



Effect of Zeolite Supplementation on the Physio-Morphological and Chemical Characteristics of *Plectranthus scutellarioides* (L.) Grown under Different Irrigation Intervals

Samah M. El-Sayed¹, Asmaa E. Abd Elhafez², and Azza A.M. Mazhar¹

¹Ornamental Plants and Woody Trees Dept. Agricultural and Biological Research Institute, National Research Centre, Dokki, Giza, Egypt

² Botanical Gardens Research Dept., Horticulture Research Institute, Agricultural Research Center, Giza, Egypt

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ABSTRACT

Water scarcity is a serious threat on a global scale because of its detrimental effects on crop productivity generally and its detrimental effects on the aesthetic appeal and decorative value of many ornamental plants, such as *Plectranthus scutellarioides* (L.) plants. Therefore, our study aimed to use zeolite as a natural, eco-friendly substance at rates 0, 30 and 60g/ kg soil to help plants develop well when they are exposed to different irrigation intervals 3, 5 and 7 days. When *P. scutellarioides* plants were irrigated every 7 days had a detrimental effect on plant growth because it reduced the values of all vegetative characteristics, RWC (%), MSI (%), and photosynthetic pigments content. Conversely, this irrigation interval (7 days) increased the antioxidant activity (%), and the levels of phenols, flavonoids, proline and lipid peroxidation increased as a result. The highest sugars content occurred when the plants were watered every 5 days. Whilst, the majority of the vegetative characteristics and chemical contents improved when the plants were treated with 30 g/kg zeolite, with the exception of the fresh and dry weights of the roots, which improved when the plants were treated with 60 g/kg zeolite at all irrigation levels. In conclusion, *P. scutellarioides* plants were found to be negatively impacted by longer drought periods; however, the application of zeolite at a level of 30 g/kg soil improved the morphological and chemical properties of the plants under all irrigation levels.

Keywords: *Plectranthus scutellarioides*, Coleus, lipid peroxidation, phenols, flavonoids, irrigation intervals, membrane stability, relative water content.

Introduction

Plectranthus scutellarioides (L.), also referred to as Coleus, is a species of leaf plant that is native to Southeast Asia and Australia. It belongs to the Lamiaceae family, which is also known as the mint or deadnettle family. It is grown all over the world and can spread to additional tropical areas (Nguyen *et al.*, 2008). Wild species may have moderately variegated leaves, but cultivated cultivars have evolved this to an extreme degree, with leaves that can be green, white, cream, yellow, pink, crimson, maroon, or dark purple in one or more colors (Nguyen and Dal Cin, 2009). The quantity of chlorophyll present in the chloroplasts of leaves is what gives them a green appearance. In addition to chlorophyll, anthocyanins water-soluble, flavonoid-biosynthetic pigments are responsible for the colors red, purple, pink, and orange. *P. scutellarioides* is commonly used as an ornamental plant with a changeable leaf color and rich leaf shape (Li *et al.*, 2021). Coleus is a perennial herbaceous foliage plant with a long history of medicinal and food uses (Desai and Thirumala, 2014). In Papua New Guinea, it is used as a food additive, while in Southeast Asia, it is considered a medicinal plant and is used to treat a variety of ailments, including dyspepsia, ophthalmia, and wound infections (Desai and Thirumala, 2014; Kartini *et al.*, 2025). Because of the therapeutic qualities of the extract of *P. scutellarioides* and its

Corresponding Author: Samah M. El-Sayed, Ornamental Plants and Woody Trees Dept. Agricultural and Biological Research Institute, National Research Centre, Dokki, Giza, Egypt
E-mail: - ensamah_83@hotmail.com

antioxidant presence, gold particles with the extract have been utilized in nanosynthesis to treat some malignant tumors (Al-Mafarjy *et al.*, 2024).

