

# **Turkish Journal of Agriculture and Forestry**

Volume 48 | Number 2

Article 11

4-1-2024

# Chitosan application improves yield and quality of rocket (Eruca sativa)

AYŞE ÖZKAN

HALİME ÜNLÜ

Follow this and additional works at: https://journals.tubitak.gov.tr/agriculture

Part of the Agriculture Commons, and the Forest Sciences Commons

# **Recommended Citation**

ÖZKAN, AYŞE and ÜNLÜ, HALİME (2024) "Chitosan application improves yield and quality of rocket (Eruca sativa)," *Turkish Journal of Agriculture and Forestry*: Vol. 48: No. 2, Article 11. https://doi.org/10.55730/1300-011X.3181

Available at: https://journals.tubitak.gov.tr/agriculture/vol48/iss2/11

This Article is brought to you for free and open access by TÜBİTAK Academic Journals. It has been accepted for inclusion in Turkish Journal of Agriculture and Forestry by an authorized editor of TÜBİTAK Academic Journals. For more information, please contact pinar.dundar@tubitak.gov.tr.



**Turkish Journal of Agriculture and Forestry** 

http://journals.tubitak.gov.tr/agriculture/

**Research Article** 

Turk J Agric For (2024) 48: 294-304 © TÜBİTAK doi:10.55730/1300-011X.3181

# Chitosan application improves yield and quality of rocket (*Eruca sativa*)

Ayşe ÖZKAN<sup>(D)</sup>, Halime ÜNLÜ<sup>(D)</sup>

Department of Horticulture, Faculty of Agriculture, Isparta University of Applied Sciences, Isparta, Turkiye

Received: 26.04.2023	٠	Accepted/Published Online: 08.02.2024	٠	Final Version: 01.04.2024
----------------------	---	---------------------------------------	---	---------------------------

Abstract: The present study aimed to investigate the effects of chitosan application on the yield and quality of rockets. "Bengi" rocket variety was used as a plant material and chitosan was applied to leaves at four different doses (0 (control)-75-150-300 ppm). The results revealed that the yield values varied between 1691-1914 g m<sup>-2</sup>, plant height between 24.33-27.92 cm, and leaf width between 4.56-5.71 cm among the applications. The total dry matter, chlorophyll, total phenolics, vitamin C, and antiradical activity values were determined to range between 7.71% and 8.68%, 34.15 and 36.68 SPAD, 104.67 and 180.84 mg 100 g<sup>-1</sup>, 126.63 and 143.51 mg 100 g<sup>-1</sup>, and 63.77% and 71.87%, respectively. The highest total phenolic and antiradical activity values were obtained at the 300 ppm application and the highest vitamin C values were obtained from the control group.  $a^{\circ}$  and  $h^{\circ}$  values varied between -17.87 to -18.71 and 124.09 to 125.78. Chitosan applications decreased nitrate accumulation compared to the control group. It was determined that the 75 ppm chitosan dose used in the study was better in terms of yield, plant height, leaf width, total dry matter, chlorophyll,  $a^{*}$ ,  $h^{*}$ , and nitrate accumulation parameters and was the recommended dose for rocket cultivation.

Key words: Chitosan, rocket, nitrate accumulation, biochemical properties, yield

#### 1. Introduction

Vegetables have important roles in the food and nutrition safety. Especially green leafy vegetables provide vital nutrients for human health and well-being. They are recognized as an exceptional source of vitamins, minerals, amino acids, phenolic compounds, folic acid, and essential fatty acids (Akan et al., 2022; Sardar et al., 2022). They have a high content of mineral nutrients such as iron and calcium. There are also significant socio-economic benefits. For example, green leafy vegetables are grown in order to support the household income of female farmers and to make use of the cold and unused periods in greenhouses (Natesh et al., 2017; Nadeem et al., 2018; Addique et al., 2022; Kul, 2022).

In recent years, with the understanding of the importance of rockets in the role of human health, there has been an increase in their production and consumption (Pasini et al., 2011; Bell et al., 2015).

Rocket (Eruca sativa), is a member of the Brassicaceae family and is widely grown in the Middle East, South Asia, Europe, and Mediterranean countries and consumed as a side dish and salad (Doležalová et al., 2013; Cavaiuolo and Ferrante, 2014; Bansal et al., 2015).

Rocket, which has a wide distribution around the world, is a winter crop grown for pure or mixed with grains, oil seeds, and legumes. It can be grown as an alternative crop with its adaptability to adverse environmental conditions and drought-resistant structure in lands where planting is delayed or other crops are not possible. (Garg and Sharma, 2014)

The rocket has a rich chemical composition and broad biological activity, a high nutritional value as a source of protein, carbohydrates, L-ascorbic acid, and mineral nutrients (Nurzyńska-Wierdak, 2015). It is an excellent source of vitamins A, C, K, riboflavin, niacin, thiamine, vitamin B-6, and pantothenic acid (Garg and Sharma, 2014).

Nowadays, in parallel with the rapid increase in the world population, there is intensive use of chemicals per unit area in order to close food deficits. This causes the deterioration of soil health and negative effects on sustainability in agriculture. In the last two decades, natural resources have been tried to be used to ensure sustainability in agricultural areas and to increase production and efficiency (Altindag et al., 2006; Celik et al., 2007; Erturk et al., 2012; Kesimci, 2013).

Changes in living conditions and the increase in the number of conscious consumers show that the available food resources need to be used more efficiently. For this, it is necessary to increase the quality as well as increase the yield in agriculture. Today, with the increase of conscious

<sup>\*</sup> Correspondence: halimeunlu@isparta.edu.tr 294

consumers, the food preferences of consumers have begun to change. Namely, consumers are turning to foods that have protective, therapeutic, and disease-reducing effects. For these reasons, efficiency-oriented production leaves its place to a production approach that includes both efficiency and quality, and it makes applications that increase quality as well as efficiency mandatory (Bayram and Elmacı, 2013).

One of the important factors in increasing the yield and quality of plants is growth regulators. Recently, there has been a focus on alternative substances that can be alternatives to plant growth regulators to increase production. For instance, biodegradable, natural, effective, and low-cost alternative substances known as plant growth regulators have become popular in the ornamental, horticultural, and agro-plant industries. Chitosan is one of the plant growth enhancer that is now being used as an alternative to plant growth regulators (Acemi, 2018).

