

Improvement of some nutritional properties, phenolic compound amounts and antioxidant enzyme levels of garden cress (*Lepidium sativum* L.) grown with controlled salinity stress application

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ABSTRACT. The present study subjected two distinct cultivars of cress (Dadaş and Bahar) to salinity treatment (0, 30, and 60 mM) to determine its effect on the properties of the cress cultivars. The Dadaş cress cultivar exhibited higher levels of antioxidant enzymes, phenolic compounds, anthocyanins, vitamins, and amino acids compared to the Bahar cress cultivar ($P \leq 0.01$). However, Bahar cress had a higher content of sucrose, fructose, and galactose compared to Dadaş cress. The salinity treatment resulted in a marked increase in the levels of antioxidant enzymes in both cultivars of cress. Moreover, the Catalase (CAT) activity in Dadaş and Bahar cress rose significantly from 12.89 to 22.45 and from 6.71 to 17.33, while the Peroxidase (POD) activity increased from 14.01 to 25.87 and from 8.95 to 18.57 (EU/g plant), respectively. Additionally, histidine levels in both cress cultivars increased under the 60 mM salinity treatment. Although the sugar content of cress cultivars increased with salinity application, interestingly, the 30 mM salinity treatment caused a decrease in the level of fructose. An increase in the levels of individual phenolic compounds was observed depending on the concentration of the salinity treatment, while a decrease in the amount of anthocyanins was noted. Finally, this research indicates that the application of salinity, an abiotic stress factor, influences the nutritional and functional compounds of cress cultivars, with salt concentration playing a significant role.

Keywords: Garden cress; abiotic stress; salinity; antioxidant enzymes; phenolic compounds; anthocyanins.

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Introduction

Plants contain high levels of natural antioxidants, which can help prevent chronic diseases, making it important to consume a diet rich in antioxidants and phenolic compounds (Conforti et al., 2009). Leafy greens are not only low in calories but are also significant sources of minerals, vitamins, and especially secondary metabolites. Consequently, there has been a growing interest in consuming healthy foods in recent years due to their benefits for human health and high nutritional value (Farha et al., 2018).

Garden cress (*Lepidium sativum* L.) is an edible, fast-growing annual plant belonging to the Brassicaceae family that can be cultivated in various parts of the world. It is also added to salads as a spice due to its nutritional properties (Rafińska et al., 2019). It is stated that garden cress could be used to treat many diseases in both traditional and modern medicine (Rafińska et al., 2019). The seeds and leafs of *Lepidium sativum* are rich in secondary metabolites such as phenolic compounds, flavonoids and glucosinolates.

Abiotic stress conditions primarily consist of various factors, including extremely high or low temperatures, drought, salinity, and UV radiation. Plants cannot cope with these factors and survive, leading to significant crop loss worldwide (Hirayama & Shinozaki, 2010). Although abiotic stress causes crop loss, it can also enhance the activity of antioxidant enzymes (Rajput et al., 2021) and increase levels of carotenoids and sugars (Atkinson et al., 2011).

Abiotic stressors are expected to escalate in the coming years as a consequence of global climate change, resulting in greater exposure of plants to these environmental factors. A better understanding of the effects

of abiotic stresses is becoming increasingly important for agricultural production and plant science. In this study, two different garden cress species (Dadas and Bahar) were exposed to abiotic stress conditions (0, 30, and 60 mM salinity). The effects of abiotic stress conditions on the concentrations of sugars, antioxidant enzymes, vitamin contents, amino acid composition, phenolic compounds, and anthocyanin compounds in garden cresses were determined.

Materials and methods

Plant material and salinity treatment

In this work, two different varieties of garden cress (Dadas and Bahar), harvested in Turkey, were selected for study. Dadaş is a recently registered variety cultivated in the eastern region of Turkey, featuring serrated and curled leaves. Bahar, on the other hand, is a common garden cress variety that is widely harvested in Turkey. The seeds of the two cress species were planted in pots filled with a mixture of soil, peat and sand (2:1:1, v/v) in a non-heated greenhouse at Atatürk University (Crop Production Application and Research Center).

Salt stress was induced in the root zone of the cress varieties using irrigation waters consisting of 0, 30, and 60 mM NaCl. The salinity process began with seed planting. Salt stress in the environment was gradually increased, beginning with an initial concentration of 25 mM until the desired concentrations were reached.

Determination leaf sugar contents of garden cress samples

Sugar standards of HPLC were supplied from Sigma-Aldrich (Shanghai, China) and calibration curves were created using these standards. Extraction process of sugars were applied as stated by Nikolidaki et al. (2017). Briefly, two grams of cress were weighted and extracted by 20 mL ethanol solution of 80% (v/v) through overnight agitation. This was followed by 150 min. of sonication. For determination of sugar contents of garden cress varieties, a HPLC system (Agilent Technologies, 1100 series, USA) was used, which was formed by a RID (1260 series), an auto-sampler, isocratic pump and a data analysis software. Purospher STAR NH₂ column (250 × 4.6 mm, 5 µm, Merck, Germany) at a flow rate of 1 mL/min was used to carry out isocratic elution. The injection capacity was 0.01 mL, while both RID and oven temperature were held at 40 °C.