Drought is one of the major environmental problems worldwide (Zhang *et al.*, 2024) and is detrimental to the growth of ornamental plants because of the increased frequency and severity of drought caused by global climate change. It is still unclear how ornamentals will change physiologically and morphologically when they are produced with little irrigation and fewer water resources. Since more focus is placed on the aesthetic appeal of landscape plants that are frequently employed in recreational areas, it is not beneficial for these plants to be exposed to water stress. Many plants will be impacted by global warming in the future, according to potential climate change scenarios (Demirel *et al.*, 2020). Studies on plants resistant and tolerant to environmental stress factors, particularly those that are drought-tolerant, are critical because cultivation is extremely challenging in situations where the water supply is limited (Fernandez *et al.*, 2020). Numerous studies have shown that longer irrigation intervals result in longer drought exposure times for the plant, which in turn causes a decline in morphological traits and modifications to physiological processes, ultimately leading to decreased productivity and lower yields, as demonstrated in *Salvia officinalis*, *Mentha piperita*, and *Andrographis paniculata* plants (Hazrati *et al.*, 2022; El-Naggar *et al.*, 2022; Rusmayadi *et al.*, 2025). Drought primarily results in decreased leaf water capacity, which decreases photosynthetic activity through stomatal closure, membrane damage, and the accumulation of reactive oxygen species (Zulfiqar *et al.*, 2021).

Zeolites were discovered in 1756 by Fredrich Cronstedt, a Swedish mineralogist (Polat *et al.*, 2004). Zeolites, hydrated alumina silicate crystals with high cationic exchange capacity, maintain moisture for a long period and improve soil physical conditions (Caspersen and Ganrot, 2018). Owing to their high porosity and crystalline structure, zeolites can absorb up to 60% of their weight (Polat *et al.*, 2004). The positive impact of zeolite on morphological, physiological, biochemical and yield parameters has already been reported in drought-exposed rice (Zheng *et al.*, 2018) and *Aloe vera* (Hazrati *et al.*, 2017). Zeolite plays a vital role in improving soil properties by enhancing water retention and nutrient availability, contributing to better plant health and resilience against water stress (Elawady, 2024). The application of zeolite as a soil amendment can lead to notable improvements in crop yield (AbdEL-Azeiz & Elsonbaty, 2024).

The aim of this work was to study the response of *Plectranthus scutellarioides* (L.) plants to different irrigation intervals, using zeolite as a natural enhancer to reduce the harmful effects when the plant's water supply is insufficient.

2. Methods

2.1. Experiment site

During two consecutive growing seasons in 2023 and 2024, the experiment was conducted at the nursery of the Botanical Gardens Research Department, Horticulture Research Institute (HRI), Agricultural Research Center (ARC), Giza, Egypt, and the chemical assessments were implemented in the laboratory of the Ornamental Plants and Woody Trees Dep., Agricultural and Biological Research Institute, National Research Centre (NRC), Egypt.

2.2. Preparing plants, experimental layout and applying treatments

Plectranthus scutellarioides seedlings with a height of 20- 25cm were obtained from the nursery of the Botanical Gardens Research Department, HRI, ARC, Giza, Egypt. The seedlings were repotted in May 2023 and 2024 in 30 cm diameter pots (one plant/pot). The pots were filled with soil consisting of clay and sand, which was analyzed according to the methods of Jackson (2005) (Table 1), and mixed with the zeolite ground into a fine powder before transplanting. The zeolite and the analysis of its characteristics (Table 2) were obtained from the fertilizer sales outlet of the Egyptian Ministry of Agriculture and Land Reclamation. The experiment was conducted as a factorial experiment in complete block design with 9 treatments with 3 replication for each treatment. The plants were watered on the basis of the proposed irrigation schedules (every 3, 5 and 7 days) and interacted with the suggested zeolite treatments (0, 30 and 60g/ kg soil), with the treatments being in the following order:

1. D₃Z₀: Irrigation interval (3 days) +zeolite 0g/ kg soil
2. D₃Z₃₀: Irrigation interval (3 days) + zeolite 30 g/kg soil
3. D₃Z₆₀: Irrigation interval (3 days) + zeolite 60 g/kg soil
4. D₅Z₀: Irrigation interval (5 days) +zeolite 0g/ kg soil
5. D₅Z₃₀: Irrigation interval (5 days) + zeolite 30 g/kg soil
6. D₅Z₆₀: Irrigation interval (5 days) + zeolite 60 g/kg soil
7. D₇Z₀: Irrigation interval (7 days) +zeolite 0g/ kg soil
8. D₇Z₃₀: Irrigation interval (7 days) + zeolite 30 g/kg soil
9. D₇Z₆₀: Irrigation interval (7 days) + zeolite 60 g/kg soil.

Table 1: Texture and chemical characterization of the soil sample

Soil Sample	Fine sand (%)	Coarse sand (%)	Silt%	Clay%					
	40.70	44.10	10.00	5.20					
Sand clay	pH (1:2.5)	EC (dS/m)	Anion (meq/l)		Cation (meq/l)				
			HCO ₃ ⁻	Cl	SO ₄ ⁻²	Ca ⁺⁺	Mg ⁺⁺	Na ⁺	K ⁺
	7.80	3.53	0.79	32.70	1.51	9.70	5.60	19.00	0.70

Table 2: Chemical analysis of zeolite

Chemical component	SiO ₂	TiO ₂	Al ₂ O ₃	Fe ₂ O ₃	K ₂ O	CaO	MgO	Na ₂ O
(%)	68.15	0.20	12.30	1.30	2.8	3.95	0.90	0.80

2.3. Recorded data

2.3.1. Morphological measurements

Plant height (cm), stem girth (cm), number of leaves, number of branches, leaf area (cm²), root length (cm), shoot fresh weight (g), shoot dry weight (g), root fresh weight (g), and root dry weight (g) were measured.

2.4. Membrane stability index (MSI %) and relative water content (RWC %)

The electrical conductivity of the leaf ions was used to compute the MSI. After 0.1 g of leaf discs were carefully washed in tap and double-distilled water, they were submerged in 10 ml of double-distilled water at 40 °C for 30 minutes. An electrical conductivity (EC) meter (C1) was used to record the electrical conductivity of the samples after the period had passed. The identical samples were then placed in a boiling water bath (100 °C) for ten minutes, and their electrical conductivity was recorded as previously mentioned (C2). The following equation was used to determine the membrane stability index (Sairam *et al.*, 1997):

$$MSI (\%) = (1 - (C1/C2)) \times 100$$

At the conclusion of each season, samples were collected from the fully grown new leaves to determine the RWC. After the fresh weight (FW) of the leaves was recorded, the leaves were submerged in deionized water for four hours. The moist surface of the turgid leaf was rapidly blotted dry prior to weighing (TW). The leaves were subsequently oven-dehydrated at 70 °C, after which their dry weight (DW) was determined. The following formula was used to determine the RWC (Lugojan & Ciulca, 2011):

$$RWC (\%) = (FW - DW / TW - DW) \times 100.$$

2.5. Chemical constituents

2.5.1. Photosynthetic pigments (mg/g FW)

In accordance with Lichtenthaler & Wellburn (1983), photosynthetic pigments (chlorophyll a, b and carotenoids) were measured by crushing 0.1 g of fresh leaves with 10 ml of 80% acetone (v/v). A UV-Vis spectrophotometer was used to measure the supernatant at wavelengths of 663, 647, and 470 nm.

2.5.2. Preparation of alcoholic extract

Fresh leaf material (0.4 g) was homogenized in 10 mL of 70% ethanol and shaken for 24 hours to yield the alcoholic plant extract (Farahani *et al.*, 2025). The total phenol content, total flavonoids, total sugars, and antioxidant activity were measured after the solution was collected.

