41.0 a**	45.0 a*
	4	15.0 a**	33.5 e	7.0 c	33.0 ab	36.0 b
	8	10.4 d	33.6 e	7.0 b	34.0 ab	38.0 ab
Rock wool	2	9.4 e	45.7 a**	9.0 a**	28.0 b	34.0 b
	4	7.7 h	45.0 a	9.0 a**	33.0 ab	37.0 ab
	8	11.2 d	42.7 b	8.0 b	29.0 ab	35.0 b
Main effects						
Growing media	Peat-perlite	12.6 a**	32.1 b	7.6 b	33.8 ab	37.4 ab
	Cocopeat	7.9	35.2 ab	7.2 b	34.4 ab	38.9 b
	Hazelnut husk	12.3 a	37.6 ab	7.4 b	36.0 b	40.0 b
	Rock wool	9.4	44.4 a*	8.3 a*	30.1 a*	35.4 a*
Doses	2 ml L^{-1}	10.6	37.5	7.7	35.1 ab	39.8 ab
	4 ml L^{-1}	10.5	37.3	7.8	32.0 b	36.4 a *
	8 ml L^{-1}	10.6	37.2	7.3	33.7 a*	37.7 ab

***p < 0.01; *p < 0.05*

Table 4. The effects of different growing media (peat-perlite, cocopeat, hazelnut husk and rock wool) and humic acid doses (2, 4 and 8 ml L^{-1}) on root volume, total dry weight, relative growth rate, N%, P% and K%.

Growing media		Doses (ml L^{-1})	Root volume (cm^3)	Total dry weight (g)	Relative growth rate (g cm^{-2} $days^{-1}$)	N (%)	P (%)	K (%)
Peat-perlite (65% + 35%)		2	5967 a**	42.5 a**	0.029 j	2.99 b	0.48 d	2.66 e
		4	3050 c	29.3 c	0.026 l	2.30 g	0.32 h	2.43 ı
		8	3410 b	21.6 g	0.034 d	2.38 d	0.30 i	2.97 c
Cocopeat		2	42 k	21.50 g	0.032 ı	2.04 h	0.24 j	2.58 f
		4	27 l	13.29 ı	0.035 c	2.87 c	0.55 b	3.66 b
		8	424 e	12.73 i	0.064 a**	3.33 a**	0.58 a	3.91 a**
Hazelnut husk		2	170 j	38.47 b	0.025 m	2.37 e	0.45 e	2.57 g
		4	337 f	23.15 e	0.027 k	1.44 k	0.50 c	2.74 d
		8	208 i	18.63 h	0.030 i	1.95 ı	0.35 g	2.45 h
Rock wool		2	268 g	25.53 d	0.032 h	1.77 j	0.38 f	1.86 k
		4	680 d	22.17 f	0.033 g	1.88 i	0.32 h	2.14 i
		8	234 ı	23.40 e	0.041 b	1.34 f	0.31 ı	1.91 j
Main Effects								
Growing media	Peat-perlite		4142 a**	26.75 b	0.030 ab	2.75 a*	0.45 a*	3.39 a*
	Cocopeat		164 b	31.10 a*	0.044 a*	1.66 b	0.33 b	1.97 b
	Hazelnut husk		238 b	23.70 b	0.028 b	2.57 a	0.37 b	2.69 ab
	Rock wool		394 b	15.84 c	0.036 ab	1.93 b	0.43 a	2.59 ab
Doses	2 ml L^{-1}		1235	24.43	0.033	2.30	0.39 ab	2.42 ab
	4 ml L^{-1}		1234	24.32	0.034	2.13	0.42 a*	2.74 ab
	8 ml L^{-1}		1236	24.30	0.036	2.26	0.38 ab	2.82 a*

***p < 0.01; *p < 0.05*

higher than other media. Similarly, the highest plant total dry weight was determined as 42.5 g in peat+perlite media, where a 2 ml L^{-1} humic acid dose was applied during the seedling period. Considering tomato plants relative growth rate values, it was determined that the highest values were obtained from the 8 ml L^{-1} dose of cocopeat (0.064 g cm^{-2} $days^{-1}$). According to the results obtained, the lowest relative growth rate (0.029 g cm^{-2} $days^{-1}$) was obtained in the application (Peat-perlite and humic acid doses; 2 ml L^{-1}) with the highest root volume. When we examined the main effect, it was determined that 0.044 g cm^{-2} $days^{-1}$ with the highest relative growth rate was in the cocopeat application, while the effect of humic acid doses on growth was not determined (Table 4). Bozkurt (2005) stated that humic and fulvic acids accelerate the plant's growth by accelerating cell division. They reported that there was rapid growth, especially in the seedling period. In this respect, the study's findings are similar to ours. In our study, the significant effects of the humic acid doses applied during the seedling period on the total dry weight of the plant could not be determined. However, the highest dry weight (31.10 g) was determined in the cocopeat media with the highest growth. Uzun et al. (1998) stated that plant growth should grow slowly and steadily.

Slow and stable growing plants accumulate dry matter in the roots and stems in their early development stages. In this way, the greening period of tomato plants that complete their development is prolonged, and an increase in photosynthesis abilities in a more extended period results an increase yield. In the study by Özer (2017), in which similar results were expressed, maximum yield was achieved by shading and prolonging the greening time of tomato plants.

Yücel (2006) reported that the best results (total emergence rates, stem diameter, stem and root length, number of leaves, seedling fresh and dry weight) were obtained in 100 mg kg^{-1} humic acid applied to peat media in cucumber and tomato seedlings. They also observed that their application increased the essential plant nutrient (NPK) uptake of tomato and cucumber seedlings. While different humic acid doses (0, 30, 60, 90 and 120 ppm) applied in beans plants did not have a significant effect on dry matter accumulation in plants, it was reported that it increased the uptake of elements such as N, P, Fe, Mn and Zn in the leaves (Sözüdoğru et al., 1996). In the study in which different doses of humic acid (0, 400, 800 and 1200 mg kg^{-1}) were applied to lettuce, it was determined that nitrate, total N, Ca, Cu, Zn and Mn contents of plants generally increased with increasing humic acid dose (Odabaş, 2019). One of the most important reasons why humic acid applications increase plant nutrient intake is that it provides better plant root system development (David et al., 1994; Demirtaş et al., 2014). In our study, where similar results were obtained, the highest plant nutrients N (2.75%), P (0.45%) and K (3.39%) values were obtained from peat + perlite media. When the effects of humic acid doses on plant nutrients were examined, the highest phosphorus content (0.42%) came to the fore in the 4 ml L^{-1} application, while the potassium content was measured with 2.82% in the 8 ml L^{-1} application (Table 4).

4. Conclusion

The effects of different doses of humic acid, an essential source of organic matter, and different growing media on tomato plants growth and nutrient uptake were statistically significant in this study. Intensive use of chemical fertilizers in tomatoes, especially during the seedling period, increases the inputs and pollutes the environment. Today, different commercial growing media are used in the seedling industry. Commercial growing media both increase the cost and create a serious waste problem. In our study, tomato seedlings grown in hazelnut husk, an important waste material, showed weak growth. Cocopeat and peat+perlite media came to the fore regarding seedling quality and growth. While a positive relationship was found among the increase in humic acid doses and the plant growth, root development and total plant dry weight values reached the highest at the lowest humic acid dose. While the humic acid doses did not have an effect on growth in general, the results were obtained in the relationship between the media and humic acid (cocopeat and 8 ml L^{-1}). However, humic acid doses showed significant effects on flowering (8 ml L^{-1}), fruit set (4 ml L^{-1}), fruit phosphorus content (4 ml L^{-1}) and fruit potassium content (8 ml L^{-1}). In future studies, different mixtures of different media should be examined in order to determine the full effect of the growing media. In addition, it is thought that the continuation of the application of humic acid to the plant from leaves or soil after planting may significantly affect the results.

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RESEARCH PAPER

HORTICULTURAL STUDIES

Evaluation of the Relations between Yield and Yield Components of Tomato (*Solanum lycopersicum* L.) Hybrids by Correlation and Path Analysis

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Abstract

Tomato is one of the most produced vegetables in the world and there are many plant breeding studies that are carried out on this vegetables species. One of the most important aims of tomato breeding is the improvement of fruit quality and yield in both open-field and greenhouse growing conditions. The knowledge of factors with regard to yield is making plant breeders work easier. In the present study, the correlations of some plant characteristics thought to be related to yield and their direct and indirect effects on yield were analyzed. This study was conducted with 14 genotypes in 2020 and a randomized complete block design was employed as an experimental design. The relationship between 12 traits and yield was determined through path coefficient analysis. It was determined that the number of days from the first fluorescence to the first fruit set time, the length under the first cluster, fruit length, fruit diameter and Brix° value have a directly negative effects on the yield. However, fruit weight, fruit number, leaf diameter, and early yield have a directly positive effect on the yield. However, early yield had a directly positive effect on the yield. According to the result of this study, in the correlation matrix, the number of days from first fruit set time to fruit ripening, internode length, fruit diameter, leaf diameter, leaf length and number of fruit per plant are insignificant. The obtained results a potentially be utilized as selection criteria in the future studies on yield.

1. Introduction

Tomato (*Solanum lycopersicum* L.) is a member of the Solanaceae family including the most important crops such as potato, tobacco and pepper in terms of global trade and agricultural production. Moreover, the Solanaceae family includes more than 3000 species that contain both field crops and vegetables. One of the most important members of the Solanaceae family is tomato that is the most produced vegetable in the world. The most important property making tomato the most produced vegetable is its nutritional value that is rich in vitamin A, vitamin C, protein, fat, carbohydrates (Kabelka et al., 2004) and other nutritional elements including phenolic and antioxidants (Seçgin et al., 2018). Tomato also has a wide usage area as the most versatile vegetable.

Even if the origin of the tomato is South America, it is produced in a wide area in the world. Especially, China, India, Türkiye and USA are globally shining out for tomato production (FAO, 2021). Although Türkiye ranks third among tomato producers in the world, the yield value of tomatoes is not at the desired level. This circumstance directly affects profitability of producers and it is specifically becoming a limitation factor for small farmers.

The main objective of tomato breeding studies is to obtain high yielding hybrid tomato varieties in the different greenhouse conditions. In a breeding program for the optimum yield level, plant selection depends on the extent of the effect of factors related

to yield and the knowledge of the interaction of these factors with each other. The only consideration of correlation coefficient is not enough for selection in breeding studies. The correlation coefficient between two parameters is not sufficient to identify cause and effect. Sometimes, the relationship between two parameters may depend on another parameter. For this reason, it is necessary to make the relationship between the traits more understandable by separating the correlation coefficient between yield and yield components into direct and indirect effects and by revealing their proportional contribution on yield for an effective selection (Wright, 1934; Gravois and Helms, 1992). The path analysis is based on multiple regression analysis and path coefficients are standardized regression coefficients (Dewey and Lu, 1959; İkiz and Şengonca, 1978).