Determination of antioxidant enzymes

Antioxidant enzymes of garden cress varieties were determined with procedure stated by Keskin et al. (2022) and Kaya et al. (2022). Garden cress samples were rinsed thrice with 50 mM Tris-HCl + 0.1 M Na₂SO₄ (pH 8.0) and homogenized with liquid nitrogen. Then, samples were mixed with buffer (10 mM Na₂N₃ + 100 mM PVP + 0.1 M Na₂SO₄ (pH 8.0) + 50 mM Tris-HCl). Finally, the centrifuge conditions were as follows: 15,000 rpm for 60 minutes at 4 °C. Enzyme extracts were added to initiate all enzymatic operations, and enzymatic activity was evaluated using a spectrophotometer (Shimadzu 1208 UV, Kyoto, Japan) at 25 °C. In this study, the following enzymes were identified as antioxidant enzymes in garden cress samples: GR (glutathione reductase), GST (glutathione S-transferase), CAT (catalase), POD (peroxidase), SOD (superoxide dismutase), APX (ascorbate peroxidase), G6PD (glucose- 6-phosphate dehydrogenase), and 6GPD (6-phosphogluconate dehydrogenase).

Determination of vitamin content

For the determination of vitamin contents (Vitamin C, A, and B complex vitamins) in garden cress varieties, the extraction procedure and methodology were carried out as specified by Keskin et al. (2022). An HPLC system equipped with an auto-sampler, PDA detector (at 270 nm), and a C-18 column was used. A volume of 20 µL of filtered extract was injected to the HPLC. A 100 mM NaH₂PO₄ buffer (pH 2.2) containing 0.8 mM C₈H₁₇NaO₃S and C₂H₃N (9:1%, v/v) at 40 °C served as the mobile phase. The flow rate was set to 0.8 mL min⁻¹.

Amino acid composition of garden cress samples

An HPLC system was utilized, consisting of a DAD detector, a Zorbax Eclipse AAA analytical column, and an auto-sampler, to measure the amino acid compositions of garden cress varieties. Fmoc and OPA were employed for inline derivatization before injection into the columns (Kaya et al., 2022). Fmoc and OPA derivatized amino acids were monitored at 262 and 338 nm, respectively. Amino acid standards were obtained from Sigma. Sarcosine and norvaline were used as internal standards for Fmoc and OPA derivatives. Amino acid concentrations were reported as pmol µL⁻¹.

Quantification of phenolic compounds of garden cress samples by RP-HPLC

To evaluate the phenolic compounds in garden cress samples, an RP-HPLC (Shimadzu, Japan) system was used, following the procedure specified by Sagdic et al. (2011). The system included an LC-10ADvp pump, a DAD detector, a CTO-10Avp column heater, an SCL-10Avp system controller, a DGU14A degasser, and a SIL-10ADvp auto-sampler. Separations were performed at 30°C on an Agilent Eclipse XDB C-18 reversed-phase column. A mobile phase consisting of 2.0% acetic acid in distilled water (A) and methanol (B) was used. Samples were passed through the column at a flow rate of 0.8 mL min⁻¹. Twenty-five mg of dried garden cress extract was dissolved in 1 mL of methanol, and 10 µL of the solution was injected. Finally, the following phenolic compounds were detected: gallic acid, vanillic acid, trans-caffeic acid, trans-p-coumaric acid, ferulic acid, kaftaric acid, (+)-catechin, (-)-epicatechin, quercetin, rutin, and tyrosol. Results are presented as µg gram⁻¹ of extract.

Quantification of anthocyanins of garden cress samples

Anthocyanins in garden cress samples were determined by HPLC according to the method of Fang et al. (2021). Briefly, anthocyanins were extracted from 2 g of lyophilized ground powder using methanol containing 0.1% HCl (v/v). The HPLC system (Agilent 1260, USA) was configured with a C18 column, using 0.3% phosphoric acid in water (A) and acetonitrile (B) as mobile phases, with a flow rate of 1 mL min⁻¹. The linear gradient of phase B was conducted according to the method outlined by Fang et al. (2021). 10 µL of garden cress extracts were injected, and anthocyanins were identified at 520 nm. The peaks obtained were compared with anthocyanin standard peaks.

Statistical analysis

The experiment was designed according to a completely randomized factorial design. Two-way ANOVA was performed using SPSS, and means were compared using Duncan's multiple range test.

Results and discussion

Sugar contents of garden cress cultivars

Sugar contents of Dadaş and Bahar cress cultivars are shown in Table 1. Xylose (3.68 ± 0.05 g 100 g⁻¹), arabinose (3.59 ± 0.20 g 100 g⁻¹), and rhamnose (2.77 ± 0.06 g 100 g⁻¹) were the sugars with the highest levels detected in the Dadaş cress cultivar, while fructose (5.33 ± 0.36 g 100 g⁻¹), sucrose (3.05 ± 0.32 g 100 g⁻¹), and xylose (2.82 ± 0.16 g 100 g⁻¹) were detected at the highest levels in the Bahar cress cultivar. Galactose and glucose were found at their lowest levels in the Dadaş and Bahar cress cultivars, respectively. Generally, salinity treatment resulted in an increase in the sugar content of both cress cultivars ($P \leq 0.01$). However, a 30 mM salinity treatment resulted in a reduction in fructose levels for both varieties of cress, followed by an increase in fructose levels with the 60 mM salinity treatment. After salinity treatment, the most significant increases were observed in sucrose and galactose levels for the Dadaş cress cultivar. The sucrose level in Dadaş cress increased from 0.62 to 7.86 g 100 g⁻¹, while the galactose level increased from 0.46 to 5.67 g 100 g⁻¹. Karazhiyan et al. (2009) reported that the sugar composition of cress seeds contains mannose, arabinose, fructose, galactose, rhamnose, glucose, and galacturonic acid. Similarly, Razmkhah et al. (2016) noted that the sugar composition of cress seed gum includes fucose, rhamnose, arabinose, galactose, glucose, xylose, mannose, and galacturonic acid. Our study aligns with the aforementioned literature, as similar sugars were detected for both cress varieties, except for sucrose. Manaa et al. (2014) found that salinity application led to an increase in the total sugar content in the shoots and roots of cress. Botella et al. (2021) reported that salinity application resulted in increased glucose and fructose levels in tomatoes. Geranpayeh et al. (2017) found that the soluble sugar content in cress increased in proportion to salinity levels. The increase in sugar content in plants due to salt stress corresponds with the impact of salt on enzymes responsible for sugar synthesis (Botella et al., 2021). Therefore, increased sugar content may enhance consumer preference.