Chitosan is a biopolymer, which is obtained by deacetylation of chitin, a polysaccharide naturally found in the exoskeletons of shellfish such as shrimp and crabs, in the wings of butterflies, and in the cell walls of fungi (Lizardi-Mendoza et al., 2016; Dotto and Pinto, 2017).

Chitosan, a natural cationic polysaccharide, is of great interest as a nontoxic renewable, biodegradable

biocompatible biopolymer used in many industries such as biomedicine, food, agriculture, pharmaceuticals, and cosmetics (Kaur and Dhillion, 2013). It causes various biochemical reactions in plants, such as stress resistance and increased productivity. It was found to depend on the chemical composition of chitosan and the timing and rate of application (Malerba and Cerena, 2016). Additionally, it has been determined that it has positive effects on the agronomic properties of vegetables. The use of chitosan is recommended to increase properties such as photosynthetic activity, vegetative growth, antioxidant activity, fruit quality, and yield (Sharif et al., 2018).

The aim of this study is to determine the effects of chitosan, which is not widely used in vegetable cultivation, and its different doses on yield and quality in rocket production.

#### 2. Materials and methods

The research was carried out in the open field ecological conditions of Sav district (Isparta, Türkiye) (37°45′58″N, 30°38′35″E, 1005 m).

During the trial period, the temperature and relative humidity ranged from 4.6 to 15.7°C and 59.1% to 77.5%, respectively. Table 1 shows the climate variables for October, November, and December months during the trial period.

Table 1. Some meteorological data for 2019.

	Monthly	Average Temperature (°C)			
Year/Month	October	November	December		
2019	15.7	9.8	4.6		
Monthly Total Precipitation (mm)					
Year/Month	October	November	December		
2019	9.9	28.6	45.3		
·	Monthly Av	erage Relative Humidity (%)			
Year/Month	October	October November I			
2019	59.1	71.6	77.5		
	Monthly Avera	ge 10 cm Soil Temperature (°	C)		
Year/Month	Tear/Month October November		December		
2019	16.3	9.8	4.9		
Monthly Average 20 cm Soil Temperature (°C)					
Year/Month	October	November December			
2019	18.4	12.1	7.2		

The soil properties of the trial area were analyzed by ISLAB Soil and Plant Analysis Laboratory. The chemical properties of the soil are summarized in Table 2.

In the experiment, 200 kg ha<sup>-1</sup> of diammonium phosphate fertilizer was applied before planting the seeds. During the vegetation period, ammonium sulfate was applied 2 times ( $20^{th}$  and  $50^{th}$  day after seed planting) in a total of 270 kg per hectare.

"Bengi" rocket variety which has lobed and toothed margins leaves was used as plant material in the study. Low molecular weight chitosan (%75–%85 deacetylated and 15 viscosity 20–300 cP, Sigma-Aldrich) was used for applications because it has many biological and physiological functions when used in agricultural production due to its strong solubility, low viscosity, and easy absorption by plants (Boamah et al., 2023). The solutions were dissolved in 0.15% acetic acid (Van et al., 2013). Before the application, 0.5% Tween 20 (Merck) was added to the solutions as a spreader-adhesive.

In the study, a total of 4 treatments with three different concentrations such as 75, 150, and 300 ppm of chitosan and control (untreated chitosan) were used. For this purpose, 12 parcels of 1 m<sup>2</sup> in size were created in the open field. The study was planned according to the randomized plot design with three replications. On October 10, 2019, planting was carried out on the plots using the pan method, with 2 g m<sup>-2</sup> rocket seeds placed in each plot.

Treatments were replicated three times in the form of foliar spraying during the developmental period. The first application was carried out after the emergence of the true leaves, and the other applications were carried out at 10-day intervals. Yield and quality analyses of the rockets harvested on 09 December 2019 were carried out at Isparta University of Applied Sciences, Faculty of Agriculture, Department of Horticulture.

#### Yield, plant height, leaf width

The rocket harvested from each replication was weighed separately on a scale with a precision of 0.01 g and the yield was determined as g m<sup>-2</sup>. The length values (cm) from the above-ground starting point to the leaf tip were determined by measuring the leaf width (cm) from the widest part of the leaf with the help of a ruler, from 10 randomly selected rocket leaves from each replication.

#### Dry matter

After determining the wet weights of the samples taken after harvest, they were dried in an oven at 65 °C until reached to a constant weight, and dry weights were determined by weighing them on a precision scale. From the wet and dry weights determined, the dry matter

(%) was determined using the Kul (2014) equation.

# Soluble solids content (SSC)

The amount of SSC of rocket leaves was determined as % (brix°) using a digital refractometer.

Analysis result	Evaluation				
0.14	Salt free				
8.05	Slightly alkaline				
1.74	Low				
3.36	Medium				
1309	Medium				
16.50	Medium				
373.32	High				
4914	High				
1328	High				
5.06	High				
2.04	Medium				
10.5	Medium				
0.84	Medium				
	0.14 8.05 1.74 3.36 1309 16.50 373.32 4914 1328 5.06 2.04 10.5				

Table 2. Soil properties of the trial area.

# Chlorophyll content

The chlorophyll content of the leaves was measured from three different locations of 10 randomly selected rocket leaves from each replication using a Minolta SPAD-502 chlorophyllmeter and the measured values were expressed as SPAD values.

# Leaf color

The color of the leaf samples was measured using a Minolta CR-400 model color meter from three different locations of 10 randomly selected rocket leaves from each replication, and the results were determined in terms of CIE  $L^*$ ,  $a^*$ ,  $b^*$ . Chroma ( $C^\circ$ ) and hue angle ( $h^\circ$ ) values were calculated according to McGuire (1992).

# Total phenolic content

For the extraction process, 20 mL of 95% ethyl alcohol was added to 5 g of the sample and crushed in a homogenizer for 2 min. After boiling the samples for 10 min, they were centrifuged at 8000 rpm and filtered through filter paper. Then, 20 mL of 80% ethanol was added and boiled for 10 min. After boiling, the samples were made up to 100 mL with 80% ethanol. After these procedures, total phenolic content analysis was carried out using the Folin-Ciocalteu reagent (Coseteng and Lee, 1987).