Alam et al. (2019) employed correlation and path analysis in their study with 23 tomato genotypes to determine 13 traits contributing to yield. In their study, correlation coefficients were determined for relationships among the traits. According to the result of this study, yield per plant (r=0.99), fruit weight (r=0.72), fruit diameter (r=0.67), number of carpel per fruit (r =0.67), and pericarp thickness (0.66) had positive and highly significant correlations with yield. They also studied cause effect relations among yield (t ha^{-1}) and its components through path coefficient analysis. Yield per plant was the most effective factor (1.018), and it was followed by number of flower per plant (0.212) and pericarp thickness (0.155). Fruit diameter (-0.279) had the most negatively direct effect on the yield, but it had positive correlation (r=0.67) with yield. Anuradha et al. (2018) carried out a study to analyze path coefficient and correlation of 13 traits that were related to yield with 40 tomato genotypes in 2017-2018 season. In their study, yield per plant, average fruit weight, yield per hectare, beta carotene and lycopene had highly positive correlations with yield. Moreover, plant height, number of primary branches per plant, number of day from sowing date to fruit set, number of fruit per plant, ascorbic acid and Brix° had significant negative correlation. Path analysis also revealed that some factors such as average fruit weight and number of fruit per plant had directly positive effect and also they had positive correlation with yield.

Kumar et al. (2013) studied on 26 tomato genotypes in India, which the tomato yield is below the world average. In their study, the correlation analysis demonstrated that number of fruit and cluster per plant were significant on yield. Path analysis revealed that fruit weight had the most positive direct effect on yield per plant, followed by number of fruit per plant, fruit diameter and number of fruit per cluster.

Sharma et al. (2019) carried out a study in 2015-2016 spring season with 27 tomato genotypes. Their study revealed that marketable fruit per plant, plant height, internode length and average fruit weight had a positive effect on yield and these criteria could be used as a selection criterion for high yield. According to the result of path coefficient analysis, number of marketable fruit per plant was the most effective parameter on yield and it was followed by average fruit yield and fruit shape index.

According to Tiwari and Apadhyay (2011), fruit weight was significant in terms of both correlation and path analysis on yield, and it can be used as a selection criterion in order to improvement of fruit yield.

Path coefficient analysis is widely used in order to determination of relationship between yield and yield components. Even if many studies have been conducted in another countries, there is no sufficient number of studies in Türkiye. Therefore, the present study on tomato breeding program was carried out to determine factors having significant effects on the yield by using correlation and path coefficient analyses.

2. Material and Methods

The present study was carried out in spring season of 2020 at Batı Akdeniz Agricultural Research Institute (BATEM) in Antalya, Türkiye whose location is at 36°C 928 N latitude and 30°C 982 E longitude and is 18 m above mean sea level. The soil structure is light and loamy.

In the present study, 12 candidate hybrid varieties improved by BATEM and 2 commercial hybrid varieties were used. Randomized complete block design (RCBD) was used as experimental design in 3 replications with 10 tomato plants in each replicate. In planting, inter row spacing (0.80 × 0.50 m) was 0.65 m and intra row was 0.60 m with double row planting. Cultural practices such as irrigation, pruning, weed management, and pesticide applications were carried out regularly.

Hybrid variety candidates and two commercial hybrid varieties used as control group were sown to plastic vials in the autumn period. Three weeks later, when the seedlings had 4-5 true leaves, the seedlings were planted in rows with 10 seedlings in each plot. Irrigation and fertilization were planned as twice a week. Sticky pheromone traps were used against plant disease and insects.

The harvest was started at third month and completed at 4 times. Observations were executed on 10 plants in each plot and consisted of 13 parameters. These parameters were; number of days to 50% flowering (NDFF), number of days from first flowering to first fruit set (NDFR), the length under the first cluster (SLFC), internode length (IL), fruit diameter (FD), fruit length (FL), leaf diameter (LD), leaf length (LL), average fruit weight (FW), average fruit number (NF), total yield of tomato plants (YP) early yield per plant tomato (EYP) and Brix° (According to UPOV criteria) (Table 1). The correlation coefficient was calculated by first

Table 1. The morphological observations methods used in the study.

No	Morphological observations	Explanation
1	Number of days to 50% flowering (NDFF)	Days to 50% flowering were determined by recording the number of days after transplanting (DAT) until 50% of plants in a plot had at least one open flower.
2	Number of days from sowing to first fruit set (NDFR)	The flowers of each plant in the plots were observed and the date of fruit set in half of the plants was recorded.
3	Stem length to first cluster (SLFC)	Flower cluster were observed in each plots and the distance between soil level and flower cluster was measured as cm.
4	First Internode length (IL)	Half of plants were observed in each plots. The internode above the first flower cluster was based on. The data were recorded as cm.
5	Fruit diameter measurement (FD)	Ten fruits were harvested from each genotype and fruit diameters were determined.
6	Fruit length (FL)	Ten fruits were harvested from each genotype and fruit lengths were determined.
7	Leaf diameter (LD)	On the 80th day after sowing, the diameters of the leaves at the 5th node from the top were measured with a ruler. Data were measured in cm.
8	Leaf length (LL)	On the 80th day after sowing, the length of the leaves in the 5th node from the top was measured with a ruler. Data were measured in cm.
9	Early yield per plant (EYP)	The data were obtained by addition of first two harvest values. It was recorded in grams by dividing by the number of plants in the plot.
10	Total yield per plant (YP)	Total weight of harvest was measure in each plot. It was recorded in grams by dividing by the number of plants in the plot.
11	Number of fruit (NF)	Harvested fruits were counted in each plots. These data were divided by number of plants in the plots and average number of fruits were determined.
12	Fruit weight (FW)	The harvested tomatoes were weighed. The average fruit weight was recorded as g by dividing by the number of plants in the plot.
13	Brix°	Digital refractometer was used a with the refractometric method (Gölükcü et al., 2018), (A. Krüss Optronic GmbH, DR6000 series, Germany).

determining the covariance of the variables and then dividing that quantity by the product of those variables' standard deviations. The coefficients were calculated by using a formula for correlation. Path coefficients were estimated according to Dewey and Lu (1959) and Singh and Chaudhary (1985), where yield (kg plot^{-1}) was kept as resultant variable and other contributing characters as causal variables. TARIST (version 5.0) computer software were used for correlation and Path analysis.

3. Results and Discussion

Fruit yield is a polygenic trait. Therefore, interaction among these genes and analyzing of their relationship with fruit yield are very important for selection criteria. For this reason, to find desired traits, plant breeders need to obtain large genetic diversity and variation among their breeding populations (Ritonga et al., 2018). Correlation coefficient among fruit yield per plant and its 12 component traits in all possible combinations are shown in Table 2.

Correlation analysis showed that fruit length (0.610*) and early fruit yield per plant (0.597*) had a positive effect on yield. The increases in these traits lead to significant increases in fruit yield per plant. Moreover, stem length to first cluster (SLFC) and number of days from sowing to first fruit flowering (NDFF) had a positive effect (0.702**) on fruit yield per plant as well. Furthermore, the result of the present study demonstrated that leaf surface (between leaf diameter and leaf length) is significant (0.968**). On the other hand, there were not only some traits that had positive effects on yield, but also some traits with negative effect. Firstly, number of days to 50% flowering (NDFF) had a negative effect (r: -0.576*). In addition, relationship between Brix^{o} value and yield per plant was inversely related (r:-0.569*). Furthermore, there was a negative relationship among fruit diameter and number of days from sowing to first fruit number (-0.664**), and internode length (-0.558*). Moreover, there was a negative correlation between number of fruit and internode length (-0.637*) and also early yield and number of days from sowing to first florescence number had a negative relationship with each other (-0.616*). According to result of the present study observed 12 observed components had positive or negative effects on the yield. While NDFR, IL, FL, FW, NF, FD and EYP have positive effect on the yield, NDFF, SLFC, LL, LD and have positive effect on the yield, NDFF, SLFC, LL, LD and Brix° had a negative effect on the yield. The results of traits effective on yield per plant on other traits are demonstrated in Table 3. Fruit weight (FW) (1.1543), number of fruit (NF) (0.4127), leaf length (LL) (0.1810) and earliness fruit yield (EYP) (0.0492) had a directly and positive effect on the yield. Fruit weight was in the positive correlation with YP, NDFR, IL and FL on fruit yield while it had negative correlation with other parameters. Its path coefficient was 1.154 and its correlation coefficient was 39%. The path coefficient of fruit number was 0.4127 and its correlation was really high with 16% rates. This parameter was in the positive correlation with SLFC (0.1535), NDFF (0.1487), Brix° (0.092) and EYP (0.0167). Leaf length was in the positive correlation with FD and FL while it was negative

Table 2. Correlation coefficient of characters contributing to yield in tomato.

Traits	NDFF	NDFR	SLFC	IL	FD	FL	LD	LL	FW	NF	Brix°	EYP
YP	-0.576*	0.502ns	-0.502ns	0.013ns	0.188ns	0.610*	-0.223ns	-0.283ns	0.187ns	0.485ns	-0.569*	0.597*
NDFF		0.091ns	0.702**	-0.532*	-0.664**	-0.309ns	0.345ns	0.286ns	-0.293ns	-0.025ns	0.429ns	-0.616*
NDFR			-0.202ns	-0.266ns	-0.065ns	0.310ns	0.147ns	0.115ns	0.327ns	0.320ns	-0.076ns	0.187ns
SLFC				-0.195ns	-0.558*	-0.004ns	0.079ns	0.052ns	-0.298ns	-0.152ns	-0.121ns	-0.489ns
IL					0.493ns	0.246ns	0.238ns	0.307ns	0.371ns	-0.637*	-0.165ns	0.156ns
FD						-0.088ns	-0.339ns	-0.270ns	0.673**	-0.444ns	-0.204ns	0.453ns
FL							0.046ns	-0.041ns	0.177ns	0.061ns	-0.519ns	0.175ns
LD								0.968**	-0.254ns	-0.189ns	0.530ns	-0.479ns
LL									-0.250ns	-0.225ns	0.531ns	-0.466ns
FW										-0.548*	-0.082ns	0.339ns
NF											-0.197ns	0.072ns
Brix°												-0.516ns

YP: Yield per plant, NDFF: The number of day from first sowing date to first florescence time, NDFR: The number of day from the first florescence to the first fruit set, SLFC: Stem length to first cluster, IL: Internode length, FD: Fruit diameter, FL: Fruit length, LD: Leaf diameter, LL: Leaf length, FW: Average fruit weight, NF: Number of fruit per plant, EYP: Early yield per plant.
*, **, and ns; significant at the $p < 0.05$, $p < 0.01$ level, and not significant, respectively.

Table 3. Effects of traits effective on yield per plant on other traits (Direct and indirect effects at levels of various component characters on yield of tomato).