Antioxidant enzymes levels of garden cress cultivars

Antioxidants, in the form of enzymes like CAT and SOD, are regarded as the primary defense against harmful oxidants (González-García et al., 2021). The investigation revealed the effects of salinity treatment on the concentrations of antioxidant enzymes, including GR, GST, CAT, POD, SOD, APX, G6PD, and 6GPD, in

the garden cress cultivars Dadaş and Bahar. The outcomes are summarized in Table 2. GST, G6PD, and 6GPD enzymes were found to be most abundant in both Dadaş and Bahar cress cultivars. However, the enzyme levels were higher in the Dadaş cress cultivar than in the Bahar cress cultivar. In fact, GST, G6PD, and APX enzymes were detected in amounts twice high in the Dadaş cress cultivar compared to the Bahar cress cultivar. The quantities of GST, G6PD, and 6GPD enzymes in the Dadaş cress cultivar were measured at 218.54, 102.75, and 86.73 EU/gr plant, respectively. Meanwhile, the Bahar cress cultivar exhibited enzyme amounts of 102.12, 49.69, and 54.29 EU/gr plant for GST, G6PD, and 6GPD, respectively. Salinity treatment increased the levels of antioxidant enzymes in both Dadaş and Bahar cress cultivars. For instance, the level of 6GPD rose from 86.73 to 172.53 EU/gr plant, POD from 21.99 to 42.54 EU/gr plant, and SOD from 14.01 to 25.87 EU/gr plant in the Dadaş cress cultivar ($P \leq 0.01$). In the Bahar cress cultivar, there was more than a twofold increase all enzyme levels. Similarly to our study, Al-Sammarraie et al. (2020) reported a significant increase in the total APX amount in *Lepidium sativum* plants after applying 100 mM salinity for 6 days or more. Furthermore, in their study, Pérez-Labrada et al. (2019) found that the application of 50 mM salinity to tomatoes resulted in a substantial increase in the levels of APX, SOD, and CAT enzymes. Likewise, González-García et al. (2021) reported that exposure to saltwater stress augmented the activity of enzymes including glutathione peroxidase, phenylalanine ammonia lyase and glutathione in bell pepper plants. When plants are subjected to saltwater stress, their stomatal conductance, transpiration, and CO₂ availability decrease, leading to alterations in the photosynthetic process. Consequently, oxidative stress is induced as a result of increased production and presence of reactive oxygen species (ROS) and/or reactive nitrogen species (RNS) (Pérez-Labrada et al., 2019). Finally, all of these factors alter both the yield of the fruit and the levels of bioactive compounds present.

Table 1. Sugar contents of garden cress cultivars by using HPLC (g 100 g⁻¹).

Cultivar	Salt (mM)	Sucrose	Glucose	Fructose	Rhamnose	Galactose	Xylose	Arabinose
Dadaş	0	0.62 ± 0.03 ^f	1.14 ± 0.03 ^c	2.75 ± 0.28 ^b	2.77 ± 0.06 ^c	0.46 ± 0.02 ^f	3.68 ± 0.05 ^e	3.59 ± 0.20 ^d
	30	6.70 ± 0.15 ^b	1.30 ± 0.01 ^c	2.48 ± 0.23 ^b	2.84 ± 0.07 ^c	5.08 ± 0.11 ^b	5.06 ± 0.28 ^b	5.07 ± 0.13 ^b
	60	7.86 ± 0.20 ^a	2.53 ± 0.02 ^a	5.33 ± 0.35 ^a	7.69 ± 0.19 ^a	5.67 ± 0.13 ^a	5.95 ± 0.09 ^a	5.74 ± 0.15 ^a
Bahar	0	3.05 ± 0.32 ^e	1.13 ± 0.05 ^c	5.33 ± 0.36 ^a	1.59 ± 0.02 ^d	2.13 ± 0.09 ^e	2.82 ± 0.16 ^f	2.07 ± 0.09 ^e
	30	4.94 ± 0.15 ^d	1.84 ± 0.12 ^b	2.48 ± 0.22 ^b	2.61 ± 0.07 ^c	3.68 ± 0.08 ^d	3.96 ± 0.11 ^d	3.60 ± 0.16 ^d
	60	5.72 ± 0.14 ^c	2.67 ± 0.25 ^a	5.33 ± 0.36 ^a	5.47 ± 0.22 ^b	4.07 ± 0.12 ^c	4.52 ± 0.10 ^c	4.26 ± 0.06 ^c

Dadaş: a regional garden cress variety in Erzurum, Bahar: commonly consumed garden cress in Turkey. Different letters in the same column indicate statistical difference ($P \leq 0.01$).