# Vitamin C content

200–300 g samples were taken and homogenized thoroughly with the help of a blender with the same amount of 2% oxalic acid. A certain amount of this mixture was weighed and made up to 100 mL with oxalic acid and filtered. By taking 10 mL of the filtrate, titration was carried out with 2.6 dichlorophenolindophenol solution. The volume was recorded and the results were determined using the formula described by Cemeroğlu (2013).

#### Nitrate accumulation

Nitrate accumulation was determined by the salicylic acid method (Cataldo et al., 1975) in the fresh samples. For this, 1 g of the sample was taken and 10 mL of distilled water was added to it and homogenized. Afterward, the samples were kept in an incubator at 45 °C for 1 h and centrifuged at 5000 rpm for 15 min. After centrifugation, 0.8 mL of 5%

salicylic acid dissolved in sulfuric acid was added to the 0.2 mL sample, and it was kept at room temperature for 20 min after mixing. Then, 19 mL of 2 N NaOH solution was added and the samples were kept at room temperature. The absorbance values of the samples and standards were read in the spectrophotometer (BOECO S-200) at a wavelength of 410 nm.

#### Antiradical activity

The antiradical activity was determined by the 2,2-diphenyl-1-picryl-hydrazine (DPPH) method (Dorman et al., 2003). After pureeing, the samples were filtered and centrifuged using a Nüve-NF 1200 centrifuge. Then, 450  $\mu$ L of Tris-HCl buffer (50 mM, pH 7.4) was added to the samples taken in the amount of 50  $\mu$ L. Finally, 1.00 mL of DPPH (0.10 mM, in methanol) solution was added to the mixture and left in the dark for 30 min. The absorbance values of the final mixtures were read in a spectrophotometer at 517 nm wavelength. The results were obtained using the formula; - % Inhibition (DPPH) = [(AbsControl-AbsSample)/AbsControl] × 100.

# Statistical analysis

In evaluating the data, Minitab (17) Inc. analysis of variance (ANOVA) was performed using the package program. Differences between the significant means were determined using Tukey's multiple range test at  $p \le 0.05$ .

# 3. Results and discussion

The effects of chitosan applied at different doses (75, 150, and 300 ppm) on the yield, plant height, leaf width, dry matter, chlorophyll, and SSC in the rocket are given in Table 3. While the treatments had significant effects on yield, plant height, leaf width, dry matter, and chlorophyll in the rocket, it was found that the effect on SSC was not significant.

As a result of the study, it was determined that the yield values of rockets varied between 1559.3 and 1913.9 g m<sup>-2</sup> according to the applications. Among the applications, the lowest value (1559.3 g m<sup>-2</sup>) was found in the 300 ppm chitosan application, while the highest yield value (1913.9 g m<sup>-2</sup>) was obtained from the 75 ppm chitosan application.

Chitosan doses	Yield (g m <sup>-2</sup> )	Plant height (cm)	Leaf width (cm)	Dry matter (%)	Chlorophyll content (SPAD)	Soluble solids content (%)
Control	1690.6 b*	24.33 b*	4.56 b*	7.71 b*	34.15 c*	4.43 <sup>n.s.</sup>
75 ppm	1913.9 a	27.92 a	5.71 a	8.68 a	36.68 a	4.88
150 ppm	1734.0 ab	26.95 a	5.30 a	8.45 a	36.24 a	4.33
300 ppm	1559.3 b	27.03 a	5.65 a	8.34 a	34.95 b	4.17

Table 3. The effects of chitosan applications on the yield, plant height, and leaf width in rocket.

\*: Means followed by different letters differ significantly at p < 0.05 (LSD test). n.s.: non significant

It has been reported that chitosan acts as a stimulator of plant defense responses, such as a stress factor in plants (Bautista-Baños et al., 2003). Chitosan binds to the cell membrane in plants, initiates a secondary messenger signal to the cell, and increases hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>) production in the chloroplast via octadecanoid and nitric oxide (NO) (Pichyangkura and Chadchawan, 2015). Transient increases in the cytokinin levels in plant tissues are also observed with the increase of H<sub>2</sub>O<sub>2</sub> (Ha et al., 2012). Mondal et al. (2012) reported that chitosan increases the basic enzyme activities in nitrogen metabolism and improves plant growth and development by improving nitrogen transport in leaves. Vasconsuelo et al. (2004) reported that chitosan applications increased the lignification of cell walls and lignin biosynthesis in plants, thus triggering shoot formation in plants. Compared to the control group, chitosan applications increased the yield values between 2.6% (150 ppm application) and 13.2% (75 ppm application). Actually, studies conducted by different researchers indicate that chitosan applications significantly increase yield values in rice (Boonlertnirun et al., 2008), strawberry (Abdel-Mawgoud et al., 2010), and pepper (Chookhongkha et al., 2012). In addition, it has been reported that chitosan applications caused an increase of yield approximately 20% in tomatoes (Walker et al., 2004), 27.9%-48.7% in okra (Mondal et al., 2012), and 157.14% in tomatoes (Rahman, 2015).

Plant height values varied between 24.33 and 27.92 cm according to the applications. While the highest plant height value was obtained from the 75 ppm dose of chitosan application, it was followed by the doses of 300 ppm (27.03 cm) and 150 ppm (26.95 cm) respectively.

In the study, when chitosan applications are compared with the control group, it is seen that they have positive effects on the plant height parameter at the rates of 10.8% (150 ppm), 11.1% (300 ppm), and 14.8% (75 ppm). Similar results have been found in wheat (Lian-Ju et al., 2014), tomato (Rahman, 2015), turmeric (Anusuya and Sathiyabama, 2016), and strawberry (Rahman et al., 2018).

Leaf width values varied between 4.56 and 5.71 cm according to the applications. While the highest leaf width value was detected in the 75 ppm dose of chitosan application, it was followed by 300 ppm and 150 ppm chitosan applications. The lowest leaf width value was obtained from the control group. In our study, it is seen that chitosan applications have positive effects on the leaf width when compared to the control group. With similar findings, Mondal et al. (2013) reported that as a result of chitosan applications at different doses (0, 50, 75, 100, and 125 ppm) in maize, leaf width increased with increase was not significant after 75 ppm. They also stated that the lowest leaf width was recorded in control plants.

Ohta et al. (1999) and Rahman et al. (2018) who applied different doses of chitosan from our study on lisianthus flower and strawberry, respectively, reported that chitosan applications had positive effects on leaf width compared to untreated plants. All these reports support our study.