No	Traits	Direct effects	Indirect effects											
			1	2	3	4	5	6	7	8	9	10	11	12
1	NDFF	-0.5070	-	-0.0503	-0.3615	0.4490	0.3659	0.0231	0.3130	0.0517	-0.3384	-0.0104	-0.4812	-0.0303
2	NDFR	-0.5511	-0.0463	-	0.1042	0.2247	0.0358	-0.0232	0.1332	0.0208	0.3775	0.1319	0.0852	0.0092
3	SLFC	-0.5147	-0.3560	0.1115	-	0.1641	0.3075	0.0003	0.0712	0.0094	-0.3441	-0.0628	0.1362	-0.0240
4	IL	-0.8434	0.2699	0.1468	0.1001	-	-0.2715	-0.0184	0.2159	0.0555	0.4278	-0.2627	0.1855	0.0077
5	FD	-0.5510	0.3367	0.0358	0.2872	-0.4156	-	0.0065	-0.3067	-0.0489	0.7766	-0.1834	0.2290	0.0223
6	FL	-0.0748	0.1567	-0.1707	0.0021	-0.2073	0.0482	-	0.0417	-0.0074	0.2049	0.0251	0.5824	0.0086
7	LD	0.9060	-0.1751	-0.0810	-0.0405	-0.2010	0.1865	-0.0034	-	0.1751	-0.2926	-0.0782	-0.5953	-0.0235
8	LL	0.1810	-0.1448	-0.0632	-0.0267	-0.2585	0.1490	0.0030	0.8768	-	-0.2885	-0.0928	-0.5957	-0.0229
9	FW	1.1543	0.1487	-0.1803	0.1535	-0.3126	-0.3707	-0.0133	-0.2297	-0.0452	-	-0.2263	0.0920	0.0167
10	NF	0.4127	0.0128	-1.7620	0.0784	0.5369	0.2449	-0.0046	-0.1716	-0.0407	-0.6328	-	0.2212	0.0035
11	Brix°	-1.1223	-0.2174	0.0419	0.0625	0.1394	0.1124	0.0388	0.4806	0.0960	-0.0947	-0.0813	-	-0.0254
12	EYP	0.0492	0.3125	-0.1032	0.2516	-0.1319	-0.2496	-0.0131	-0.4338	-0.0844	0.3912	0.0297	0.5787	-

NDFF: The number of day from first sowing date to first florescence time, NDFR: The number of day from the first florescence to the first fruit set, SLFC: Stem length to first cluster, IL: Internode length, FD: Fruit diameter, FL: Fruit length, LD: Leaf diameter, LL: Leaf length, FW: Average fruit weight, NF: Number of fruit per plant, EYP: Early yield per plant.

correlation with other parameters. Number of days from sowing to first fruit set (NDFR) had a negative effect on the yield. Its path coefficient was -0.507 and its the raito of direct effect to yield was 17%. These results were consistent with the results in the literature (Rasheed et al., 2017; Alam et al., 2019). Moreover, according to Ritonga et al (2018), fruit weight and number of fruit directly effect the yield and the result of the present study correspond to their study.

According to result of the present study, fruit weight (-0.5510, 17%) and fruit length (-0.0748, %4) had a negative path coefficient so it can be said that their effects were negative. On the contrary, fruit weight (1.1543, 39%) and fruit number (0.4127, 16%) were the most positively effective parameters on the yield. Even if fruit diameter had a negative effect on yield, fruit length did not have a similar effect. Fruit number (0.4127, 16%) had a positive effect on the yield. In this context, the result of this study is consistent with the result of Mohanty et al. (2003). On the other hand, some obtained results were not similar with the other studies. According to literature there is an ongoing debate on fruit length and diameter (Ritonga et al., 2018; Alam et al., 2019; Sanchez et al., 2019; Sharma et al., 2019, Singh et al., 2021). These results show that the value of the leaf diameter path coefficient was 0.0960 and its the raito of direct effect to yield was 32.8%. In addition, the path coefficient of the leaf length was found to be 0.1810 and the direct effect ratio was 6.6%. There is no previous study on the effects of leaf length and leaf diameter on yield. The path coefficient showing the direct effect of the average fruit weight on the yield was 1.154 and the ratio in the ratio of direct effect to yield was 39%, the

path coefficient for the number of fruit per plant were 0.427 and the ratio was quite high with 16%. Path and correlation coefficients that was used for demonstration of direct and indirect effect on yield show that the number of day from first sowing date to first florescence time had a negative effect on the yield. Its path coefficient was -0.507 and the ratio of direct effect to yield was 17%. These results were similar with the results of Rasheed et al. (2017) and Alam et al. (2019). Moreover, according to Ritonga et al. (2018), fruit weight and number of average fruit number per plant affect the yield directly. The results of the present study were similar with those of Ritonga et al. (2018).

According to result of the present study, fruit diameter (-0.5510, 17%) and fruit length (-0.0748, 4%) had a negative path coefficient so it can be said that they effect yield negatively. However, it is demonstrated that number of fruit (0.4127, %16) and fruit weight (1.1543, %39) were the most effective factors on the yield. Although increasing of fruit diameter has negative effect on the yield, fruit length has no similar effect on the yield. Number of fruit per plant (0.4127, %16) had a positive effect on the yield. In this context, this study has similar results with Mohanty et al. (2003). On the contrary, the results of this study are not similar with those of other studies. The literature on this topic clearly showed that there is an ongoing debate on aspect ratio of fruit, so some researchers say that it has a positive effect on yield, while others say that it has a negative effect on yield (Ritonga et al., 2018; Sharma et al., 2019; Sanchez et al., 2019; Alam et al., 2019; Sing et al., 2021).

According to results, the path coefficient of leaf diameter was 0.9060 and correlation coefficient was 32.8%. Furthermore, the path coefficient of leaf length was 0.1810 and its correlation coefficient rate was 6.6%. No other study was found on leaf diameter and leaf length.

The path coefficient of Brix° was -1.1223 and its the ratio of direct effect to yield was 44%. These results display clearly that Brix° had a negative effect on the yield. There are many studies that have similar results. For example, this value was -0.4098 in Reddy et al. (2013), -0.5027 in Anuradha et al. (2018), -0.26 in Alam et al. (2019), -0.19 in Sing et al. (2021) which were consistent with our results.

4. Conclusion

All observed parameters had a positive or negative effect on tomato yield. Results display that stem length to first cluster had a negative effect on yield. The result of fruit dimension and fruit weight demonstrate that as fruit volume increased, the number of fruit per plant decreased. The cumulative effect of fruit dimensions (fruit diameter and fruit length) and fruit weight can be seen in the fruit weight. Average fruit weight had a positive effect on the yield. In addition to this, when fruit volume increased, Brix° value decreased and this circumstances had a positive effect on the yield.

According to the result of this study, fruit weight and fruit dimension had a direct effect on the yield and other parameters had an indirect effect on the yield. Therefore, it can be said that the used parameters in this study can be used as a selection criterion in breeding program.

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RESEARCH PAPER

Determination of *Phytophthora* Species Causing Root and Crown Rot on Tomatoes Grown in Antalya Province and Reactions of Some Tomato Genotypes against *Phytophthora nicotianae*

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Abstract

Antalya province is the main center of vegetable production in Türkiye. Tomato comes first in terms of crops cultivated under greenhouse. *Phytophthora* species causing root and crown rot are among the factors negatively affecting tomato yield and quality. This research aimed to determine the prevalence of root and crown rot of tomatoes grown in Antalya province and to identify *Phytophthora* species causing disease. During surveys performed in 170 tomato greenhouses, plant and soil samples were taken from the areas where root and crown rot, stem blight, and drying symptoms were observed. Disease prevalence and incidence in the investigated greenhouses were 25.88% and 4.87%, respectively. *Phytophthora* symptoms were not observed in the greenhouses in Demre and Kepez districts, while the highest disease prevalence was found in Elmalı district with 75%. Eighty of 84 *Phytophthora* isolates were identified as *P. nicotianae* and four as *P. capsici*, according to their cultural, morphological and molecular characterisation. Virulence of 18 selected isolates were determined by using stem inoculation technique and all isolates caused lesions with different lengths on tomato stems. The reactions of 22 tomato genotypes in the gene pool of BATEM against *P. nicotianae* were also investigated and the genotype DT-15 was found as the most susceptible genotype with the largest lesions, while A-286 was the most resistant genotype. This study formed the basis for further studies on tomato breeding and integrated disease management.

1. Introduction

Tomato (*Solanum lycopersicum* L.), belonging to Solanaceae family, is an important vegetable both for fresh consumption and as an agricultural raw material. Tomato production constitutes about 55% of the total vegetable production of Antalya province and is an important source of income for the farmers (Karaköse et al., 2022). Plant diseases cause significant economic losses in agriculture and pose a major threat to global food security (Kroon, 2010). Pests and diseases can also cause yield and quality losses in tomato production worldwide. Tomato is susceptible to more than 200 diseases and yield losses can reach 70-95% (Lukyanenko, 1991; Ma et al., 2023).

Phytophthora genus has many pathogenic species and is among the most important plant pathogens all over the world (Erwin and Ribeiro, 2005; Brasier et al., 2022; Giachero et al., 2022). These species cause various destructive diseases on many plant species, from vegetable or ornamental plants to fruit and forest trees. Most species cause root or crown rot on plants (Agrios, 2005; Erwin and Ribeiro, 2005). Infected plants show drought and nutrient deficiency symptoms at the beginning, then the plants quickly weaken and become vulnerable to attack by other pathogens.

Phytophthora root and crown rot destroys its hosts in almost all parts of the world, having waterlogged soils with relatively low (15-23°C) temperatures (Agrios, 2005). Diseases caused by *Phytophthora* species have become more important with the increasing trade in plant materials, especially with the ornamental plant trade (Cacciola and Gullino, 2009; Ebrahimzadeh and Dolar, 2019).

Various studies showed that *P. nicotianae (=P. parasitica*), *P. capsici, P. cryptogea, P. arecae, P. citricola, P. mexicana, P. erythroseptica, P. cactorum*, *P. drechsleri* (Blancard, 2012) and *P. syringae* (Hyder et al., 2019) cause root and crown rot on tomatoes. *Phytophthora nicotianae* has a very wide host spectrum. Since its first description on tobacco was in 1896, it has been reported to cause root rot, crown rot, leaf blight, stem canker, tip blight and fruit rot on about 255 plant species from 90 families (Erwin and Ribeiro, 2005; Cline et al., 2008; Minuto et al., 2008; Gilardi et al., 2013; 2014; Gupta et al., 2022).

Tomato production, made in Antalya for many years, has become possible in all seasons with the increase of highland greenhouse cultivation in the province. Root and crown rots are the most important diseases threatening tomato production. These diseases caused by soil-borne pathogens are common in tomato growing areas. *Phytophthora* species are the most important group of agents related to root and crown rot. However, since their isolation is difficult and require special media, they cannot be isolated from diseased plants. Considering that the disease is caused by different pathogens, unnecessary fungicide applications are made. Thus, the disease cannot be controlled successfully. Studies on root and crown rot disease caused by *Phytophthora* species on tomato plants are very limited in Türkiye. Since they are not considered as significant pathogens of tomatoes, studies on the breeding of resistant cultivars against this group of pathogens have been neglected. No detailed research has been done on the reactions of tomato genotypes against *Phytophthora* species. In this study, surveys were performed in the tomato-growing areas of Antalya province and plant and soil samples were collected. *Phytophthora* species were isolated from the collected samples by using selective media and identified according to their morphological and molecular features. Virulence variations among species and isolates were also determined by pathogenicity tests and reactions of tomato genotypes against the most common species were determined.

2. Material and Methods

2.1. Field studies

Surveys were performed in Aksu, Alanya, Demre, Elmalı, Finike, Gazipaşa, Kaş, Kepez, Korkuteli, Kumluca and Serik districts of Antalya province, where tomato cultivation is common and over 500 hectares land, during 2019-2021 vegetation period. According to the simple random sampling method, selected greenhouses were examined for disease symptoms and root and crown rot prevalence and incidences were determined (Bora and Karaca, 1970). A total of 170 greenhouses were investigated and soil and plant samples with root and crown rot, wilting and drying symptoms were collected. Samples were brought to the Mycology Laboratory of the Plant Health Department of the Batı Akdeniz Agricultural Research Institute (BATEM) and investigated for the presence of *Phytophthora* species.