Table 2. Antioxidant enzymes levels of garden cress cultivars by using HPLC (EU/gr plant).

Cultivar	Salt (mM)	GR	GST	G6PD	6GPD	CAT	POD	SOD	APX
Dadaş	0	9.53 ± 0.37 ^d	218.54 ± 4.74 ^d	102.75 ± 0.85 ^d	86.73 ± 4.51 ^e	12.89 ± 0.59 ^c	21.99 ± 0.54 ^e	14.01 ± 0.77 ^e	11.39 ± 0.10 ^d
	30	13.95 ± 0.74 ^b	296.13 ± 9.18 ^b	125.00 ± 3.45 ^c	150.55 ± 4.06 ^b	15.98 ± 0.32 ^b	33.35 ± 0.93 ^b	20.39 ± 1.89 ^b	13.35 ± 0.53 ^c
	60	18.09 ± 1.05 ^a	395.66 ± 7.59 ^a	191.62 ± 3.94 ^a	172.53 ± 2.42 ^a	22.45 ± 1.80 ^a	42.54 ± 1.11 ^a	25.87 ± 0.24 ^a	21.39 ± 1.13 ^a
Bahar	0	5.80 ± 0.43 ^e	102.12 ± 6.05 ^e	49.69 ± 0.63 ^f	54.29 ± 0.32 ^f	6.71 ± 0.26 ^d	14.29 ± 0.27 ^f	8.95 ± 0.27 ^f	5.55 ± 0.27 ^f
	30	10.48 ± 0.08 ^d	211.04 ± 8.19 ^d	90.79 ± 2.57 ^e	104.59 ± 2.31 ^d	11.62 ± 0.23 ^c	24.83 ± 0.77 ^d	15.66 ± 0.23 ^d	9.71 ± 0.38 ^e
	60	12.91 ± 0.16 ^c	270.69 ± 2.52 ^c	140.26 ± 2.04 ^b	122.28 ± 5.10 ^c	17.33 ± 0.43 ^b	29.72 ± 1.30 ^c	18.57 ± 0.47 ^c	15.55 ± 0.35 ^b

Dadaş: a regional garden cress variety in Erzurum, Bahar: commonly consumed garden cress in Turkey. Different letters in the same column indicate statistical difference ($P \leq 0.01$). GR: Glutathione reductase, GST: Glutathione S-transferase, CAT: Catalase, POD: Peroxidase, SOD: Superoxide dismutase, APX: Ascorbate peroxidase, G6PD: Glucose- 6-phosphate dehydrogenase, 6GPD: 6-phosphogluconate dehydrogenase.

Vitamin contents of garden cress cultivars

In this study, the amounts of vitamins A, B1, B2, B6, and C in garden cress species were determined, and the results are presented in Figure 1. Garden cress is noted for its high vitamin A and C contents. The highest and lowest vitamin amounts for both Dadaş and Bahar garden cress cultivars were found to be B2 and C vitamins, respectively. In comparison, the Dadaş garden cress variety had a higher vitamin content for all vitamins detected than the Bahar garden cress variety. The salinity treatment led to increased vitamin content in both garden cress cultivars, with higher salt concentrations resulted in significantly greater vitamin levels ($P \leq 0.01$). As noted for the untreated garden cress varieties, vitamins B2 and C were present in the highest and lowest amounts, respectively, in both cultivars following the salinity treatment. The amount of vitamin B2 in the Dadaş garden cress cultivar increased from 84.59 mg kg⁻¹ to 142.26 mg kg⁻¹, while the amount of vitamin C increased from 8.59 mg kg⁻¹ to 13.49 mg kg⁻¹ as a result of the salinity treatment. For the Bahar garden cress cultivar, the amount of vitamin B2 increased from 42.03 mg kg⁻¹ to 95.73 mg kg⁻¹, and the amount of vitamin C rose from 4.87 mg kg⁻¹ to 9.65 mg kg⁻¹ due to the

salinity treatment. In general, the application of salinity treatment increased vitamin levels in garden cress varieties by more than 1.5 times. Similarly to our study, Islam et al. (2019) stated that the application of 12.5 mM NaCl resulted in an approximately 1.17-fold increase in the amount of vitamin C in wheat microgreen extracts. Likewise, Sarker et al. (2022) reported that increasing salinity application led to higher levels of vitamin C in *Amaranthus* leaves. However, Botella et al. (2021) indicated that salinity treatment did not result in increased vitamin C levels in tomatoes. In contrast, the combined use of salinity and heat applications did enhance the vitamin C levels in tomatoes. Additionally, the application of Cu NPs to the tomato plant under salinity conditions raised vitamin C levels (Pérez-Labrada et al., 2019).