As a result of the study, when Table 3 was examined based on the applications, it was determined that the dry matter values of the rocket varied between 7.71% and 8.68%. While the lowest value was found in the control (7.71%) among the applications, the highest dry matter value was obtained from the 75 ppm chitosan application.

It was determined that all applications increased the dry matter content between 8.2% and 12.6% as compared to the control group. In the study, it was observed that chitosan applications positively affected the chlorophyll content. As we know, the increase in chlorophyll induces photosynthesis and the growth of plants. In addition, chitosan promotes growth by increasing cellular activity in plants (Notario-Pérez et al., 2022). Data on yield, plant height, and leaf width indicate that plant growth is taking place and this may lead to an increase in dry matter content. Also, some researchers working on chitosan reported that chitosan increases the dry matter content of rice (Boonlertnirun et al., 2008; Ahmed et al., 2012) and okra (Mondal et al., 2012). Also, Bistgani et al. (2017) reported that chitosan increased the dry matter content by 15%-20% compared to the control by reducing the negative effect of stress in the thyme grown under drought stress.

Chlorophyll is the main pigment in green plants and performs basic functions in the life cycle of plants (Alaca et al., 2021). Chlorophyll SPAD values varied between 34.15 and 36.68 according to the applications. While the highest chlorophyll SPAD value was detected in the 75 ppm dose of chitosan, it was followed by 150 ppm and 300 ppm chitosan applications, respectively. The lowest chlorophyll SPAD value was obtained in the control group.

In our study, it was concluded that the chitosan applications had a positive effect on the amount of chlorophyll in the rocket when compared to the control group. Similarly, Xu and Mau (2018) in lettuce, Dzung et al. (2011) in coffee, Mondal et al. (2012) in okra, and Malekpoor et al. (2016) in basil reported that chitosan applications increased the chlorophyll content.

According to the applications, SSC values varied between 4.17% and 4.88%. The effects of chitosan applications on the SSC in rockets were not found significant. In support of our findings, it was reported that chitosan applications increased the amount of SSC, but this increase had no significant effect on the tomatoes (El-Tantawy, 2009), strawberries (Abdel-Mawgoud et al., 2010) and squash (Ibraheim and Mohsen, 2015). Sultana et al. (2017) found that chitosan applications increased the content of SSC in eggplant and decreased the content of SSC in tomatoes.

The effects of chitosan applications (75, 150, and 300 ppm) on the leaf color values of rocket are given in Table 4. It is seen that the  $a^*$  and  $h^\circ$  values of the leaf color are significant, while the effects on the  $L^*$ ,  $b^*$ , and  $C^*$  values are not significant.

According to the study, the  $L^*$  values ranged from 43.38 to 43.97 depending on the applications.

The lowest  $a^*$  value was found in the control (-17.87) among the applications, while the darkest green color was obtained from the 75 ppm chitosan application (-18.71). Supporting our findings, Chibu and Shibayama (2001) found that tomato and lettuce leaves turned darker green as a result of chitosan applications. Also, it has been determined that chitosan was effective in reducing color changes during storage in the studies on asparagus (Qiu et al., 2013) and orange (Ngoc et al., 2022) species. Fasciglione et al. (2020) reported that the chitosan coating had a positive effect on  $a^*$  and  $C^*$  values during the storage process of lettuce.

When Table 4 is examined, it has been determined that the  $b^*$  values vary between 25.97 and 26.40 and the  $C^*$  values vary between 31.89 and 32.01 according to the applications.

In this study, the  $h^{\circ}$  value was determined as 124.09 in the control group. When we look at the 75 ppm chitosan application, it was determined that the  $h^{\circ}$  value increased to 125.78. This positive effect of chitosan application on h<sup>o</sup> value was also detected in pepper leaves (Chookhongkha et al., 2012).

The effect of chitosan applied at different doses on the total phenolic, antiradical activity, vitamin C, and nitrate accumulation in the rocket was found to be significant (Figure).

According to the applications, the values for the total phenolic ranged from 104.67 to 180.84 mg 100 g<sup>-1</sup>. While the lowest value was found in the control group (104.67 mg 100 g<sup>-1</sup>), the highest total phenolic content was obtained from the 300 ppm chitosan application (180.84 mg 100 g<sup>-1</sup>).

Malerba and Cerena (2016) reported that elicitors increase the production of secondary metabolites by stimulating defense or stress-induced responses in plants. It has been reported that chitosan is used as an abiotic elicitor (stimulant) in the production of secondary metabolites in the plant cell and tissue cultures. Thus, it increases the amount of phenolic compounds and proteins by increasing the defense responses against biotic and abiotic stresses in plants and activating some genes in the signaling pathways (Kurtuluş and Vardar, 2020). These reports explain the increases in the total phenolic content of 7.5% (75 ppm application), 9.0% (150 ppm application), and 72.8% (300 ppm application) as compared to the control group in our study. As a matter of fact, in the studies conducted by different researchers, it was determined that chitosan applications increased the phenolic content of sweet basil (Kim et al., 2005), sunflower (Cho et al., 2008), eggplant (Mandal, 2010) and Hypericum perforatum (Brasili et al., 2014). In addition to this, it was reported that chitosan applications increased the phenolic content of tea (Srisornkompon et al., 2014) up to 9% compared to the control group.

It was determined that the antiradical activity values of rockets varied between 63.77% and 71.87%. In the study, it was determined that chitosan applications increased the antiradical activity by 12.7% (300 ppm application) compared to the control group. Similar results have been determined in lettuce sprouts (Viacava and Roura, 2015), in spinach (Singh, 2016), in sage (Vosoughi et al., 2018), in strawberries (Rahman et al., 2018), and in tomatoes (Hernández-Hernández et al., 2018). Also, Negrao et al. (2021) reported that antioxidant activity increased with the increase of phenolic compounds.