2.2. Isolation of *Phytophthora* species from plant and soil samples

The roots of the diseased plants were washed under tap water and small tissue pieces with lesions taken from the roots, crown and stem were directly transferred onto a selective medium (PARP-Corn meal agar amended with pimaricin, ampicillin, rifampicin and pentachloronitrobenzene) (Jeffers and Martin, 1986). Cultures were incubated at 20±1°C in the dark for 2-3 days. Colonies were then examined under a microscope and agar plugs with coenocytic mycelia were cut from the edges of the colonies and transferred to carrot juice agar (CA) (200 ml boiled carrot juice, 800 ml distilled water and 20 g agar) (Kurbetli et al., 2020). The baiting technique was used to isolate *Phytophthora* species from the soil samples. Green tomato fruits and fresh tomato leaves were used as traps. Small tissue pieces taken from the fruits and leaves with lesions were then transferred onto selective medium as mentioned above.

2.3. Identification of the *Phytophthora* isolates

The *Phytophthora* isolates were identified according to their colony types, and morphology and sizes of sporangia (Erwin and Ribeiro, 2005; Gallegly and Hong, 2008). Soil extract (1.5%) or rain water was used to induce sporangia formation (Jeffers, 2006) and carrot juice agar amended with β-sitosterol (30 mg L^{-1}) to induce a sexual structures (Latorre et al., 2001; Gallegly and Hong, 2008). The presence and morphology of hyphal swellings and chlamidospores were investigated in two week-old cultures. The growth of heterothallic species at 35°C on carrot agar and potato dextrose agar was investigated after 5-7 days of incubation. Colony morphologies of the isolates on different media (carrot agar, V8 juice agar, malt extract agar, cornmeal agar and potato dextrose agar) were also determined.

2.4. DNA isolation, PCR and DNA sequencing

Isolates were grown on carrot agar at 24±1°C in the dark for one week. Mycelia were taken by a

sterile scalpel, transferred to Eppendorf tubes and crushed using the TissueLyser instrument (Qiagen, Tokyo, Japan). DNA was extracted by using the Wizard Genomic DNA Kit (Promega, Madison, WI, USA) according to the manufacturer's instructions. DNA sequence of the internal transcribed spacer (ITS) regions of the isolates was amplified by PCR by using universal primers ITS4 (R 5' TCC TCC GCT TAT TGA TATGC 3') and ITS6 (F 5' GAA GGT GAA GTC GTA ACA AGG 3') (White et al., 1990; Kroon et al., 2004). DNA sequences of the PCR products were analyzed by BMLABOSİS (Ankara-Türkiye) and compared with the sequences listed in GenBank (NCBI-National Center for Biotechnology Information) to verify the morphological identifications of the isolates. DNA sequences were submitted to GenBank.

2.5. Pathogenicity test

Virulence of the *Phytophthora* species was determined by stem inoculation technique. A total of 18 isolates, 15 of which were *P. nicotianae* (Dalpem, Dalbel-1, Dtur-5, Dtur-6, Dakadlı-1, Dakadlı-2, Dakgökdere-2, Dal7-3, Dkare-1, Dkare-4, Dkumsyn, Dgazi, DSermer-1, DSermer-2, DKO 20-4) and 3 *P. capsici* (DTur-1, DTur-3, and DTur-4) were used in the test. Stems of tomato plants (cv. Batem Özçelik) with 6-7 leaves were decapitated by a sterile scalpel and 2.5 mm agar discs taken from one-week-old pathogen culture were placed on the injury. Then the inoculums were covered with aluminum foil to keep the inoculum on the stem and maintain humidity on the inoculation site. Sterile agar discs were used for control plants. Plants were incubated at 24±1°C and isolates causing lesions on tomato stems 5 days after inoculation were determined as pathogens. Lengths of the lesions were measured and small pieces taken from the lesions were transferred onto a selective medium to reisolate the pathogens (Pochard et al., 1976; Messaouda et al., 2015).

2.6. Determination of the reactions of tomato genotypes

To determine the susceptibilities of tomato genotypes against *Phytophthora* root and crown rot, 22 genotypes in the gene pool of BATEM (DT-7, DT-9, DT-15, DT-31, DT-50, DT-62, DT-90, DT-233, DT-253, DT-257, DT-284, DT-289, DT-296, DT-630, BH-4, TY-83, TY-84, YS-580, YS-583, FHG-470, A-286, Batem Özçelik) were used. *Phytophthora nicotianae* isolate Dakadlı-1, which was found as the most virulent isolate in the pathogenicity test, was inoculated to the genotypes by the stem inoculation technique. Plants were grown at 24±1°C in greenhouse conditions and lesion lengths were measured every 5 days after inoculations. In addition, necrosis progression (mm day^{-1}) was found by dividing the differences between two successive measurements by days between them (Sağır, 1984; Sağır and Yıldız, 1988) and statistically evaluated. Death started in susceptible genotypes 10 days post inoculation (dpi) due to the necrosis covering whole plant. Since the sensitivity levels of tomato genotypes began to dicriminate 10 dpi, genotypes were classified as resistant, moderately resistant, susceptible and very susceptible, based on the necrosis progressions in this period (Göçmen, 2006).

2.7. Statistical analyses

Obtained data were subjected to one-way ANOVA and means were compared by Tukey's HSD test using SPSS (Version 23.0) program.

3. Results and Discussion

During surveys, investigations were made in 170 tomato greenhouses and plant and soil samples from the areas with root and crown rot symptoms were studied. Mean disease prevalence was 25.88% and incidence was 4.87% in the surveyed greenhouses. There was no plant with disease symptoms in the greenhouses in Demre and Kepez districts, while disease prevalence and incidence were highest in Elmalı district (Table 1). In Elmalı and Korkuteli districts, tomato production is mainly

Table 1. Number of tomato greenhouses investigated in Antalya province, greenhouses infected with root and crown rot disease, disease prevalence and incidence rates.

Districts	Number of greenhouses surveyed	Number of greenhouses with disease symptoms	Disease prevalence (%)	Disease incidence (%)
Aksu	35	7	20.00	1.86
Alanya	10	3	30.00	3.08
Demre	11	-	-	-
Elmalı	12	9	75.00	4.88
Finike	10	2	20.00	4.55
Gazipaşa	7	1	14.29	0.12
Kaş	17	5	29.41	3.38
Kepez	10	-	-	-
Korkuteli	12	8	66.67	3.41
Kumluca	23	2	8.70	0.41
Serik	23	7	30.43	2.71
Total/Mean	170	44	25.88	4.87

performed in plateaus during summer and period of time is not sufficient for solarization and soil fumigation. The reason why disease prevalence was higher in these areas may be the lack of pre-plant applications preventing soil-borne pathogens. Results of the study made by Perez et al. (2004) supported this thought. They applied metham sodium and solarization in the seed beds after inoculations of *P. nicotianae* and *R. solani.* As a result of the experiment, disease symptoms were not observed in these seed beds, while high rates of pathogens were observed in the controls.

As a result of isolations, a total of 84 *Phytophthora* isolates, 53 of them from the plant and 31 from the soil samples, were obtained. Isolates were evaluated according to their cultural and morphological criterias and 80 of them were identified as *P. nicotianae (= P. parasitica)*, and the remaining 4 were *P. capsici.* Identification of *Phytophthora* species are rather difficult due to the morphological similarities among the species (Waterhouse et al., 1983). Therefore, identifications were supported by molecular techniques. As a result of the comparison of the ITS sequences of our isolates with those of the *P. nicotianae* and *P. capsici* isolates in the GenBank, they showed 99-100% similarity. Our isolates were registered in the GenBank with the accession numbers OQ415883 (*P. nicotianae*) and OQ415886 (*P. capsici*).

Phytophthora nicotianae and *P. capsici* were isolated both from the plant and soil samples in the same greenhouses as only species and found virulent in the pathogenicity test. However, *P. nicotianae* was more common with a 95.24% isolation rate. Our results are compatible with the previous findings reporting these species as pathogens affecting tomato (Kırbağ and Turan, 2006; Gilardi et al., 2013). Bolkan (1985) mentioned that *Phytophthora* root rot caused by *P. capsici* or *P. parasitica* was an important disease in tomato-growing areas in California, and *P. parasitica* was responsible for more than 85% of the disease. Similarly, Colla (2012) stated that *P. capsici* and *P. nicotianae* were commonly found in tomato areas, but *P. nicotianae* was more important. *P. nicotianae* was also reported as the dominant species in Brazil, Egypt, Tunisia and South Africa (Panabières et al., 2016). In Türkiye, symptoms of root and crown rot disease caused by *P. nicotianae* have especially been observed during the early season in tomato areas and the damage caused by this pathogen has constantly been increasing (Altın et al., 2018). Some researchers noted that the root infections related to *P. nicotianae* were more severe in summer and early autumn (Alvarez et al., 2009). However, some others indicated that the pathogen caused severe epidemics on grafted tomatoes especially spring and summer months (Minuto et al., 2008; Garibaldi and Gullino, 2010). *Phytophthora nicotianae* susceptibility was observed on rootstocks of *S. lycopersicum* × *S. hirsutum* and other *S. lycopersicum* hybrids (Gilardi et al., 2011). In our surveys, the pathogen was similarly isolated from the grafted tomato plants.

As a result of necrosis length measurements made at 5 dpi, it was observed that the pathogen started to reproduce and form necrotic areas in the plant tissues. Due to necrosis covering the whole plant, death started at 10 dpi, and on the 20th day, necrosis covered all over the stems and susceptible plants died. However, the necrosis progression rate slowed or stopped in the resistant and moderately resistant genotypes. Differences among the genotypes in terms of mean necrosis lengths, measured during 20 days, were statistically significant (Table 2).

DT-15 was found as the most susceptible tomato genotype with the largest lesions, while A-286 was the most resistant one. Necrosis progression was also higher on the genotype DT-15 in the first 10 dpi, then it slowed and stopped after 15 days (Table 3). Similar to the necrosis lengths, daily necrosis progression was also slower on genotype A-286, especially during the first 10 dpi.

According to the classification made at 10 dpi, when the sensitivity levels of tomato genotypes began to discriminate, DT-9 and DT-15 genotypes were classified as highly susceptible, while Batem Özçelik genotype was susceptible. DT-7, YS-583, FH6-470 and A-286 genotypes were classified as resistant, and the remaining genotypes were moderately resistant (Table 4).

Control of soil-borne pathogens like *Phytophthora* species is rather difficult, since they can survive in soil and have wide host ranges. Chemical control has negative side effects on the environment and human health, while physical methods are more expensive and laborius (Ma et al., 2023). Therefore, the development of resistant cultivars against *Phytophthora* root and crown rot disease will make a significant contribution to tomato breeders and growers (Bolkan, 1985). In fact, it is focused on the selection and breeding of resistant cultivars against the disease, in many countries. However, disease symptoms and genetic factors controlling pathogen resistance can vary depending on the region and the virulence of the pathogen (Naegele, 2013).

4. Conclusion

This research showed that root and crown rot diseases caused by *Phytophthora* species are common in the tomato greenhouses in Antalya province, with the highest prevalence rate in Elmalı district. As a result of the isolations made from the plant and soil samples, *P. nicotianae* was found to be the most common agent causing the disease. A greenhouse trial showed differences among the resistance levels of 22 tomato genotypes found in the gene pool of BATEM, against the pathogen. The results obtained in this study will contribute to the integrated management of the disease by

Table 2. Mean lesion lengths on tomato genotypes 5-20 days after inoculation with *Phytophthora nicotianae*.