2.5.3. Total flavonoids (mg of RE/g FW)

The aluminum chloride (AlCl₃) colorimetric method were used (Chang *et al.*, 2002). A solution of ethanolic rutin was prepared to obtain a calibration curve. Two milliliters of methanolic leaf tissue extract was mixed with 2 mL of 2% AlCl₃ and incubated for 30 min. at room temperature, and the absorbance was measured at 415 nm using UV-Vis spectrophotometer. mg of rutin equivalents per g of fresh weight (mg of RE/g FW) was used to express the overall flavonoid content.

2.5.4. Total phenols (mg of GAE/g FW)

The Singleton & Rossi (1965) method was used to estimate the total phenol content. Folin-Ciocalteu reagent and sodium carbonate (NaHCO₃) at a 14% concentration were added to the ethanol extract of fresh leaf tissue. Using the gallic acid standard curve (mg of GAE/g FW), the optical density was determined at a wavelength of 765 nm via a UV-Vis spectrophotometer.

2.5.5. DPPH radical scavenging assay for determination of antioxidant activity (%)

The 2,2-diphenyl-1-picrylhydrazyl (DPPH) test was used to assess the ability of the extracts to scavenge free radicals (Zhu *et al.*, 2009). A 0.1 ml ethanol extract of the sample was combined with a 0.1 ml DPPH solution. The absorbance at 517 nm was measured after 30 minutes of incubation in the dark at room temperature. The radical scavenging activity (antioxidant activity %) was computed as:

$$\text{Antioxidant activity (\%)} = (A_0 - A_t / A_0) \times 100$$

where A₀ represents the absorbance of the control sample and A_t represents the absorbance of the treated sample.

2.5.6. Total sugars (mg/g FW)

In accordance with Dubois *et al.* (1956), the sulfuric-phenol colorimetric method was used to determine the total sugars concentration. One millilitre of 5% phenol solution was added to one millilitre of the ethanolic extract of the plant sample; five millilitres of concentrated solution were then added, properly mixed, and left for fifteen minutes. A UV-Vis spectrophotometer was used to measure the absorbance of the yellow-orange color at a wavelength of 490 nm. Different concentrations of pure glucose were used to create a standard curve.

2.5.7. Proline (µg/g FW)

The method described by Bates *et al.* (1973) was used to determine the proline content. After a fresh leaf sample (0.5 g) was homogenized in 10 mL of 3% sulfosalicylic acid, the sample was centrifuged for 20 minutes at 4 °C at 10,000 rpm. Two milliliters of the supernatant were combined with two milliliters of glacial acetic acid and two milliliters of acid ninhydrin solution (made by dissolving 1.25 g of ninhydrin in 30 mL of glacial acetic acid and adding 20 mL of 6 M phosphoric acid). After one hour of incubation in a hot water bath at 95 °C, the mixture was immediately transferred to an ice bath. The mixture was then vortexed for 20 seconds after 4 mL of toluene was added. A UV-Vis spectrophotometer was used to separate the chromophore-containing toluene phase, and the absorbance was measured at 520 nm.

2.5.8. Lipid peroxidation (MDA $\mu\text{mol/g FW}$)

In accordance with Rao and Sresty (2000), lipid peroxidation ($\mu\text{M/g F.W.}$) was calculated. Lipid peroxidation was assessed via the thiobarbituric acid (TBA) test, which measures the amount of malondialdehyde (MDA). Five percent trichloroacetic acid (TCA) was used to extract fresh leaf samples (0.5 g), which were then centrifuged for ten minutes at 4000 rpm. Two milliliters of a 0.6% thiobarbituric acid (TBA) solution were combined with two milliliters of the extract. After 30 minutes of heating the final volume in a water bath at 95 °C, the absorbance of the generated color was measured at 532, 600, and 450 nm.