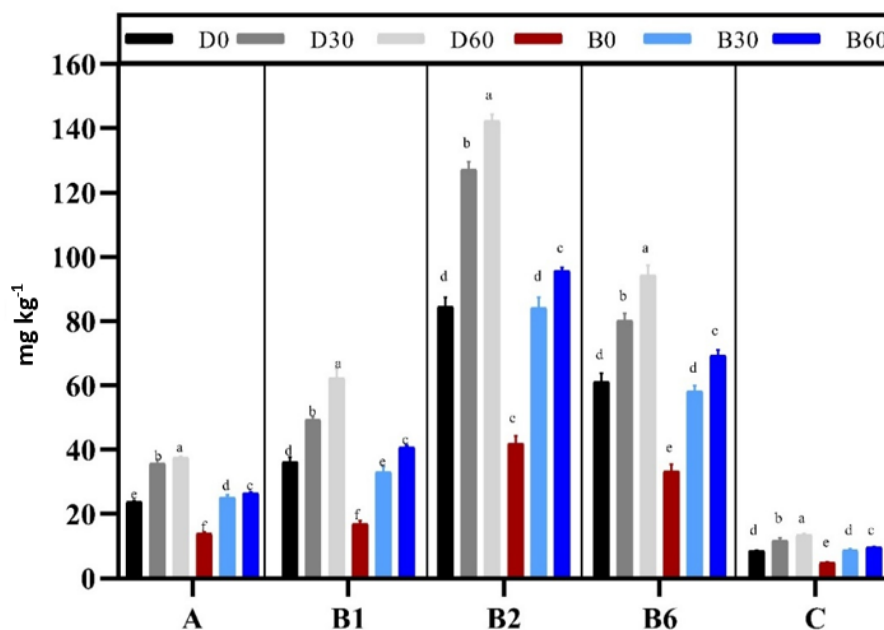


Figure 1. Vitamin contents of garden cress cultivars by using HPLC. D0: Dadaş garden cress 0 mM salinity treatment, D30: Dadaş garden cress 30 mM salinity treatment, D60: Dadaş garden cress 60 mM salinity treatment, B0: Bahar garden cress 0 mM salinity treatment, B30: Bahar garden cress 30 mM salinity treatment, B60: Bahar garden cress 60 mM salinity treatment. Different letters in the same column indicate statistical difference ($P < 0.01$).

Amino acid composition of garden cress cultivars

Table 3 summarizes the amino acid content of garden cress cultivars. Aspartate, arginine, and serine were detected at the highest levels in both Dadaş and Bahar garden cress cultivars, while glycine, glutamine, and phenylalanine were found at the lowest levels. Overall, the research found that Dadaş garden cress had a higher concentration of individual amino acids compared to Bahar garden cress. Nevertheless, only the levels of arginine, alanine, and tryptophan were higher in Bahar garden cress. Essential amino acids were detected at varying levels in both cultivars. In both Dadaş and Bahar garden cress cultivars, histidine was the essential amino acid with the highest concentration, whereas phenylalanine had the lowest concentration. Furthermore, the Dadaş cress cultivar is rich in essential amino acids, with higher quantities than the Bahar cress cultivar for all essential amino acids, except tryptophan. Divergent outcomes were observed in amino acid levels following salinity treatment. The application of 30 mM and 60 mM salinity to Dadaş and Bahar garden cress cultivars resulted in reduced levels of aspartate, glutamate, asparagine, serine, alanine, cystine, leucine, and sarcosine. Furthermore, as the concentration of salt increased, the amounts of these amino acids decreased. While the 30 mM salinity treatment resulted in a decreased levels of glutamine, tyrosine, valine, and isoleucine, the 60 mM salinity treatment increased their levels. The amounts of histidine and glycine were found to be higher than those of the control group due to the 60 mM salinity treatment. Hassan et al. (2011) identified the individual amino acids present in garden cress, with glutamic detected at the highest level and cystine at the lowest. Joseph et al. (2015) and Benitez et al. (2016) reported that salinity treatment enhanced proline accumulation in rice. A study on *Arabidopsis* demonstrated that the contents of alanine, arginine, glycine, histidine, isoleucine, leucine, lysine, phenylalanine, proline, serine, threonine, tyrosine, and valine increased in response to abiotic stress (salt), while the contents of asparagine, aspartic acid, glutamine, and glutamic acid decreased (Hildebrandt, 2018). Patel et al. (2022) exposed peanut plants to various abiotic stress

conditions. Salinity treatment resulted in increased levels of valine, proline, glycine, serine, and phenylalanine but decreased levels of threonine, aspartic acid, glutamic acid, asparagine, glutamine, hydroxyproline, and lysine. Numerous studies in the literature demonstrate that abiotic stress conditions lead to an increase in proline amino acid content. However, our research revealed that the amount of proline in Dadaş garden cress decreased following exposure to salinity, while the proline content in Bahar garden cress increased with 30 mM salinity application but decreased with 60 mM salinity application.

Table 3. Amino acids composition of garden cress cultivars by using HPLC (pmol μL^{-1}).