Tomato genotypes	Lesion length (mm)			
	5th day	10th day	15th day	20th day
DT-7	20.30 ce*	30.20 df	36.70 fh	41.60 eg
DT-9	35.27 a	80.47 a	87.73 a	87.73 a
DT-15	35.67 a	81.40 a	94.91 a	94.91 a
DT-31	32.07 ab	52.00 bc	65.73 b	72.73 b
DT-50	31.53 ab	48.60 bc	58.80 be	65.93 bd
DT-62	28.20 abc	47.73 bc	56.60 be	61.27 bd
DT- 90	31.27 ab	51.00 bc	58.27 be	63.47 bd
DT- 233	27.53 ad	43.53 bd	50.60 cf	56.20 ce
DT-253	31.13 ab	51.47 bc	63.00 bd	71.87 b
DT-257	30.13 ab	50.33 bc	62.33 bc	72.60 b
DT-284	33.53 a	49.27 bc	60.13 be	69.60 bc
DT-289	24.13 bd	40.40 ce	49.60 df	55.87 ce
DT- 296	28.87 ab	47.67 bc	63.00 bd	72.67 b
DT-630	29.67 ab	46.20 bc	59.07 be	68.20 bc
BH-4	31.07 ab	47.67 bc	59.20 be	67.07 bd
TY-83	31.07 ab	48.47 bc	60.60 be	67.40 bd
TY-84	31.27 ab	50.07 bc	63.40 bd	71.53 b
YS-580	29.80 ab	48.13 bc	62.20 bd	72.33 b
YS-583	24.60 bd	39.40 ce	46.80 eg	52.93 df
FH6-470	19.27 de	27.33 ef	34.47 gh	39.80 fg
A-286	15.13 e	23.13 f	30.13 h	35.13 g
BATEM Özçelik	30.93 ab	56.00 b	64.40 bc	70.87 b

* Means in the same column shown by the same letters are not statistically different from each other according to Tukey test (P=0.05).

Table 3. Necrosis progression on the tomato genotypes measured for 20 days in 5-day intervals following inoculation with *Phytopthora nicotianae*.

Tomato genotypes	Necrosis progression (mm day^{-1})			
	0-5 days	5-10 days	10-15 days	15-20 days
DT-7	4.05	1.99	1.29	0.99
DT-9	7.05	9.04	1.45	0.00
DT-15	7.13	9.15	2.70	0.00
DT-3	6.41	3.99	2.75	1.40
DT-50	6.31	3.41	2.04	1.43
DT-62	5.64	3.91	1.77	0.93
DT- 90	6.25	3.95	1.45	1.04
DT- 233	5.51	3.20	1.41	1.12
DT-253	6.23	4.07	2.31	1.77
DT-257	6.03	4.04	2.40	2.05
DT-284	6.71	3.15	2.17	1.89
DT-289	4.83	3.25	1.84	1.25
DT- 296	5.77	3.76	3.07	1.93
DT-630	5.93	3.31	2.57	1.83
BH-4	6.21	3.32	2.31	1.57
TY-83	6.21	3.48	2.43	1.36
TY-84	6.25	3.76	2.67	1.63
YS-580	5.96	3.67	2.81	2.03
YS-583	4.92	2.96	1.48	1.23
FH6-470	3.85	1.61	1.43	1.07
A-286	3.03	1.52	1.40	1.00
BATEM Özçelik	6.19	5.01	1.68	1.29

preventing unnecessary fungicide use, and will ensure the protection of environment and human health.

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Table 4. Susceptibilities of the tomato genotypes according to the necrosis regression rates on the 10th day after inoculation.

Tomato genotypes	Mean necrosis regression rate (mm day^{-1})	Sensitivity level
DT-7	1.99	Resistant
DT-9	9.04	Highly susceptible
DT-15	9.15	Highly susceptible
DT-31	3.99	Moderately resistant
DT-50	3.41	Moderately resistant
DT-62	3.91	Moderately resistant
DT- 90	3.95	Moderately resistant
DT- 233	3.20	Moderately resistant
DT-253	4.07	Moderately resistant
DT-257	4.04	Moderately resistant
DT-284	3.15	Moderately resistant
DT-289	3.25	Moderately resistant
DT- 296	3.76	Moderately resistant
DT-630	3.31	Moderately resistant
BH-4	3.32	Moderately resistant
TY-83	3.48	Moderately resistant
TY-84	3.76	Moderately resistant
YS-580	3.67	Moderately resistant
YS-583	2.96	Resistant
FH6-470	1.61	Resistant
A-286	1.52	Resistant
BATEM Özçelik	5.01	Susceptible

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RESEARCH PAPER

The Effect of the Application of Mycorrhiza on Vegetative Growth, Mineral Element Intake, and Some Biochemical Characteristics of Strawberry Seedlings under Lime Stress

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Abstract

This study aims to determine the effects of vesicular–arbuscular mycorrhiza (VAM) applications on vegetative growth, mineral element intake, and some biochemical characteristics of strawberry seedlings grown under lime stress conditions. The experiment was conducted with frigo seeds of "Albion" strawberry cultivar in pots filled with 1% lime mixture and 1:1 ratio of peat and perlite. In the uprootings performed in three different stages (four leaved, blooming, and fruit stages) to examine the biochemical effects of mycorrhiza applications against the lime stress, vegetative growth criteria (leaf chlorophyll and anthocyanin content, area, crown diameter, fresh and dry plant weights) and mineral contents in the plant parts (leaf, crown, and root) were determined. The proline, total phenolic content, and malondialdehyde (MDA), end product of the lipid peroxidation, analyses were conducted on the leaf samples taken in these uprooting. In all three stages, an increase in crown diameter and leaf area was determined. In uprooting periods, proline and total phenolic amounts increased, and, on the other hand, MDA decreased. Microelement intake, which decreased with the lime application, was detected to be increased with mycorrhiza applications. At the end of the experiment, mycorrhiza application was observed to lessen the effect of lime stress on strawberry seedlings.

1. Introduction

Strawberry (*Fragaria* × *ananassa* Duch.) of the Fragaria kind, the Rosaceae family, is grown in many parts of the world. Due to its taste, health benefits, and suitability for food technology, the production amount has been increasing year by year. The fact that it has no marketing issues encourages growers to strawberry cultivation. While the highest yield is obtained in temperate climatic conditions, the yield decreases as becoming distant from the sea, towards the upcountry (Yılmaz et al., 2006; Balcı et al., 2017). In upcountry areas where the continental climate is dominant, high soil pH is one of the most significant limiting factors for strawberry cultivation. Strawberries are susceptible to high pH level. When cultivation is performed in these areas, severe chlorosis and a decrease in vegetative growth are observed (Yılmaz, 2009).

Mycorrhizal fungi maintain their lives in plant roots by locating in the plant's root surface, root fibers, cells, and intercellular spaces. Mycorrhizal fungi are beneficial to soil microorganisms and are of great importance for healthy plant growth and soil fertility. The role of these fungal species, which play a very important role in 85% of the world's vegetation and establish symbiotic relationships with roots, in agricultural production is rapidly getting stronger day by day (Erzurumlu and Karar, 2014). Many studies demonstrate that mycorrhizal applications have beneficial effects for plants against to biotic or abiotic stress conditions

(Bavaresco and Fogher, 1995; Gianinazzi and Schuepp, 1995; Yano and Takaki, 2005; Sinclair et al., 2014; Latef et al., 2016). Strawberries, which have an important place in world fruit growing, are generally considered among the varieties that respond positively to mycorrhizae. The studies conducted to analyze the effects of the use of mycorrhiza on the growth and yield of strawberries have revealed positive effects in terms of plant development, fruit quality, and early yield in strawberries (Sharma and Adholeya, 2004; Ertan et al., 2007; Bayozen and Yıldız, 2008; Cekic and Yılmaz, 2011). While there are studies stating that mycorrhiza applications positively support vegetative growth, yield, and mineral substance intake in strawberries under stress conditions such as low pH, salinity, high phosphorus content in the soil, and drought (Gupta and Krishnamurthy, 1996; Stewart et al., 2005; Matsubara et al., 2009; Borowicz, 2010; Sinclair et al., 2014; Koç et al., 2016), very few studies examining the effects of mycorrhiza application on biochemical characteristics in the strawberries cultivated under stress conditions (Koç, 2015; Koç et al., 2016; Bahmanbiglo and Eshghi, 2021). There are not enough studies on the effects of mycorriza applications on strawberry under high pH conditions. This study aimed to determine the effects of mycorrhiza applied on strawberries in high pH conditions on vegetative growth, biochemical characteristics, and mineral element intake.

2. Material and Method

This study was carried out in a greenhouse without climate control, located on the field of Yozgat Bozok University (1111 m, 39°35'7" N and 35°09'35" E). In our experiment, the "Albion" cultivar, which is one of the day-neutral strawberries, was used. Albion, which is very productive and high quality, is successfully grown in areas with high altitudes (Balcı et al., 2017). The experiment was carried out by filling the peat perlite mixture at the rate of 1:1 in the 5-liter of pots (265×210 mm). Lime ($CaCO_3$) addition was not done to the pots in the 0% and 1% lime was added to the group to which lime stress was applied in terms of weight. The initial and final pH of the cultivation media in the experiment was determined as 7.74 and 8.41, respectively. The pH values of the environment were determined according to Kacar (2012). The frigo seedlings belonging to the Albion strawberry cultivar were planted in the pots on 28.03.2018. The strawberry seedlings were once fertilized with "Nutritect 18-18-18 TE" commercial fertilizer (15.05.2018).

2.1. Application of mycorrhiza

Preparate in commercial powder form containing mycorrhiza (9 different *Glomus* species) at the rate of 23%, named Endo Roots Soluble, belonging to the Bioglobal firm was used in our study. The fungi and their rates in the content of the preparate were as *Glomus intraradices* (21%), *Glomus aggregatum (*20%), *Glomus mosseage (*20%), *Glomus clarum (*1%), *Glomus monosporus* (1%), *Glomus deserticola (1%*), G*lomus brasilianum (*1%), *Glomus etunicatum (*1%), and *Gigaspora margarita (*1%). Mycorrhiza application to seedlings was performed by soaking plant roots in the solution, which was prepared by mixing the packet of 250 gram-powder with the 10-liter sugared water, for an hour before the planting and inoculating with the fungi. The remaining solution was put in the plant root area as sap. The plants in the control group, on the other hand, were kept in the water for an hour.

2.2. Taking leaf samples

Leaf sampling was performed in three different periods with the aim to determine the VAM effect on lime stress in strawberry seedlings in different development stages. The first leaf sample was taken in the period when the strawberry seedlings had 4 leaves (25.04.2018), the second sample was taken in the blooming period two months after the planting (25.06.2018), and the last sample was taken three months after the planting in the fruiting stage (26.0.2018). Vegetative growth parameters and mineral element contents were determined by uprooting plants in these periods. For mineral element contents analysis, the plants were separated into roots, crowns, and leaves with petioles, and washed. These parts of the plant were oven-dried at 70°C. For biochemical analysis, the leaves that had taken their full size during these periods were cut off, then immediately placed in ice, and kept at -20°C until the analysis. Mineral element analysis was made by the Yozgat Bozok University Application and Research Center of Science and Technology (BILTEM).