2.5.9. Data analysis

The average of the obtained data for each treatment was subjected to statistical analysis via CoStat (CoHort software, Monterey, CA, USA), where the treatment means and standard deviation ($\pm\text{SD}$) were compared for significance via Duncan's new multiple range test (DMRT), where $p < 0.05$ (Duncan, 1955). The graphs were created via Microsoft Excel 2013.

3. Results

3.1. Vegetative growth

The results recorded in Figure (1 A-J) explained the response of a *Plectranthus scutellarioides* plant to irrigation at different intervals. During the two seasons revealed that a significant decrease was detected in most plant traits as a result of increasing intervals between irrigations; the greatest reduction in all morphological parameters occurred at the time of irrigation every 7 days. On the contrary, the highest values for most of these parameters were obtained due to irrigation every three days, with 83.92 ± 3.49 and 87.47 ± 3.38 cm for plant height, 68.00 ± 3.77 and 61.45 ± 4.42 for the number of leaves/plant, 6.00 ± 0.91 and 6.11 ± 0.53 for the number of branches/plant 27.66 ± 2.12 and 29.52 ± 2.40 cm² for the leaf area, 25.83 ± 2.66 and 26.12 ± 2.44 cm for the root length, 108.46 ± 4.79 and 113.00 ± 4.17 g for the shoot fresh weight and 12.15 ± 0.80 and 12.67 ± 0.77 g for the shoot dry weight, respectively, in both seasons. On the other hand, the highest values of stem girth (3.32 ± 0.30 and 3.45 ± 0.24 cm), fresh weight of roots (14.84 ± 0.75 and 15.61 ± 0.81 g) and dry weight of roots (3.67 ± 0.52 and 4.16 ± 0.38 g) were obtained when the plants were irrigated every five days during 2023 and 2024 seasons, respectively. The data in Figure (2 A-J) illustrate that zeolite had different effects on all the vegetative growth parameters. The highest values of plant height (80.98 ± 3.20 and 84.39 ± 3.45 cm), stem girth (3.27 ± 0.27 and 3.33 ± 0.25 cm), number of leaves (67.67 ± 3.78 and 61.89 ± 4.05), number of branches (5.47 ± 0.91 and 6.22 ± 0.72), root length (25.91 ± 2.37 and 28.98 ± 2.54 cm), shoot fresh weight (104.47 ± 4.72 and 107.93 ± 3.83 g) and shoot dry weight (11.18 ± 0.82 and 11.61 ± 0.65 g) were detected, respectively, in the two seasons when plants treated with zeolite at rate 30g/ kg soil. However, the highest values of root fresh weight (14.86 ± 0.75 and 15.63 ± 0.67 g) and root dry weight (3.69 ± 0.43 and 4.16 ± 0.39 g) were obtained with the application of zeolite at 60 g/kg soil. The leaf area (cm²) was the largest when plants treated with zeolite at 60 g/kg, followed by zeolite at 30 g/kg without any significant differences between both treatments, respectively, in both seasons.

The data inserted in Tables (3& 4) revealed that the plant height increased significantly in the first season when the plants were irrigated every 3 days and treated with zeolite at a rate of 60 g/kg soil, while in the second season when the plants were irrigated every 3 days and treated with zeolite at 30 g/kg soil. No. of leaves, No. of branches, root length, shoot fresh and dry weight significantly affected in plants treated with irrigation every 3 days + zeolite at 30 g/kg soil. In addition, stem girth, root fresh weight and root dry weight were positively affected by irrigation every 5 days + zeolite at 30 g/kg soil. Leaf area improved when irrigated every 3 days and treated with zeolite at 30 or 60 g/kg soil in both seasons. Most of the recorded parameters showed a highly reduction when plants irrigated every 7 days without the application of zeolite.