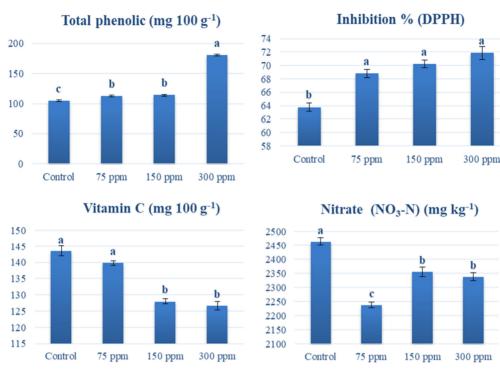
According to the applications, the vitamin C values varied between 126.63 and 143.51 mg 100 g<sup>-1</sup>. The highest vitamin C content was found in the control group (143.51 mg 100 g<sup>-1</sup>), while the lowest vitamin C content was obtained from the 300 ppm application (126.63 mg 100 g<sup>-1</sup>).

According to our investigation, applying chitosan to rocket plants reduced their vitamin C levels as compared

Chitosan Doses	$L^{\star}$	a*	$\boldsymbol{b}^{*}$	$C^*$	h°
Control	43.84 <sup>n.s.</sup>	-17.87 a*	26.40 <sup>n.s.</sup>	31.89 <sup>n.s.</sup>	124.09 b*
75 ppm	43.38	–18.71 b	25.97	32.01	125.78 a
150 ppm	43.71	-18.25 a	26.25	31.97	124.80 b
300 ppm	43.97	-17.99 a	26.34	31.90	124.33 b

Table 4. Effects of chitosan applications on the color values of rocket.

\*: Means followed by different letters differ significantly at p < 0.05 (LSD test). n.s.: non significant



**Figure.** The effects of chitosan applications on the total phenolic, antiradical activity, vitamin C, and nitrate accumulation values in rocket [The bars represent mean values of three replicates with standard error at p < 0.05 level].

to the control plants. Similar findings were determined in the tomato and eggplant (Sultana et al., 2017). Moreover, Zoldners et al. (2005) reported that the presence of chitosan significantly accelerates the oxidation of ascorbic acid and this effect varies depending on the chitosan/ ascorbic acid ratio.

According to the applications, the nitrate values varied between 2238.0 and 2463.6 mg kg<sup>-1</sup>. While the highest nitrate content was found in the control group, the lowest nitrate value was found in the 75 ppm chitosan application.

The presence of high doses of nitrates and nitrites in foods adversely affects human health. Nitrates in foods can be reduced to nitrites. It is known that nitrite causes methemoglobinemia and produces carcinogenic nitrosamines together with secondary and tertiary amines (Bryan and Van Grinsven, 2013). Toxic effective limits of nitrate vary according to source and country. Santamaria (2006) reports that these limits vary between 2500 and 4000 mg kg<sup>-1</sup> for the rocket.

In our study, when chitosan applications were compared with the control group, it was determined that the nitrate content decreased by 4.4% (150 ppm chitosan application), 5.1% (300 ppm chitosan application) and 9.2% (75 ppm chitosan application) rates. Similar to these results, Chung et al. (2001) found that chitosan applications in lettuce increased the nitrate reductase activity and decreased the nitrate content by 14 through 18% compared to the control group. Also, Mondal et al. (2011, 2012, 2016) reported that chitosan applications in spinach, okra, and tomatoes similarly increased the nitrate reductase activity.

#### 4. Conclusion

The results from this study showed that foliar application of chitosan at the early growth stage promotes plant growth and development which resulted in increased yield in rocket. Moreover, it could be concluded that chitosan had a significant effect not only on growth and yield but also on biochemical properties such as total phenolic, antiradical activity, and nitrate accumulation which are important for human health. Therefore, when the results were evaluated together, it was determined that 75 ppm dose among the chitosan concentrations was suitable for rocket cultivation and that chitosan can be used as an ecofriendly compound to increase the yield and quality of the crops.

#### Acknowledgment

This study is derived from a master's thesis performed by the first author in the Institute of Graduate Education at Isparta University of Applied Sciences.

No potential conflict of interest was reported by the authors.

#### References

- Abdel-Mawgoud AMR, Tantawy AS, El-Nemr MA, Sassine YN (2010). Growth and yield responses of strawberry plants to chitosan application. European Journal of Scientific Research 39 (1): 170-177.
- Acemi A (2018). Kitosan ile bazı bitki büyüme düzenleyicilerin Serapias vomeracea'nın in vitro fizyolojisi üzerine etkilerinin karşılaştırılması. Doktora Tezi, Kocaeli Üniversitesi Fen Bilimleri Enstitüsü Biyoloji Anabilim Dalı, Kocaeli, Türkiye (in Turkish).
- Ahmed R, Hossain ME, Ullah MA, Gazi AR, Rahman MZ (2012). Effect of chitosan on growth and yield of rice cv. BRRI dhan29. Journal of Agroforestry and Environment 7 (2): 33-35.
- Akan S (2022) Morphological characterisation and volatile analysis of Turkish garlic genotypes. Turkish Journal of Agriculture and Forestry 46 (4): 424-440. https://doi.org/10.55730/1300-011X.3015
- Alaca K, Okumuş E, Bakkalbaşı E, Javidipour I (2021). Phytochemicals and antioxidant activities of twelve edible wild plants from Eastern Anatolia, Turkey. Food Science and Technology 42: e18021. https://doi.org/10.1590/fst.18021
- Anusuya S, Sathiyabama M (2016). Effect of chitosan on growth, yield and curcumin content in turmeric under field condition. Biocatalysis and Agricultural Biotechnology 6: 102-106. https://doi.org/10.1016/j.bcab.2016.03.002.
- Addique M, Kausar A, Iqra I, Akhter N, Mujahid N, Parveen A, Zaman Q, Hussain S (2022) Amino acids application alleviated salinity stress in spinach (*Spinacia oleracea* L.) by improving oxidative defense, osmolyte accumulation, and nutrient balance. Turkish Journal of Agriculture and Forestry, 46 (6): 875-887. https://doi.org/10.55730/1300-011X.3049.
- Altindag M, Sahin M, Esitken A, Ercisli S, Guleryuz M et al. (2006) Biological control of brown rot (*Moniliana laxa* Ehr.) on apricot (*Prunus armeniaca* L. cv. Hacihaliloglu) by *Bacillus*, *Burkholderia* and *Pseudomonas* application under in vitro and in vivo conditions. Biological Control 38: 369-372.
- Bansal P, Medhe S, Ganesh N, Srivastava MM (2015). Antimelanoma potential of *Eruca sativa* seed oil and its bioactive principles. Indian Journal of Pharmaceutical Sciences 77 (2): 208-217. https://doi.org/10.4103/0250-474x.156600
- Bautista-Baños S, Hernández-López M, Bosquez-Molina E, Wilson CL (2003). Effects of chitosan and plant extracts on growth of *Colletotrichum gloeosporioides*, anthracnose levels and quality of papaya fruit. Crop Protection 22 (9): 1087-1092. https://doi. org/10.1016/S0261-2194(03)00117-0
- Bayram SE, Elmacı ÖL (2013). Gübrelemenin meyve ve sebzelerin fonksiyonel özellikleri üzerine etkileri. Adnan Menderes Üniversitesi Ziraat Fakültesi Dergisi 10 (1): 25-31 (in Turkish).
- Bell L, Oruna-Concha MJ, Wagstaff C (2015). Identification and quantification of glucosinolate and flavonol compounds in rocket salad (*Eruca sativa, Eruca vesicaria, and Diplotaxis tenuifolia*) by LC-MS: Highlighting the potential for improving nutritional value of rocket crops. Food Chemistry 172 (1): 852-861. https://doi.org/10.1016/j.foodchem.2014.09.116 PMID:25442630