Salt (mM)	Dadaş			Bahar		
	0	30	60	0	30	60
Aspartate	1445.82 \pm 26.70	1312.18 \pm 47.23	1247.66 \pm 65.98	1331.48 \pm 35.32	1142.94 \pm 58.26	984.51 \pm 7.61
Glutamate	234.66 \pm 5.00	153.00 \pm 3.88	142.10 \pm 6.13	192.90 \pm 1.44	164.00 \pm 3.81	113.99 \pm 4.39
Asparagine	355.61 \pm 7.17	257.47 \pm 2.60	247.78 \pm 8.71	321.88 \pm 11.06	266.25 \pm 5.41	255.23 \pm 5.56
Serine	555.34 \pm 7.36	429.58 \pm 20.97	385.88 \pm 11.02	521.83 \pm 24.98	519.43 \pm 6.08	451.62 \pm 9.75
Glutamine	141.11 \pm 6.07	113.12 \pm 10.77	115.62 \pm 12.84	114.07 \pm 6.09	54.34 \pm 3.95	60.50 \pm 1.24
Histidine	521.40 \pm 24.15	493.07 \pm 15.78	546.91 \pm 16.57	469.27 \pm 38.98	475.50 \pm 11.38	489.30 \pm 19.70
Glycine	57.35 \pm 5.10	47.69 \pm 3.46	67.64 \pm 2.20	32.70 \pm 1.43	59.81 \pm 0.60	66.97 \pm 2.34
Threonine	316.76 \pm 11.77	278.28 \pm 55.60	256.31 \pm 25.32	291.82 \pm 32.71	192.93 \pm 15.75	217.52 \pm 37.78
Arginine	1193.85 \pm 152.23	1209.12 \pm 170.83	1019.99 \pm 12.35	1336.55 \pm 184.64	1068.21 \pm 144.64	977.57 \pm 227.02
Alanine	253.00 \pm 54.43	203.64 \pm 47.37	182.64 \pm 27.13	262.82 \pm 43.59	234.41 \pm 14.01	200.57 \pm 13.73
Tyrosine	269.88 \pm 12.83	207.71 \pm 12.16	249.55 \pm 26.30	201.09 \pm 28.03	117.81 \pm 18.65	129.17 \pm 28.97
Cystine	260.93 \pm 41.05	227.73 \pm 60.08	203.62 \pm 11.95	220.58 \pm 34.68	202.71 \pm 37.14	150.46 \pm 26.17
Valin	458.77 \pm 91.15	334.34 \pm 20.39	365.60 \pm 61.47	340.27 \pm 48.26	216.96 \pm 32.58	236.93 \pm 41.50
Methionine	290.41 \pm 34.56	207.20 \pm 7.17	236.23 \pm 19.47	272.18 \pm 50.69	298.47 \pm 27.69	235.96 \pm 29.79
Tryptophane	238.11 \pm 44.94	193.96 \pm 77.21	229.71 \pm 10.85	336.57 \pm 184.52	372.05 \pm 132.71	371.91 \pm 85.97
Phenylalanine	191.31 \pm 17.09	148.76 \pm 20.81	152.81 \pm 36.98	146.99 \pm 19.51	133.26 \pm 4.00	118.01 \pm 13.08
Isoluecine	351.48 \pm 17.12	277.88 \pm 44.11	282.00 \pm 34.56	284.94 \pm 13.70	218.79 \pm 7.88	219.86 \pm 16.67
Leucine	212.81 \pm 9.26	184.02 \pm 53.15	180.39 \pm 26.91	170.96 \pm 17.26	170.17 \pm 6.60	158.07 \pm 17.42
Lysine	286.65 \pm 41.13	213.27 \pm 24.52	232.59 \pm 64.78	208.52 \pm 40.09	153.12 \pm 13.60	151.33 \pm 20.16
Hydroxyproline	207.87 \pm 6.83	194.40 \pm 44.92	185.76 \pm 25.29	180.11 \pm 72.68	76.12 \pm 9.09	81.37 \pm 13.76
Sarcosine	330.26 \pm 56.91	275.14 \pm 39.43	229.52 \pm 33.18	287.98 \pm 43.34	188.36 \pm 14.98	174.30 \pm 10.01
Proline	285.83 \pm 31.87	241.53 \pm 51.50	210.91 \pm 44.01	284.38 \pm 42.76	301.74 \pm 9.86	253.53 \pm 33.94

Dadaş: a regional garden cress variety in Erzurum, Bahar: commonly consumed garden cress in Turkey.

Individual phenolic compounds of garden cress cultivars

Phenolic compounds are responsible for properties such as color and aroma and are involved in plant defense mechanisms. Extensive research is being conducted to reveal the beneficial effects of these compounds on various diseases, particularly cancer and heart disease (González-Chavira et al., 2018). The individual phenolic compound contents of garden cress cultivars are presented in Table 4. The level of phenolic compounds in the Dadaş garden cress cultivar was found to be higher than that in the Bahar garden cress cultivar for the individual phenolic compounds analyzed. Among the detected phenolic compounds, kaftaric acid ($9.13 \pm 0.09 \mu\text{g L}^{-1}$) and tyrosol ($4.22 \pm 0.52 \mu\text{g L}^{-1}$) were the predominant phenolic compounds in the Dadaş and Bahar garden cress cultivars, respectively.

Table 4. Individual phenolic compounds of garden cress cultivars by using HPLC ($\mu\text{g L}^{-1}$).