Table 3: Effects of different irrigation intervals (D) and zeolite (Z) applications on several morphological traits of *Plectranthus scutellarioides* during the 2023 and 2024 seasons

Treatments	Plant height (cm)		Stem girth (cm)		No. of leaves		No. of branches		Leaf area (cm ²)	
	2023	2024	2023	2024	2023	2024	2023	2024	2023	2024
<i>D₃+Z₀</i>	78.83±3.49 ^{cd}	81.23±4.05 ^c	2.63±0.17 ^d	2.50±0.28 ^{de}	61.67±3.51 ^{cd}	59.67±4.72 ^{bc}	5.33±0.58 ^{abc}	5.00±1.00 ^{cd}	22.67±2.53 ^{bcd}	25.29±2.04 ^b
<i>D₃+Z₃₀</i>	82.67±2.63 ^{bc}	96.50±3.20 ^a	3.38±0.16 ^{ab}	3.68±0.28 ^{ab}	75.33±3.22 ^a	71.67±4.94 ^a	6.67±1.15 ^a	7.33±0.58 ^a	29.75±1.60 ^a	32.49±2.73 ^a
<i>D₃+Z₆₀</i>	90.27±4.35 ^a	84.67±2.88 ^{bc}	3.17±0.27 ^{bc}	2.86±0.36 ^{cd}	67.00±4.58 ^{bc}	53.00±3.61 ^{cd}	6.00±1.00 ^{ab}	6.00±0.00 ^{bc}	30.56±2.22 ^a	30.77±2.44 ^a
<i>D₅+Z₀</i>	75.33±3.70 ^{de}	70.00±3.50 ^{de}	2.83±0.33 ^{cd}	3.15±0.24 ^c	52.00±4.00 ^{ef}	43.67±4.94 ^{ef}	4.00±1.00 ^{cde}	4.33±0.58 ^{de}	21.33±2.66 ^{cd}	18.95±2.35 ^{cd}
<i>D₅+Z₃₀</i>	85.50±4.78 ^{ab}	90.33±4.02 ^b	3.63±0.36 ^a	3.89±0.18 ^a	71.67±4.51 ^{ab}	65.33±3.05 ^{ab}	6.00±1.00 ^{ab}	7.00±1.00 ^{ab}	24.69±2.31 ^{bc}	21.63±1.76 ^c
<i>D₅+Z₆₀</i>	72.23±4.31 ^{ef}	73.60±3.15 ^d	3.50±0.22 ^{ab}	3.30±0.31 ^{bc}	65.67±3.22 ^{bc}	50.33±4.51 ^{de}	4.67±0.58 ^{bcd}	5.67±0.58 ^c	25.78±3.28 ^b	26.08±2.51 ^b
<i>D₇+Z₀</i>	68.40±3.10 ^{fg}	59.17±2.66 ^g	2.84±0.26 ^d	2.13±0.23 ^e	37.67±2.88 ^g	34.33±4.72 ^g	2.67±0.57 ^c	3.00±0.00 ^f	14.56±1.35 ^c	13.34±1.20 ^e
<i>D₇+Z₃₀</i>	74.77±3.20 ^{de}	66.33±3.11 ^{ef}	2.80±0.28 ^{cd}	2.43±0.29 ^{de}	56.00±3.61 ^{de}	48.67±4.16 ^{de}	3.67±0.58 ^{de}	4.33±0.58 ^{de}	14.87±1.46 ^c	17.76±1.13 ^d
<i>D₇+Z₆₀</i>	62.67±2.97 ^g	61.80±3.70 ^{fg}	2.57±0.21 ^d	2.26±0.23 ^c	46.33±4.72 ^f	39.00±4.36 ^{fg}	3.33±0.58 ^{de}	3.67±0.58 ^{ef}	19.56±2.38 ^d	15.83±0.93 ^{de}

The means (± SD) were compared via Duncan's multiple range test (p< 0.05). The different letters in each column indicate a substantial effect, whereas values with the same letters do not.