- Bistgani ZE, Siadat SA, Bakhshandeh A, Pirbalouti AG, Hashemi M (2017). Interactive effects of drought stress and chitosan application on physiological characteristics and essential oil yield of *Thymus daenensis* Celak. The Crop Journal 5 (5): 407-415. https://doi.org/10.1016/j.cj.2017.04.003
- Boamah PO, Onumah J, Aduguba WO, Santo KG (2023). Application of depolymerized chitosan in crop production: A review. International Journal of Biological Macromolecules 235: 123858. https://doi.org/10.1016/j.ijbiomac.2023.123858
- Boonlertnirun S, Boonraung C, Suvanasara R (2008). Application of chitosan in rice production. Journal of Metals, Materials and Minerals 18 (2): 47-52.
- Brasili E, Pratico G, Marini F, Valletta A, Capuani G et al. (2014). A non-targeted metabolomics approach to evaluate the effects of biomass growth and chitosan elicitation on primary and secondary metabolism of *Hypericum perforatum* in vitro roots. Metabolomics 10 (6): 1186-1196. https://doi.org/10.1007/ s11306-014-0660-z
- Bryan NS, Van Grinsven H (2013). The role of nitrate in human health. Advances in Agronomy 119: 153-182. https://doi. org/10.1016/B978-0-12-407247-3.00003-2
- Cataldo DA, Maroon M, Schrader LE, Youngs VL (1975). Rapid colorimetric determination of nitrate in plant tissue by nitration of salicylic acid. Communications in Soil Science and Plant Analysis 6 (1): 71-80. https://doi. org/10.1080/00103627509366547
- Cavaiuolo M, Ferrante A (2014). Nitrates and glucosinolates as strong determinants of the nutritional quality in rocket leafy salads. Nutrients 6 (4): 1519-1538. https://doi.org/10.3390/ nu6041519
- Celik A, Ercisli S, Turgut N (2007) Some physical, pomological and nutritional properties of kiwifruit cv. Hayward. International Journal of Food Sciences and Nutrition, 58 (6): 411-418.
- Cemeroğlu B (2013). Meyve ve Sebze İşleme Endüstrisinde Temel Analiz Metotları. Ankara Biltav Yayınları (in Turkish).
- Chibu H, Shibayama H (2001). Effects of chitosan applications on the growth of several crops. Chitin and Chitosan in Life Science, Kodansha Scientific LTD.
- Cho MH, No HK, Prinyawiwatkul W (2008). Chitosan treatments affect growth and selected quality of sunflower sprouts. Journal of Food Science 73 (1): 70-77. https://doi.org/10.1111/j.1750-3841.2007.00607.x
- Chookhongkha N, Miyagawa S, Jirakiattikul Y, Photchanachai S (2012). Chili growth and seed productivity as affected by chitosan. In: Proceedings of the International Conference on Agriculture Technology and Food Sciences; Manila, Philippines. pp. 17-18.
- Chung JB, Park SG, Park S (2001). Suppression of nitrate accumulation in lettuce by application of mg and micronutrients. Korean Journal of Environmental Agriculture 20 (5): 340-345. https:// doi.org/10.3390/agronomy10081208

- Coseteng MY, Lee CY (1987). Changes in apple polyphenoloxidase and polyphenol concentrations in relation to degree of browning. Journal of Food Science 52 (4): 985-989. https://doi. org/10.1111/j.1365-2621.1987.tb14257.x
- Doležalová I, Duchoslav M, Dušek K (2013). Biology and yield of rocket (*Eruca sativa* Mill.) under field conditions of the Czech Republic (Central Europe). Notulae Botanicae Horti Agrobotanici Cluj-Napoca 41 (2): 530-537. https://doi. org/10.15835/nbha4129281
- Dorman HJD, Peltoketo A, Hiltunen R, Tikkanen MJ (2003). Characterisation of the antioxidant properties of de-odourised aqueous extracts from selected Lamiaceae herbs. Food Chemistry 83 (2): 255-262. https://doi.org/10.1016/S0308-8146(03)00088-8
- Dotto GL, Pinto LAA (2017). General considerations about chitosan. Frontiers in Biomaterials 3: 3-33. https://doi.org/10.2174/9781 6810848551170301
- El-Tantawy EM (2009). Behavior of tomato plants as affected by spraying with chitosan and aminofort as natural stimulator substances under application of soil organic amendments. Pakistan Journal of Biological Sciences 12 (17): 1164-1173. https://doi.org/10.3923/pjbs.2009.1164.1173
- Erturk Y, Ercisli S, Cakmakci R (2012) Yield and growth response of strawberry to plant growth-promoting rhizobacteria inoculation. Journal of Plant Nutrition, 35 (6): 817-826.
- Fasciglione G, Goñi MG, Yommi AK, Perez-Bravo JJ, Ortueta R et al. (2020). Revaluation of waste from fishing industry through generation of chitosan coatings to improve quality and extend shelf-life of minimally processed lettuce. Postharvest Biology and Technology 170: 111310. https://doi.org/10.1016/j. postharvbio.2020.111310
- Garg G, Sharma V (2014). *Eruca sativa* (L.): Botanical description, crop improvement, and medicinal properties. Journal of Herbs, Spices and Medicinal Plants 20 (2): 171-182. https://doi.org/10. 1080/10496475.2013.848254
- Ha S, Vankova R, Yamaguchi-Shinozaki K, Shinozaki K, Tran LSP (2012). Cytokinins: Metabolism and function in plant adaptation to environmental stresses. Trends in Plant Science 17 (3): 172-179. https://doi.org/10.1016/j.tplants.2011.12.005
- Hernández-Hernández H, González-Morales S, Benavides-Mendoza A, Ortega-Ortiz H, Cadenas-Pliego G et al. (2018). Effects of chitosan–PVA and Cu nanoparticles on the growth and antioxidant capacity of tomato under saline stress. Molecules 23 (1): 178. https://doi.org/10.3390/molecules23010178
- Ibraheim SKA, Mohsen AAM (2015). Effect of chitosan and nitrogen rates on growth and productivity of summer squash plants. Middle East Journal 4 (4): 673-681.
- Kaur S, Dhillion GS (2013). The versatile biopolymer chitosan: potential sources, evaluation of extraction methods and applications. Critical Reviews in Microbiology 40 (2): 155-175. https://doi.org/10.3109/1040841X.2013.770385.
- Kesimci E (2013). Sera koşullarında bitki büyümesini artırıcı rizobakterlerin marulda verim, verim unsurları ve besin elementi içeriklerine etkileri. Yüksek Lisans Tezi, Selçuk Üniversitesi Fen Bilimleri Enstitüsü Bahçe Bitkileri Anabilim Dalı, Konya, Türkiye (in Turkish).