Phenolic compounds	Dadaş			Bahar		
	0 (mM)	30 (mM)	60 (mM)	0 (mM)	30 (mM)	60 (mM)
Gallic acid	5.81 \pm 0.19 ^d	7.06 \pm 0.21 ^c	10.73 \pm 0.05 ^a	2.52 \pm 0.33 ^f	4.91 \pm 0.26 ^e	7.65 \pm 0.14 ^b
Vanillic acid	7.61 \pm 0.24 ^c	8.99 \pm 0.18 ^b	13.29 \pm 0.13 ^a	3.50 \pm 0.39 ^e	5.98 \pm 0.10 ^d	8.98 \pm 0.15 ^b
Trans-caffeic acid	4.71 \pm 0.10 ^d	5.74 \pm 0.06 ^c	8.50 \pm 0.25 ^a	2.22 \pm 0.16 ^f	4.00 \pm 0.20 ^e	6.16 \pm 0.08 ^b
Trans-p-coumaric acid	5.83 \pm 0.22 ^c	8.18 \pm 0.29 ^b	10.87 \pm 0.32 ^a	3.07 \pm 0.18 ^d	6.04 \pm 0.26 ^c	7.90 \pm 0.23 ^b
Ferulic acid	1.84 \pm 0.04 ^b	1.88 \pm 0.03 ^b	1.98 \pm 0.06 ^a	1.43 \pm 0.03 ^d	1.56 \pm 0.03 ^c	2.04 \pm 0.07 ^a
Kaftaric acid	9.13 \pm 0.09 ^c	9.75 \pm 0.30 ^c	16.03 \pm 0.52 ^a	4.08 \pm 0.45 ^e	6.87 \pm 0.17 ^d	11.66 \pm 0.38 ^b
Catechin	9.00 \pm 0.52 ^c	9.43 \pm 0.35 ^c	15.79 \pm 1.09 ^a	3.78 \pm 0.18 ^e	6.61 \pm 0.16 ^d	11.48 \pm 0.79 ^b
Epicatechin	4.95 \pm 0.08 ^c	6.89 \pm 0.07 ^b	9.32 \pm 0.40 ^a	2.39 \pm 0.22 ^d	4.70 \pm 0.09 ^c	6.64 \pm 0.20 ^b
Quercetin	3.74 \pm 0.23 ^b	3.76 \pm 0.08 ^b	3.99 \pm 0.15 ^b	3.15 \pm 0.10 ^c	3.16 \pm 0.15 ^c	4.35 \pm 0.10 ^a
Rutin	3.64 \pm 0.06 ^d	4.12 \pm 0.19 ^c	6.22 \pm 0.11 ^a	1.75 \pm 0.02 ^f	2.99 \pm 0.14 ^e	4.71 \pm 0.20 ^b
Myricetin	2.56 \pm 0.13 ^d	3.16 \pm 0.06 ^c	6.29 \pm 0.10 ^a	1.31 \pm 0.06 ^f	2.30 \pm 0.05 ^e	4.57 \pm 0.21 ^b
Tyrosol	8.64 \pm 0.37 ^d	10.02 \pm 0.11 ^c	16.94 \pm 1.01 ^a	4.22 \pm 0.52 ^f	7.46 \pm 0.24 ^e	12.32 \pm 0.73 ^b

Dadaş: a regional garden cress variety in Erzurum, Bahar: commonly consumed garden cress in Turkey. Different letters in the same column indicate statistical difference ($P \leq 0.01$).

On the other hand, ferulic acid ($1.84 \pm 0.04 \mu\text{g L}^{-1}$) and myricetin ($1.31 \pm 0.06 \mu\text{g L}^{-1}$) were the least detected phenolic compounds in the Dadaş and Bahar garden cress cultivars, respectively. Similar to our study, vanillic acid, caffeic acid, coumaric acid and ferulic acid were identified as individual phenolic compounds in garden cress (Elguera et al., 2013). Additionally, gallic acid, ferulic acid, and quercetin were reported in another study (Santos et al., 2014). Furthermore, from the enzymatic extract of garden cress seed meal, gallic acid, ferulic acid, catechin, quercetin, and rutin were identified (Younos & Akl, 2022). Many factors, such as cultivar, environmental conditions, water availability, germination, and maturity, affect the phenolic compounds of plants (Santos et al., 2014). Salinity treatment caused a substantial increase in the quantity of phenolic compounds in all garden cress cultivars ($P \leq 0.01$), although the increase in quercetin for the Dadaş garden cress cultivar was not statistically significant ($P > 0.01$). After treating for salinity, the amount of phenolic compounds increased to varying degrees depending on the compound. With a 60 mM salinity treatment, an increase of approximately 1.5 to 3 times the initial amounts of individual phenolic compounds was detected. Myricetin was identified as the phenolic compound with the highest increase for both Dadaş and Bahar garden cress cultivars. In contrast, there was only a slight increase in ferulic acid and quercetin for both garden cress cultivars. Following the 60 mM salinity treatment, tyrosol was detected at the highest levels among the phenolic compounds for Dadaş and Bahar garden cress cultivars (16.94 ± 1.01 and $12.32 \pm 0.73 \mu\text{g L}^{-1}$, respectively). Abiotic stress conditions may lead to an increase in phenolic compounds by affecting the phenylpropanoid biosynthetic pathway (Graziani et al., 2022). Islam et al. (2019) reported that 12.5 and 25 mM salinity applications increased the total phenolic contents of wheat microgreen extract, while 50 mM and 100 mM salinity application decreased total phenolic contents. Botella et al. (2021) noted that salinity treatment slightly decreased the concentrations of hydroxycinnamic acid, flavanones and phloretin, increased flavonols, and did not affect the concentration of homovallinic acid in tomato fruits.