Table 4: Effects of different irrigation intervals (D) and zeolite (Z) applications on several morphological traits of *Plectranthus scutellarioides* during the 2023 and 2024 seasons

Treatments	Root length (cm)		Shoot FW (g)		Shoot DW (g)		Root FW (g)		Root DW (g)	
	2023	2024	2023	2024	2023	2024	2023	2024	2023	2024
<i>D₃+Z₀</i>	23.33±2.80 ^b	22.70±2.66 ^c	96.27±3.49 ^c	101.58±4.34 ^d	10.16±0.84 ^c	10.72±0.98 ^d	7.23±0.44 ^g	10.82±0.66 ^f	1.81±0.06 ^{de}	2.80±0.23 ^{efg}
<i>D₃+Z₃₀</i>	31.83±3.01 ^a	34.67±2.29 ^a	120.63±5.19 ^a	127.00±3.60 ^a	14.23±0.85 ^a	14.99±0.69 ^a	12.83±0.58 ^d	13.61±0.69 ^{bcd}	3.32±0.31 ^{abc}	3.52±0.33 ^{bcd}
<i>D₃+Z₆₀</i>	22.33±2.16 ^b	21.00±2.38 ^{cd}	108.49±6.69 ^b	110.42±4.62 ^c	12.07±0.72 ^b	12.29±0.64 ^c	15.99±0.68 ^b	14.68±0.88 ^b	4.23±0.55 ^a	4.06±0.42 ^b
<i>D₅+Z₀</i>	24.77±2.37 ^b	28.17±3.12 ^b	84.66±5.26 ^d	86.35±3.67 ^c	8.07±0.70 ^d	8.23±0.64 ^{ef}	11.49±0.62 ^c	13.06±0.93 ^{cde}	2.94±0.45 ^{bc}	3.35±0.47 ^{cde}
<i>D₅+Z₃₀</i>	24.23±1.65 ^b	31.87±2.80 ^{ab}	111.85±5.35 ^b	118.65±3.11 ^b	12.85±0.79 ^b	13.63±0.63 ^b	14.73±0.73 ^c	14.05±0.77 ^{bc}	3.85±0.58 ^{ab}	3.82±0.31 ^{bc}
<i>D₅+Z₆₀</i>	18.20±1.99 ^{cd}	16.80±1.31 ^{de}	95.37±4.11 ^c	91.22±3.91 ^e	9.33±0.74 ^{cd}	8.92±0.77 ^c	18.30±0.91 ^a	19.72±0.74 ^a	4.23±0.54 ^a	5.32±0.37 ^a
<i>D₇+Z₀</i>	16.00±2.25 ^d	15.50±2.15 ^e	71.53±5.56 ^f	69.52±4.06 ^g	5.93±0.59 ^e	5.76±0.68 ^h	6.03±0.87 ^g	9.35±0.57 ^g	1.45±0.42 ^e	2.25±0.29 ^g
<i>D₇+Z₃₀</i>	21.67±2.46 ^{bc}	20.40±2.52 ^{cd}	80.94±3.63 ^{de}	78.15±4.79 ^f	6.46±0.81 ^e	6.20±0.64 ^{gh}	6.54±0.68 ^{gh}	11.76±0.93 ^{ef}	1.60±0.17 ^{de}	2.71±0.40 ^{fg}
<i>D₇+Z₆₀</i>	17.80±1.95 ^{cd}	16.00±2.72 ^e	76.21±3.64 ^{ef}	71.66±4.50 ^{fg}	6.59±0.58 ^e	7.11±0.65 ^{fg}	10.29±0.67 ^f	12.48±0.60 ^{de}	2.61±0.21 ^{cd}	3.10±0.37 ^{def}

The means (± SD) were compared via Duncan's multiple range test (p< 0.05). The different letters in each column indicate a substantial effect, whereas values with the same letters do not.