2.3. Evaluated criteria

In three different periods, on the leaves just before the removal, the chlorophyll content (SPAD) was determined with the chlorophyll meter (Konica Minolta SPAD-502 Plus Brand Chlorophyll Meter model) and the anthocyanin content (ACI) with the anthocyanin meter (Opti Science ACM-200 Plus Anthocyanin Meter model). After the plants were removed, the leaf area (cm^2 $plant^{-1}$) was determined using the ADC BioScientific Area Meter AM 300 model leaf area meter. Crown diameter (mm) using a digital compass was determined. Leaf fresh and dry weights were determined on a precision balance. For mineral elements, in leaf samples, extraction was prepared according to Falandysz et al. (2001). Mineral elements were determined by using the ICP-MS (ICAP-QC). The amount of proline was calculated according to the method of Bates et al. (1973) and the results were given as

nmol proline g^{-1} (fresh weight). The total phenolic amount was determined according to Singleton and Rossi (1965) by using the Folin Ciocalteu Colorimetric method and the results were given in gallic acid equivalent (mg g^{-1}). The lipid peroxidation was calculated according to Yong et al. (2008) and the calculated results were given as µmol g^{-1} in fresh weight.

2.4. Evaluation of the data

The experiment was set up with three repeats (10 plants in each repeat), two applications (0% and 1% lime application), according to the experimental design in the randomized parcels. For calculating the averages of all data obtained during the research, "Microsoft Office XP EXCEL" was used and the statistical analysis of applications was performed using a t-test, and Cohen's d effect size analysis techniques were used. The d value expressing the effect size was evaluated as 0.20-0.50 small, 0.51-0.80 medium, 0.81-1.00 large (Cohen, 1988). Interactions one ANOVA test SPSS 20.0 package. As a result of the statistical analysis, Duncan Multiple Range Test (Duncan Multiple Comparison Test) was applied by using the same package program to determine the difference between the media. The significance level between the differences in the statistical evaluation of the results was determined as 0.05.

3. Results and Discussion

3.1. Vegetative growth parameters

Vegetative growth parameters were measured in mycorrhizal strawberries exposed to lime stress. While the statistical effect of mycorrhiza and lime applications was insignificant in all three removals, the VAM × lime interactions were significant (Table 1). VAM applications were observed to have positive effects on the leaf area. In every uprooting, the leaf areas decreased with the effect of lime stress (Table 1). VAM application was detected to support the growth of leaf area in plants exposed to lime stress. In a study, of strawberries and pistachio, in which VAM was applied and which were exposed to drought stress, the leaf area has been reported to be positively affected (Borkowska, 2002; Abbaspoura et al., 2012). The thickest crown diameter was measured in mycorrhiza-applied plants (Table 1). Although a decrease in crown diameter was observed in plants exposed to lime stress, VAM applications reduced the severity of these decreases. The negative effect of lime stress on leaf weights can be seen clearly (Table 1). When the fresh weights of the leaves were examined, mycorrhiza applications were observed to increase the fresh weight of the leaves (except for the first uprooting) (Table 1). A similar effect was seen in the dry leaf weights.

The chlorophyll and anthocyanin contents obtained in three extractions are given in Table 2. VAM application on chlorophyll and anthocyanin content in the first removal was found to be insignificant. It was determined that the lime application had a small effect in terms of chlorophyll (d=0.48) and anthocyanin contents (d=0.39). The VAM × lime interaction was significant. (Table 2). In the removal made during the flowering period, while the VAM application had a moderate (d=0.52) effect on chlorophyll content, the lime application was ineffective. In anthocyanin content, while the VAM application was ineffective, the lime application showed a small effect (d=0.30). Considering the VAM × lime interaction, it was found to be significant in the chlorophyll content and insignificant in the anthocyanin content. In the final removal, when the chlorophyll content was examined, it was determined that the VAM application had a small (d=0.29) effect, while the lime application was ineffective. On the anthocyanin content, the VAM application was found to have a small (d=0.27) effect on lime application. The VAM × lime interaction was very important in both examined parameters.

In all three uprootings, the highest content of chlorophyll substances was detected in mycorrhiza-applied plants (Table 2). While there was a

Table 1. The effects of VAM on the some vegetative growth criteria of strawberry under calcareous conditions.

Application		Beginning of flowering				Flowering				Harvesting			
		LA (cm^2)	CD (mm)	FW (g)	DW (g)	LA (cm^2)	CD (mm)	FW (g)	DW (g)	LA (cm^2)	CD (mm)	FW (g)	DW (g)
Mycorrhiza	VAM	155	10.5	7.1	1.9	248	10.8	7.8	4.7	294	12.5	10.0	3.2
	N-VAM	121	9.6	6.0	1.4	200	8.5	6.3	3.9	248	11.1	10.5	3.7
$CaCO_3$	0% $CaCO_3$	148	10.8	6.2	1.6	254	10.3	7.5	4.8	293	11.9	11.4	3.7
	1% $CaCO_3$	128	9.3	6.9	1.8	195	9.1	6.6	3.8	250	11.7	9.1	3.2
VAM×$CaCO_3$	VAM×0% $CaCO_3$	162 a	11.6 a	7.6 ns	2.1a	293 a	11.2 a	8.7 a	5.4 ns	307 a	13.1 a	11.8 a	5.4 a
	VAM×1% $CaCO_3$	135 bc	9.4 b	6.5	1.7b	204 b	9.3 ab	6.4 b	3.9	278 b	11.2 b	11.0 ab	3.9 c
	N-VAM×0% $CaCO_3$	149 b	10.0 b	6.1	1.5b	215 ab	10.5 ab	7.0 b	4.1	281 ab	12.8a	11.1 a	4.1 b
	N-VAM×1% $CaCO_3$	107 c	9.2 b	5.9	1.4b	186 b	7.7 c	6.1 b	3.6	219 c	11.0 b	8.2 c	3.6 c
Significance													
VAM		Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
$CaCO_3$		Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns
VAM×$CaCO_3$		*	*	Ns	*	*	*	*	Ns	*	*	*	*

LA: Leaf area, CD: Crown diameter, FW: Leaf fresh weight, DW: Leaf dry weight, VAM: Vesicular–arbuscular mycorrhiza, N-VAM: Non vesicular–arbuscular mycorrhiza, $CaCO_3$: Lime

* Significant at the $p < 0.05$ level, Ns: Not significant. Mean followed by different letters within columns differ significantly ($p < 0.05$).

Table 2. The effects of VAM on the chlorophyll and anthocyanin content of strawberries under calcareous conditions.

Application		Beginning of flowering		Flowering		Harvesting	
		Chl (SPAD)	Ant (ACI)	Chl (SPAD)	Ant (ACI)	Chl (SPAD)	Ant (ACI)
Mycorrhiza	VAM	45.2	9.8	48.5	9.6	43.2	8.3
	N-VAM	42.9	8.1	46.1	9.3	39.8	9.5
$CaCO_3$	0% $CaCO_3$	45.9	9.9	47.8	9.7	43.8	9.0
	1% $CaCO_3$	42.1	8.0	46.8	9.1	39.1	8.8
VAM×$CaCO_3$	VAM×0% $CaCO_3$	47.3 a	11.2 a	48.8 a	10.0 ns	46.8 a	10.0 a
	VAM×1% $CaCO_3$	43.0 bc	8.4 b	46.2 ab	9.1	39.7 bc	8.5 b
	N-VAM×0% $CaCO_3$	44.5 ab	8.6 b	46.8 ab	9.5	40.9 b	9.1 a
	N-VAM×1% $CaCO_3$	41.2 c	8.0 b	45.5 b	9.0	38.7 c	8.2 b
Significance							
VAM		Ns	Ns	**	Ns	*	Ns
$CaCO_3$		*	*	Ns	*	Ns	*
VAM×$CaCO_3$		*	*	*	Ns	**	**

Chl: Chlorophyll; Ant: Anthocyanin, VAM: Vesicular–arbuscular mycorrhiza, N-VAM: Non vesicular–arbuscular mycorrhiza, $CaCO_3$: Lime
* Significant at the $p < 0.05$ level, ** Significant at the $p < 0.01$ level, Ns: Not significant. Mean followed by different letters within columns differ significantly ($p < 0.05$).

decrease in chlorophyll content with lime stress, the VAM application to plants exposed to stress was observed to significantly preserve their chlorophyll content as well. In their study, Akay and Kararslan (2012) stated that mycorrhiza applications contributed to the chlorophyll content of the plants, especially in poor soils.

When the anthocyanin content in the leaf was examined, mycorrhiza applications were observed to increase the anthocyanin content (except for the second uprooting). The anthocyanin content (Farrant, 2000; Johnston et al., 2007), known for protecting the chlorophyll structure, increased with mycorrhiza application, in line with the chlorophyll content (Table 2).

3.2. Effects on some biochemical characteristics

Some biochemical parameters obtained as a result of the experiment are given in Table 3. At the beginning of flowering, the VAM application had a small effect (d=0.39) on proline content, while the lime application had no effect. When the MDA content was examined, while the VAM application was moderately effective (d=0.66), the lime application was ineffective. In total phenolic substance content, while the VAM application was ineffective, the lime application was moderately effective (d=0.76). The VAM × lime interaction was very important in the investigated parameters. When evaluated on the second removal date, the VAM application had a small (d=0.34 and d=0.35) effect on proline and MDA contents, while it had a moderate (d=0.52) effect on total phenolic substance content. The lime application had little effect on proline and total phenolic content (d=0.39 and d=0.33, respectively) while it had no effect on MDA content. The VAM × lime interaction was very significant in all criteria examined. While no effect was observed on the proline and MDA content of the VAM application during the harvesting period, the effect on the total phenolic content was large (d=0.93). While the effect of lime application on proline and total phenolic substance application was not determined, its effect on MDA content was large (d=0.88). During this uprooting period, the VAM × lime interaction was very important for the evaluated parameters.

Mycorrhiza applications were determined to have significant effects on the amount of the proline throughout the experiment (Table 3).

In the first uprooting period, the highest proline amount (50 nmol proline g^{-1}) was obtained from the mycorrhiza-applied in the control group (Table 4). In the second and third uprooting periods, the highest amount of proline was detected in the mycorrhiza-applied plants in lime stress (30 and 70 nmol proline g^{-1}, respectively). When all experimental groups were evaluated in all uprooting periods, mycorrhiza applications were observed to increase the amount of proline in both groups. Proline, the amount of which is increased in stressful environments in the plant cell (Chen and Murata, 2002; Vardharajula et al., 2011; Cetin and Daler, 2017), is one of the significant osmolites in abiotic stress conditions (Rontein et al., 2002; Aktaş and Akça, 2015). Data we obtained in our research are compatible with many studies reporting that mycorrhiza applications applied in various stress conditions increase the amount of proline in different plants (Krishna et al., 2006; Campanelli et al., 2013; Hazzoumi et al., 2015; Latef et al., 2016).

The mycorrhiza applications were observed to have significant effects on the total phenolic content throughout the experiment (Table 3). When data were examined, the phenolic contents were observed to increase depending on stress in all uprooting periods. The mycorrhiza applications were also detected to positively affect the increase in phenolic content in strawberry seedlings (Table 3).

In plants under stress conditions, phenylpropanoid biosynthesis increases by the means of the PAL enzyme, and many secondary metabolites including phenolic compounds are synthesized (Koç, 2015; Pešaković et al., 2016; Aviova et al., 2017; Cetin and Daler, 2017). There are studies reporting an increase in phenolic compounds when mycorrhiza-applied strawberry

Table 3. The effects of VAM on some the biochemical criteria of strawberry under calcareous conditions.