- Kim HJ, Chen F, Wang X, Rajapakse NC (2005). Effect of chitosan on the biological properties of sweet basil (*Ocimum basilicum* L.). Journal of Agricultural and Food Chemistry 53 (9): 3696-3701. https://doi.org/10.1021/jf0480804
- Kul R (2014). Balık gübresi, mineral gübre ve kombinasyonlarının marulda (*Lactuca sativa* L.) bitki gelişimi ve besin elementi içeriği üzerine etkisi. Yüksek Lisans Tezi, Atatürk Üniversitesi Fen Bilimleri Enstitüsü Bahçe Bitkileri Anabilim Dalı, Erzurum, Türkiye (in Turkish).
- Kul R (2022). Integrated application of plant growth promoting rhizobacteria and biochar improves salt tolerance in eggplant seedlings. Turkish Journal of Agriculture and Forestry, 46 (5): 677-702. https://doi.org/10.55730/1300-011X.3035
- Kurtuluş G, Vardar F (2020). Kitosanın özellikleri, uygulama alanları, bitki sistemlerine etkileri. International Journal of Advances in Engineering and Pure Sciences 32 (3): 258-269. https://doi. org/10.7240/jeps.635430 (in Turkish).
- Li Y, Ren J, Zhang G, Li X (2021). High-barrier and antibacterial films based on PET/SiO x for food packaging applications. Food Science and Technology 41: 763-767. https://doi.org/10.1590/ fst.37720
- Lian-ju M, Yueying L, Lanlan W, Xuemei L, Ting L et al. (2014). Germination and physiological response of wheat (*Triticum aestivum*) to pre-soaking with oligochitosan. International Journal of Agriculture and Biology 16 (4): 766-770.
- Lizardi-Mendoza J, Mona WMA, Valencia FMG (2016). Chemical characteristics and functional properties of chitosan. In: Bautista-Baños S, Romanazzi G, Jiménez-Aparicio A (Editors). Chitosan in the preservation of agricultural commodities. Academic Press. pp. 3-31. https://doi.org/10.1016/B978-0-12-802735-6.00001-X
- Malekpoor F, Pirbalouti AG, Salimi A (2016). Effect of foliar application of chitosan on morphological and physiological characteristics of basil under reduced irrigation. Research on Crops 17 (2): 354-359. https://doi.org/10.5958/2348-7542.2016.00060.7.
- Malerba M, Cerena R (2016). Chitosan effects on plant systems. International Journal of Molecular Sciences 17: 996. https:// doi.org/10.3390/ijms17070996
- Mandal S (2010). Induction of phenolics, lignin and key defense enzymes in eggplant (*Solanum melongena* L.) roots in response to elicitors. African Journal of Biotechnology 9 (47): 8038-8047. https://doi.org/10.5897/AJB10.984
- McGuire RG (1992). Reporting of objective color measurements. HortScience 27: 1254-1255. http://doi.org/10.21273/ HORTSCI.27.12.1254
- Mondal MMA, Malek MA, Puteh AB, Ismail MR, Ashrafuzzaman M et al. (2012). Effect of foliar application of chitosan on growth and yield in okra. Australian Journal of Crop Science 685: 918-921.
- Mondal MMA, Puteh AB, Dafader NC, Rafii MY, Malek MA (2013). Foliar application of chitosan improves growth and yield in maize. Journal of Food, Agriculture and Environment 11 (2): 520-523.