Anthocyanins contents of garden cress cultivars

Table 5 provides the individual anthocyanin contents of garden cress cultivars. Both Dadaş and Bahar garden cress cultivars exhibited the highest levels of malvidin-3-glucoside-p-coumaryl (481.42 ± 80.90 , $\text{g } 100 \text{ g}^{-1}$), while levels of cyanidin-3-glucoside (5.89 ± 0.77 and 4.29 ± 0.56 , $\text{g } 100 \text{ g}^{-1}$, respectively) were the lowest for both cultivars. Generally, salinity treatment of garden cress cultivars resulted in a reduction of anthocyanin content. Furthermore, with increased salt concentration, the reduction in anthocyanin content became more pronounced.

Table 5. Anthocyanins contents of garden cress cultivars by using HPLC ($\text{g } 100 \text{ g}^{-1}$).

	Dadaş			Bahar		
	0 (mM)	30 (mM)	60 (mM)	0 (mM)	30 (mM)	60 (mM)
Delphinidin-3-glucoside	14.63 ± 0.76	8.84 ± 0.96	7.62 ± 0.77	10.09 ± 0.53	6.10 ± 0.66	3.14 ± 0.42
Cyanidin-3-glucoside	5.89 ± 0.77	4.20 ± 0.06	3.25 ± 0.58	4.29 ± 0.56	3.05 ± 0.04	1.74 ± 0.05
Petunidin-3-glucoside	19.15 ± 1.98	13.91 ± 1.11	10.45 ± 1.66	13.93 ± 1.44	10.12 ± 0.81	5.85 ± 0.62
Peonidin-3- glucoside	95.52 ± 10.18	67.58 ± 6.05	56.59 ± 9.21	65.85 ± 7.01	46.59 ± 4.17	21.12 ± 2.33
Malvidin-3-glucoside	153.30 ± 14.22	82.41 ± 1.18	83.50 ± 12.39	111.49 ± 10.34	59.93 ± 0.86	36.64 ± 1.22
Peonidin-3- glucoside acetyl	209.74 ± 7.21	128.09 ± 14.55	109.05 ± 5.99	144.59 ± 4.97	88.30 ± 10.03	45.44 ± 6.19
Malvidin-3-glucoside acetyl	98.27 ± 10.21	86.83 ± 7.16	53.77 ± 8.34	71.47 ± 7.42	63.15 ± 5.20	36.28 ± 2.07
Malvidin-3-glucoside-p-coumaryl	481.42 ± 80.90	139.10 ± 11.71	148.29 ± 16.60	481.42 ± 80.90	149.10 ± 11.71	403.21 ± 65.12

Dadaş: a regional garden cress variety in Erzurum, Bahar: commonly consumed garden cress in Turkey.

The 60 mM salinity treatment led to a decrease in the amount of delphinidin 3-glucoside, from 14.63 to $7.62 \text{ g } 100 \text{ g}^{-1}$ in the Dadaş cress variety. Similarly, a decrease in the amount of cyanidin-3-glucoside, from 5.89 to $3.25 \text{ g } 100 \text{ g}^{-1}$, as well as Malvidin-3-glucoside, from 153.30 to $83.50 \text{ g } 100 \text{ g}^{-1}$, was noted. The Bahar cress cultivar showed similar decreases in anthocyanin levels with salinity treatment. However, the Dadaş cress cultivar exhibited higher amounts of malvidin-3-glucoside and malvidin-3-glucoside-p-coumaryl with the application of 60 mM salinity compared to 30 mM salinity. Malvidin-3-glucoside increased from 82.41 ± 1.18 to $83.50 \pm 12.39 \text{ g } 100 \text{ g}^{-1}$, and malvidin-3-glucoside-p-coumaryl increased from 139.10 ± 11.71 to $148.29 \pm 16.60 \text{ g } 100 \text{ g}^{-1}$ for 30 and 60 mM salinity treatments, respectively. For the Bahar garden cress cultivar, a greater level of malvidin-3-glucoside-p-coumaryl was found at a salinity of 60 mM compared to 30 mM. Al-Sammarraie et al. (2020) reported a reduction in the total antioxidant level of garden cress with the application of 100 mM NaCl. However, an increase in antioxidant levels was observed with increasing application time. In a separate study, Islam et al. (2019) found a decrease in the total anthocyanin levels of

wheat microgreen extracts with the application of 12.5 and 100 mM NaCl. On the other hand, the application of 25 and 50 mM NaCl resulted in an increase in the total anthocyanin level of wheat microgreen extract. Anthocyanin biosynthesis occurs via the phenylpropanoid pathway (Islam et al., 2019), with potential impacts on anthocyanin release due to salinity treatment.

Conclusion

Cress contains antioxidant enzymes, vitamins, and phenolic compounds that have numerous positive effects on health. As a result, cress is frequently used as a spice and consumed in salads. While abiotic stress conditions can harm plants, leading to crop loss, in some cases, they can reveal beneficial substances or increase their concentrations. Controlled salinity treatment was administered to Bahar garden cress, which is commonly consumed in Turkey, and Dadaş cress, a regional variety from Erzurum. This treatment resulted in a significant increase in the levels of phenolic compounds, antioxidant enzymes, vitamins, and sugars in both cress varieties. However, a reduction in anthocyanin levels was observed. The findings indicate that salinity treatment can positively influence the nutritional and functional compounds of cress, leading to beneficial health effects. Additionally, this study will help predict the changes that may occur in products such as garden cress due to abiotic stresses exacerbated by climate change.

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