Application		Beginning of flowering			Flowering			Harvesting		
		P nmol g^{-1}	MDA µmol g^{-1}	TP GAEmg g^{-1}	P nmol g^{-1}	MDA µmol g^{-1}	TP GAEmg g^{-1}	P nmol g^{-1}	MDA µmol g^{-1}	TP GAEmg g^{-1}
Mycorrhiza	VAM	40.0	0.67	3.4	30.0	1.2	5.3	60.0	2.4	4.1
	N-VAM	30.0	1.6	3.4	20.0	2.1	3.4	50.0	3.7	3.6
$CaCO_3$	0% $CaCO_3$	30.0	1.1	3.1	20.0	2.0	3.6	60.0	0.75	3.9
	1% $CaCO_3$	30.0	1.2	3.8	30.0	1.3	5.1	60.0	5.4	3.9
VAM×$CaCO_3$	VAM×0% $CaCO_3$	50.0 a	0.53 b	3.0 b	20.0 b	1.2 b	5.0 b	60.0 ab	0.75 c	4.2 a
	VAM×1% $CaCO_3$	30.0 b	0.98 ab	3.9 a	30.0 a	1.5 b	5.6 a	70.0 a	0.75 c	4.1 a
	N-VAM×0% $CaCO_3$	20.0 b	1.4 a	3.1 b	20.0 b	1.5 b	2.2 d	60.0 ab	4.1 b	3.6 b
	N-VAM×1% $CaCO_3$	30.0 b	1.4 a	3.7 a	20.0 b	2.7 a	4.6 c	40.0 b	6. 7 a	3.7 b
Significance										
VAM		*	**	Ns	*	*	**	Ns	Ns	***
$CaCO_3$		Ns	Ns	**	*	Ns	*	Ns	***	Ns
VAM×$CaCO_3$		**	*	**	**	**	**	**	**	*

P: Proline, MDA: Malondialdehyde, TP: Total phenolic, VAM: Vesicular–arbuscular mycorrhiza, N-VAM: Non vesicular–arbuscular mycorrhiza, $CaCO_3$: Lime
* Significant at the p < 0.05 level, ** Significant at the p < 0.01 level, *** Significant at the p < 0.001 level, Ns: Not significant. Mean followed by different letters within columns differ significantly (p < 0.05).

Table 4. The effects of VAM on some mineral element content of strawberry under calcareous conditions in leaves.

Application		Beginning of flowering					Flowering					Harvesting				
		Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)	Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)	Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)
Mycorrhiza	VAM	2.8	0.11	0.66	300	12.6	2.4	0.16	0.46	224	21.1	2.5	0.12	0.41	316	25.7
	Non VAM	2.7	0.03	0.55	245	9.3	2.3	0.12	0.47	224	20.3	2.5	0.09	0.43	262	14.0
$CaCO_3$	0% $CaCO_3$	2.2	0.08	0.61	320	12.9	2.2	0.20	0.49	244	27.2	2.2	0.16	0.41	314	24.0
	1% $CaCO_3$	3.3	0.06	0.60	225	9.0	2.4	0.08	0.44	205	14.2	2.8	0.05	0.43	265	15.7
VAM×$CaCO_3$	VAM×0% $CaCO_3$	2.5 c	0.16 a	0.74 a	340 a	13.8 a	2.5 a	0.23 a	0.49 a	264 a	26.6 a	2.0 d	0.18 a	0.46 a	358 a	29.6 a
	VAM×1% $CaCO_3$	3.0 b	0.04 b	0.58 c	259 c	11.3 b	2.3 b	0.09 c	0.43 b	222 b	15.6 b	2.9 a	0.07 c	0.35 c	269 b	21.9 b
	N-VAM×0% $CaCO_3$	1.8 d	0.08 b	0.63 b	299 b	11.9 b	2.0 c	0.17 b	0.50 a	226 b	27.8 a	2.4 c	0.15 b	0.46 a	275 b	18.5 c
	N-VAM×1% $CaCO_3$	3.5 a	0.02 b	0.48 d	191 d	6.7c	2.5 a	0.07 d	0.44 b	187 c	12.8 c	2.7 b	0.03 d	0.40 b	255 b	9.5 d
Significance																
VAM		Ns	**	Ns	Ns	*	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	*	**
$CaCO_3$		**	Ns	Ns	**	**	Ns	***	***	**	***	**	***	Ns	*	*
VAM×$CaCO_3$		**	**	**	**	**	**	**	**	**	**	**	**	**	**	**

Ca:Calcium, P:Phosphate, Mg:Magnesium, Fe:Iron, Zn:Zinc, VAM:Vesicular–arbuscular mycorrhiza, N-VAM:Non vesicular–arbuscular mycorrhiza, $CaCO_3$:Lime
* Significant at the p < 0.05 level, ** Significant at the p < 0.01 level, *** Significant at the p < 0.001 level, Ns: Not significant. Mean followed by different letters within columns differ significantly (p < 0.05).

seedlings are exposed to different stresses (Koç, 2015).

The mycorrhiza application in lime stress was detected to have a significant effect on the total MDA content throughout the experiment (Table 3). When data were examined, while the amount of MDA increased in stress conditions, mycorrhiza applications were observed to decrease significantly the amount of the MDA content. When plants are exposed to stress, first of all, the effects of malondialdehyde (MDA), one of the end products of lipid peroxidation, on membranes are clearly observed (Hodges et al., 1999), and it is known that MDA content increases when exposed to stress in almost all plants (Krupa et al., 1986; Quarti et al., 1997; Nouairi et al., 2006; Zhang et al., 2008; Büyük et al., 2012; Yekbun and Kabay, 2017). As in our research, there are many studies in which mycorrhiza applications reduce the MDA content and preserve the membrane permeability (Baozhong et al., 2010; Koç, 2015; Moradtalab et al., 2019).

3.3. Some nutrient element contents

Many factors affect the obtainability of nutrients in the soil and their usefulness to the plant. Soil reaction is one of the most important of these factors (Yakupoğlu et al., 2010). Leaf mineral element contents are given in Table 4. In the first removal, the VAM application had a moderate (d=0.59) effect on P content and a small (d=0.36) effect on Zn content. The lime application had a moderate effect on Ca, Fe, and Zn contents (d=0.77, d= 0.72, and d=0.52, respectively). VAM application had no effect on the mineral element content in the uprooting during the flowering period. The lime application had a large effect on P, Mg, and Zn contents (d=0.89, d=0.92, and d=0.96, respectively) and moderately effect on Fe content (d=0.53). In the final removal, the VAM application had a small effect on Fe content (d=0.44) and moderately effect on Zn content (d=0.33). The lime application had a medium effect on Ca content (d=0.74), large effect on P content (d=0.90) and a small effect on Fe and Zn content (d=0.36 and 0.33). VAM × lime interaction was very important in leaf element contents in all removals.

In the experiment, the Ca content in the leaves was found to be in the range of 1.83-3.46% at beginning of flowering, 2.47-2.03% in flowering, and 2.94-1.98% in harvesting (Table 4). The Ca content of the leaves increased with the increase in the air

Table 5. The effects of VAM on some mineral element content of strawberry under calcareous conditions in the crown.

Application		Beginning of flowering					Flowering					Harvesting				
		Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)	Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)	Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)
Mycorrhiza	VAM	2.2	0.20	0.59	718	23.6	1.6	0.44	0.27	726	43.6	2.5	0.11	0.44	639	47.6
	Non VAM	2.0	0.15	0.49	355	23.0	1.6	0.16	0.22	634	36.7	2.1	0.11	0.36	530	20.9
$CaCO_3$	0% $CaCO_3$	1.5	0.17	0.50	760	23.8	1.2	0.42	0.31	1057	44.7	2.0	0.13	0.40	846	48.3
	1% $CaCO_3$	2.7	0.19	0.58	313	22.9	2.0	0.19	0.19	302	35.7	2.6	0.08	0.40	323	20.2
VAM×$CaCO_3$	VAM×0% $CaCO_3$	1.6 c	0.20 a	0.55 b	1043 a	24.2 a	1.3 c	0.63 a	0.29 d	1064 a	63.5 a	2.0 d	0.14 a	0.40 b	896 a	67.3 a
	VAM×1% $CaCO_3$	2.8 a	0.20 a	0.63 a	393 c	23.1 a	2.0 a	0.25 b	0.33 a	389 b	42.6 b	2.2 a	0.08 b	0.48 a	381 c	28.0 b
	N-VAM×0% $CaCO_3$	1.4 d	0.18 b	0.45 d	477 b	23.4 a	1.2 d	0.21 c	0.32 b	1051 a	44.7 b	2.0 d	0.13 a	0.40 b	795 b	29.3 b
	N-VAM×1% $CaCO_3$	2.6 b	0.13 c	0.53 c	232 d	22.6 b	1.9 b	0.12 d	0.30 c	216c	28.8 c	2.2 a	0.08 b	0.33 c	264 d	12.5 c
Significance																
VAM		Ns	*	**	*	Ns	Ns	**	Ns	Ns	*	Ns	Ns	**	Ns	*
$CaCO_3$		***	Ns	*	**	Ns	***	*	Ns	***	**	**	***	Ns	***	*
VAM×$CaCO_3$		**	**	**	**	**	**	**	**	**	**	**	**	**	**	**

Ca: Calcium, P: Phosphate, Mg: Magnesium, Fe: Iron, Zn: Zinc, VAM: Vesicular–arbuscular mycorrhiza, N-VAM: Non vesicular–arbuscular mycorrhiza, $CaCO_3$: Lime

* Significant at the $p < 0.05$ level, ** Significant at the $p < 0.01$ level, *** Significant at the $p < 0.001$ level, Ns: Not significant. Mean followed by different letters within columns differ significantly ($p < 0.05$).

temperature. Daugaard (2001) reported that the amount of Ca accumulated in the leaves increased with the increase in the air temperature. The Ca content in the strawberry leaves in the range of 0.20-1.50% was considered sufficient (May and Pritss, 1990).

In the present study, the P content in the leaves was found to be in the range of 0.02-1.14% at beginning of flowering, 0.07-0.23% in flowering, and 0.03-0.18% in harvesting (Table 4). To get sufficient efficiency in strawberry cultivation, the amount of P in the leaf to be 0.25-0.40% has been reported to be enough (May and Pritts, 1990). The VAM-applied plants were seen to be in or close to this range. It was observed that the addition of lime to the growing medium decreased the P uptake, whereas the application of mycorrhiza alleviated the effect of this decrease.

Looking at the Mg content of the leaves, it was found to be in the range of 0.74%-0.48% at beginning of flowering, 0.50-0.43% in flowering, and 0.46-0.35% in harvesting. May and Pritts (1990) stated that Mg content between the range of 0.20-0.50% was sufficient (Table 4). It is seen that Mg decreased with the addition of lime to the growth medium and that Mg intake increased with mycorrhiza application (Table 4).

We determined in our study that Fe in the leaves changed in the range of 340.15-190.50 ppm at beginning of flowering, 260.35-187.1 ppm in flowering, and 358.21-254.76 ppm in harvesting. The Fe content in strawberry leaves has been reported to change in the range of 70-1383.20 ppm (May and Pritts, 1990; Stanisavljevic et al., 1997). It was detected that the addition of lime to the growing medium decreased the Fe uptake, whereas the application of mycorrhiza alleviated the effect of this decrease.

Throughout our experiment, the Zn content of the leaves changed in the range of 13.83-6.71 ppm at beginning of flowering, 26.64-12.81 ppm in flowering, and 29.55-9.53 ppm in harvesting (Table 4). Ersoy and Demirsoy (2006) stated that the Zn content of the leaves was in the range of 22.9–54.9 ppm. May and Pritts (1990) determined that the Zn content of the leaves in the range of 20-50 ppm was sufficient for normal development and growth in strawberries. It decreased lime stress, mycorrhiza applications increased Zn intake.