- Mondal MMA, Rana IK, Dafader NC, Haque ME (2011). Effect of foliar application of chitosan on growth and yield in Indian spinach. Journal of Agroforestry and Environment 5 (1): 99-102.
- Mondal M, Puteh AB, Dafader NC (2016). Foliar application of chitosan improved morphophysiological attributes and yield in summer tomato (*Solanum lycopersicum*). Pakistan Journal of Agricultural Sciences, 53 (2): 339-344. https://doi. org/10.21162/PAKJAS/16.2011
- Muniz ADS, Carvalho GA, Raices RSL, Souza SLQD (2022). Organic vs conventional agriculture: evaluation of cadmium in two of the most consumed vegetables in Brazil. Food Science and Technology 42: e106721. https://doi.org//10.1590/fst.106721.
- Nadeem MA, Habyarimana E, Çiftçi V, Nawaz MA, Karaköy T et al. (2018) Characterization of genetic diversity in Turkish common bean gene pool using phenotypic and whole-genome DArTseq-generated silicoDArT marker information. PLoS ONE, 13(10): e0205363.
- Natesh N, Abbey L, Asiedu S (2017). An overview of nutritional and antinutritional factors in green leafy vegetables. Horticulture International Journal 1 (3-9): 58-65. https://doi.org/10.15406/ hij.2017.01.00011
- Negrao LD, Sousa PVDL, Barradas AM, Brandão ADCAS, Araújo MADM et al. (2021). Bioactive compounds and antioxidant activity of crisphead lettuce (*Lactuca sativa* L.) of three different cultivation systems. Food Science and Technology 41: 365-370. https://doi.org/10.1590/fst.04120
- Ngoc LS, Van PTH, Nhi TTY, Dung NA, Manh TD (2022). Effects of dipping time in chitosan (CS) and polyvinyl alcohol (PVA) mixture to quality of orange fruits during storage. Food Science and Technology 42: 1-7. https://doi.org/10.1590/fst.114221
- Notario-Pérez F, Martín-Illana A, Cazorla-Luna R, Ruiz-Caro R, Veiga MD (2022). Applications of chitosan in surgical and post-surgical materials. Marine drugs 20 (6): 396. https://doi. org/10.3390/md20060396
- Nurzyńska-Wierdak R (2015). Nutritional and energetic value of *Eruca sativa* Mill. leaves. Acta Scientiarum Polonorum-Hortorum Cultus 14 (4): 191-199.
- Ohta K, Taniguchi A, Konishi N, Hosoki T (1999). Chitosan treatment affects plant growth and flower quality in *Eustoma grandiflorum*. HortScience 34 (2): 233-234. https://doi. org/10.21273/hortsci.34.2.233
- Pasini F, Verardo V, Cerretani L, Caboni MF, D'Antuono LF (2011). Rocket salad (*Diplotaxis* and *Eruca* spp.) sensory analysis and relation with glucosinolate and phenolic content. Journal of the Science of Food and Agriculture 91 (15): 2858-2864. https:// doi.org/10.1002/jsfa.4535 PMID:21725983
- Pichyangkura R, Chadchawan S (2015). Biostimulant activity of chitosan in horticulture. Scientia Horticulturae 196: 49-65. https://doi.org/10.1016/j.scienta.2015.09.031
- Qiu M, Jiang H, Ren G, Huang J, Wang X (2013). Effect of chitosan coatings on postharvest green asparagus quality. Carbohydrate Polymers 92 (2): 2027-2032. https://doi.org/10.1016/j. carbpol.2012.11.070

- Rahman MA (2015). Performance of Modified Chitosan Treated Tomato (*Lycopersicon esculentum*) Seedlings on the Growth and Yield of Tomato (Doctoral Dissertation). Sher-E-Bangla Agricultural University Department of Soil Science.
- Rahman M, Mukta JA, Sabir AA, Gupta DR, Mohi-Ud-Din M et al. (2018). Chitosan biopolymer promotes yield and stimulates accumulation of antioxidants in strawberry fruit. PLoS One 13 (9): E0203769. https://doi.org/10.1371/journal.pone.0203769
- Santamaria P (2006). Nitrate in vegetables: toxicity, content, intake and EC regulation. Journal of the Science of Food and Agriculture 86: 10-17. https://doi.org/10.1002/jsfa.2351.
- Sardar H, Irshad M, Anjum MA, Hussain S, Ali S et al. (2022) Foliar application of micronutrients improves the growth, yield, mineral contents, and nutritional quality of broccoli (Brassica oleracea L.). Turkish Journal of Agriculture and Forestry, 46 (6):791-801. https://doi.org/10.55730/1300-011X.3043
- Sharif R, Mujtaba M, Rahman MU, Shalmani A, Ahmad H et al. (2018). The multifunctional role of chitosan in horticultural crops; a review. Molecules 23 (4): 872. http://doi.org/10.3390/ molecules23040872
- Singh S (2016). Enhancing phytochemical levels, enzymatic and antioxidant activity of spinach leaves by chitosan treatment and an insight into the metabolic pathway using DART-MS technique. Food Chemistry 199: 176-184. https://doi. org/10.1016/j.foodchem.2015.11.127
- Srisornkompon P, Pichyangkura R, Chadchawan S (2014). Chitosan increased phenolic compound contents in tea (*Camellia* sinensis) leaves by pre-and post-treatment. Journal of Chitin and Chitosan Science 2 (2): 93-98. https://doi.org/10.1166/ jcc.2014.1056
- Sultana S, Islam M, Khatun MA, Hassain MA, Huque R (2017). Effect of foliar application of oligo-chitosan on growth, yield and quality of tomato and eggplant. Asian Journal of Agricultural Research 11 (2): 36-42. https://doi.org/10.3923/ajar.2017.36.42
- Van SN, Minh HD, Anh AN (2013). Study on chitosan nanoparticles on biophysical characteristics and growth of robusta coffee in greenhouse. Biocatalysis and Agricultural Biotechnology 2 (4): 289-294. https://doi.org/10.1016/j.bcab.2013.06.001
- Vasconsuelo A, Giulietti AM, Boland R (2004). Signal transduction events mediating chitosan stimulation of anthraquinone synthesis in *Rubia tinctorum*. Plant Science 166 (2): 405-413. https://doi.org/10.1016/j.plantsci.2003.10.007
- Viacava G.E, Roura SI (2015). Principal component and hierarchical cluster analysis to select natural elicitors for enhancing phytochemical content and antioxidant activity of lettuce sprouts. Scientia Horticulturae 193: 13-21. https://doi. org/10.1016/j.scienta.2015.06.041
- Vosoughi N, Gomarian M, Pirbalouti AG, Khaghani S, Malekpoor F (2018). Essential oil composition and total phenolic, flavonoid contents, and antioxidant activity of sage (*Salvia officinalis* L.) extract under chitosan application and irrigation frequencies. Industrial Crops and Products 117: 366-374. https://doi. org/10.1016/j.indcrop.2018.03.021

- Walker R, Morris S, Brown P, Gracie A (2004). Evaluation of Potential for Chitosan to Enhance Plant Defense. A Report for the Rural Industries Research and Development Corporation, Australia. RIRDC Publication.
- Xu C, Mou B (2018). Chitosan as soil amendment affects lettuce growth, photochemical efficiency, and gas exchange. Horttechnology 28 (4): 476-480. https://doi.org/10.21273/ HORTTECH04032-18
- Zoldners J, Kiseleva T, Kaiminsh I (2005). Influence of ascorbic acid on the stability of chitosan solutions. Carbohydrate Polymers 60 (2): 215-218. https://doi.org/10.1016/j.carbpol.2005.01.013