The mineral element contents of the crown obtained during the experiment are given in Table 5. It was determined that the VAM application had a small (d=0.21) effect on the P content of the crown, medium (d=0.60) on the Mg content, and a small (d=0.36) effect on the Fe content in the crown during the uprooting before flowering. It was determined that the lime application had a large (d=0.98) effect on the Ca content of the crown, a small (d=0.39) effect on the Mg content, and a moderate (d=0.54) effect on the Fe content. In the second extraction, the VAM application had a moderate (d=0.51) effect on P content and a small (d=0.43) effect on Zn content. The lime application, on the other hand, had a large (d=0.99 and 0.97) effect on Ca and Fe content, moderate (d=0.55) on Zn content, and small (d=0.35) on P content. In the harvesting period, the VAM application had a medium (d=0.54) effect on the Mg content and a small (d=0.48) effect on the Zn content. Lime application, on the other hand, affected the Ca content at a moderate (d=0.53), P and Fe content at a large level (d=0.99 and d=0.96, respectively), and the Zn content at a small level (d=0.48). The VAM × lime interaction is essential in terms of crown mineral content in all dismantling.

In our study, the Ca content of the crown was seen to be on the level of 2.79-1.35% at beginning of flowering, 1.98-1.15% in flowering, and 2.20-1.96% in harvesting (Table 5). The Ca content of the crown was observed to be relatively low in the uprootings made during the blooming period, compared to other uprootings. The reason for this has been thought to be Ca transferring to other organs (particularly flowers) in this period (Ersoy and Demirsoy, 2006). Throughout our experiment, the P content of the crown was seen to be on the level of 0.20-0.13% at beginning of flowering, 0.63-0.12% in flowering, and 0.14-0.08% in harvesting

(Table 5). It has been stated in the studies conducted on strawberries that P content on the level of 0.21-0.35% in the crown is sufficient (Ersoy and Demirsoy, 2006; Demirsoy et al., 2010; Demirsoy et al., 2012). When data were examined, the P content of the crown was detected to be mildly low in the harvesting period. It is known that towards the end of development, plants take relatively less P from the soil and transfer the P that they absorb at the beginning of development to the fruit (Kacar, 2012).

Looking at the Mg content of the crown, it was determined as 0.63-0.45% at beginning of flowering, 0.33-0.29% in flowering, and 0.48-0.33% in harvesting (Table 5). In studies conducted on strawberries, the amount of Mg in the crown has changed in the range of 0.09% and 0.19% (Stanisavljevic et al., 1997; Demirsoy et al., 2010).

In our study, the Fe contents in the strawberry crown changed in the range of 1043.14-232.42 ppm at beginning of flowering, 1063.63-216.36 ppm in flowering, and 896.28-264.12 ppm in harvesting (Table 5). In the study of Ersoy and Demirsoy (2006), the Fe content of the strawberry crown was reported to change in the range of 408.3-2362.3 ppm. May et al. (1994) in their study conducted in New York determined the Fe content of the crown in the Earlyglow strawberry kind as approximately 300-1300 ppm.

Throughout our research, the Zn content of the strawberry crown was in the range of 24.15-22.57 ppm at beginning of flowering, 63.50-28.81 ppm in flowering, and 67.26-12.48 ppm in harvesting (Table 5). In a previous study, the Zn content was determined as 225.6-48 ppm in the crown throughout the experiment period (Ersoy and Demirsoy, 2006). May et al. (1994) have determined the Zn content of the crown in the range of 160-250 ppm.

The root mineral substance contents obtained in our study are given in Table 6. It was determined that VAM application had a moderate effect on P content (d=0.67) in the root, large and small effects on Mg and Fe content (d=0.98 and d=0.96, respectively) (d=0.37) in the root during flowering, and a moderate effect on Ca content (d=0.58). The lime application, on the other hand, had a moderate (d=0.53 and d=0.78) effect on Mg and Zn contents, and a small (d=0.48) effect on Fe content. In the final removal, the VAM application had a moderate (d=0.68) effect on the Zn content. The lime application had a large (d=0.97 and d=0.91, respectively) effect on Ca and Fe content. During the experiment, the VAM × lime interaction significantly affected the root mineral element contents.

While the content of Ca in roots in our experiment was seen between the range of 2.69-1.44% at beginning of flowering, 2.04-1.73% in flowering, and 3.06-1.53% in harvesting (Table 6). The amount of Ca decreased in the blooming period, similarly to the crown.

In the experiment, the P content changed in the range of 0.07-0.02% at beginning of flowering, 0.10-0.03% in flowering, and 0.08-0.05% in harvesting (Table 6). While Demirsoy et al. (2010) have determined the P content of the root as 0.33-0.22%, Stanisavljevic et al. (1997) have determined it as 0.09%. As the pH of the growth medium increases, P intake decreases (Hazelton and Murphy, 2007), and also mycorrhiza supports the intake of P (Giri et al., 2003; Tüfenkci et al., 2006; Sönmez et al., 2013).

The Mg content obtained in the uprootings performed at different times throughout the experiment is seen in Table 6. The Mg content in the roots of strawberry seedlings was determined in the range of 0.93-0.37% at beginning of flowering, 0.72-0.51% in flowering, and 0.72-0.64% in harvesting (Table 6). When studies conducted on strawberries were analyzed, the Mg content in the root has been in the range of 0.15-0.23% (Stanisavljevic et al., 1997; Ersoy and Demirsoy, 2006; Demirsoy et al., 2012).

Table 6. The effects of VAM on some mineral element content of strawberry under calcareous conditions in the root.

Application		Beginning of flowering					Flowering					Harvesting				
		Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)	Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)	Ca (%)	P (%)	Mg (%)	Fe (ppm)	Zn (ppm)
Mycorrhiza	VAM	2.4	0.05	0.89	3034	63.2	1.8	0.08	0.62	5218	48.9	2.3	0.07	0.68	5094	66.2
	Non VAM	2.0	0.02	0.39	1911	53.1	2.0	0.06	0.54	2814	40.2	2.2	0.07	0.66	4411	52.2
$CaCO_3$	0% $CaCO_3$	1.7	0.05	0.67	3118	64.0	1.9	0.06	0.64	5558	52.5	1.6	0.06	0.69	6388	63.1
	1% $CaCO_3$	2.6	0.03	0.60	1827	52.4	2.0	0.08	0.52	2474	36.5	2.9	0.07	0.65	3117	55.4
VAM×$CaCO_3$	VAM×0% $CaCO_3$	2.0 c	0.07 a	0.93 a	4192 a	66.2 a	1.7 d	0.10 a	0.72 a	7809 a	54.3 a	1.5 d	0.05	0.72 a	6467 a	72.5 a
	VAM×1% $CaCO_3$	2.7 a	0.04 b	0.84 b	1876 c	60.3 b	1.9 c	0.07 a	0.52 c	2627 c	43.4 c	3.1 a	0.08	0.64 c	3721 b	60.0 b
	N-VAM×0% $CaCO_3$	1.4 d	0.02 c	0.41 c	2045 b	61.8 b	2.0 b	0.03 b	0.56 b	3307 b	50.6 b	1.6 c	0.08	0.66 b	6310 a	53.7 c
	N-VAM×1% $CaCO_3$	2.6 b	0.02 c	0.37 d	1778 d	44.4 c	2.0 a	0.09 a	0.51 c	2321 d	29.7 d	2.7 b	0.06	0.67 b	2513 c	50.7 c
Significance																
VAM		Ns	**	***	***	*	**	Ns	Ns	Ns	Ns	Ns	Ns	Ns	Ns	**
$CaCO_3$		***	Ns	Ns	*	*	Ns	Ns	**	*	**	***	Ns	Ns	***	Ns
VAM×$CaCO_3$		**	**	**	**	**	**	**	**	**	**	**	Ns	**	**	**

Ca: Calcium, P: Phosphate, Mg: Magnesium, Fe: Iron, Zn: Zinc, VAM: Vesicular–arbuscular mycorrhiza, N-VAM: Non vesicular–arbuscular mycorrhiza, $CaCO_3$: Lime

* Significant at the $p < 0.05$ level, ** Significant at the $p < 0.01$ level, *** Significant at the $p < 0.001$ level, Ns: Not significant. Mean followed by different letters within columns differ significantly ($p < 0.05$).

The Fe content of the strawberry roots was determined in the range of 4191.71-1777.45 ppm at beginning of flowering, 7808.60-2321.20 ppm in flowering, and 6467.10-2512.95 ppm in harvesting (Table 6). In the study of Ersoy and Demirsoy (2006), the Fe content of the root changed in the range of 987.10–2643.30 ppm. In their study, May et al. (1994) determined the Fe content of the root in strawberries in the range of 900-2700 ppm. It is known that, as the pH of the environment increases, the plants' Fe intake decreases (Hazelton and Murphy, 2007). While Fe intake decreases with lime stress, mycorrhiza application was observed to increase the Fe intake.

The Zn content of the strawberry roots was determined in the range of 66.21-44.36 ppm at beginning of flowering, 54.39-29.71 ppm in flowering, and 72.45-50.70 ppm in harvesting (Table 6). In another study, the Zn content of the root changed in the range of 45.9–160.2 ppm (Ersoy and Demirsoy, 2006). May et al. (1994) determined in their study that the Zn content of the roots of the strawberry is in the range of approximately 110-140 ppm.

In all uprootings, the Ca content in the plant parts was observed to increase in direct proportion with the addition of Ca in the growth medium in the pots. Mg uptake is the highest at pH 7-8.5 (Kacar, 2012), therefore, since the pH range of our growth media was within these levels in our experiment, it is thought that the Mg content was higher than that of other studies.

The mycorrhiza applications were determined to significantly affect iron intake in lime stress. Significant differences were found between the applications in terms of the Fe content of the leaf throughout the experiment period. While lime stress decreases the Fe intake in the leaves, crowns, and roots, mycorrhiza applications were detected to increase the Fe intake (Table 4, 5, 6). The effect of mycorrhiza applications in lime stress on the Zn content of the leaves, crowns, and roots was significant (Table 4, 5, 6). While the intake of Zn, which is one of the microelements whose intake by plants is affected by the pH of the growth medium (Hazelton and Murphy, 2007). Many studies have revealed that mycorrhizal applications support the growth and development of the plant by supporting the intake of mineral elements in plants growing in stress conditions (Medeiros et al., 1994; Almaca, 2014; Latef et al., 2016).

4. Conclusion

Strawberries are cultivated in many parts of the world due to their adaptation ability. The most important factor in the increasing importance of strawberry in the world and in Türkiye, especially in recent years, is the breeding of varieties suitable for different climatic and soil conditions. However, despite the newly developed varieties, soil pH causes significant problems in strawberry cultivation. Mycorrhizal fungi in the soil play an active role in plant growth and use of nutrients in the environment. When the data obtained from our experiment were examined, it was determined that the amount of chlorophyll and leaf area decreased with the application of lime and increased to the same values as the plants in normal growing conditions with the application of VAM to the growing medium with lime. This situation was also detected in Fe intake. In addition, while mycorrhiza application to strawberry seedlings exposed to lime stress increased proline and total phenolic contents, it decreased MDA content. The use of mycorrhiza can be recommended to reduce the negative effects of strawberry cultivation in calcareous soils far from the sea.

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