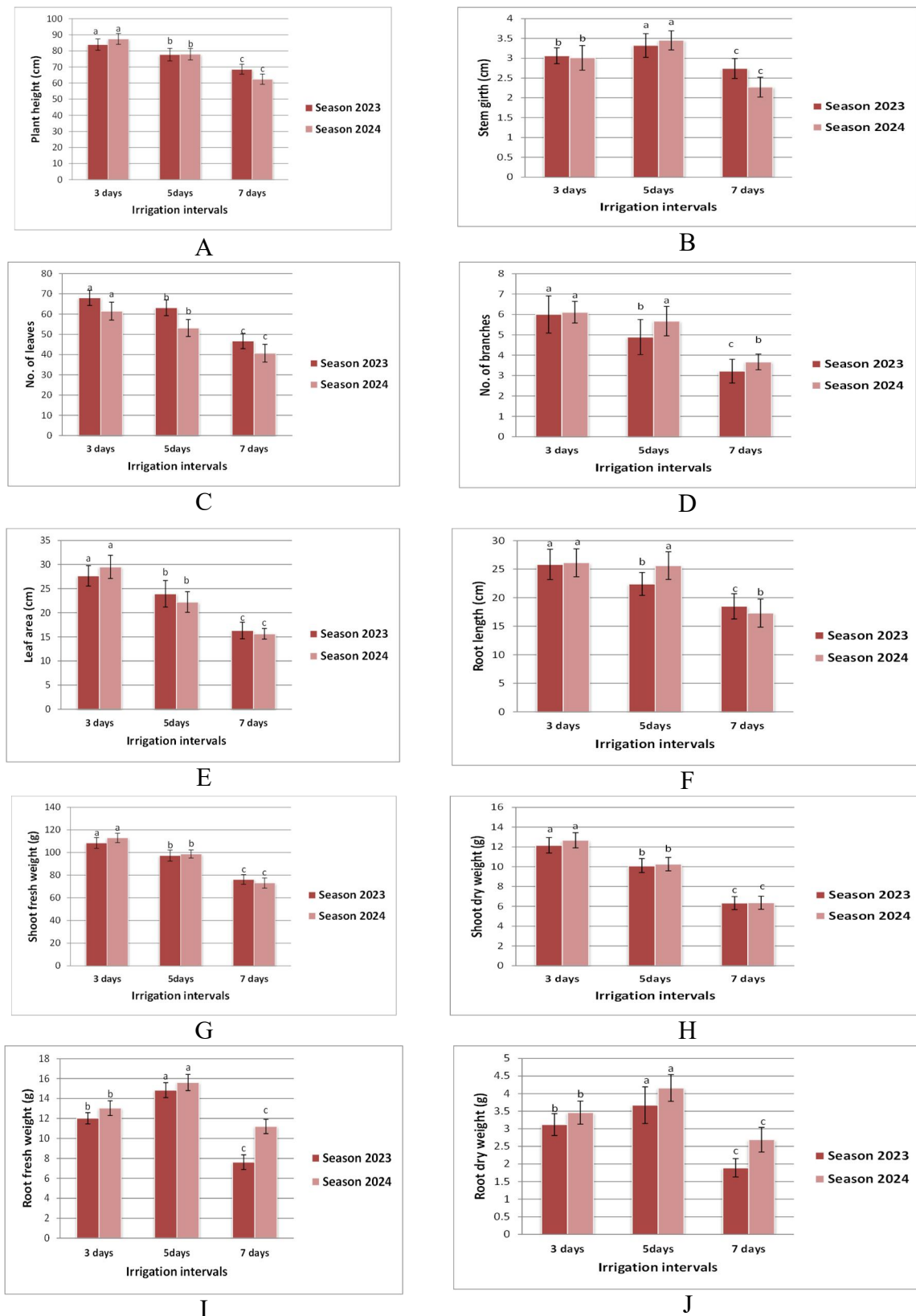


Fig. 1. Effects of different irrigation intervals (days) on the morphological traits of *Plectranthus scutellarioides* during the 2023 and 2024 seasons

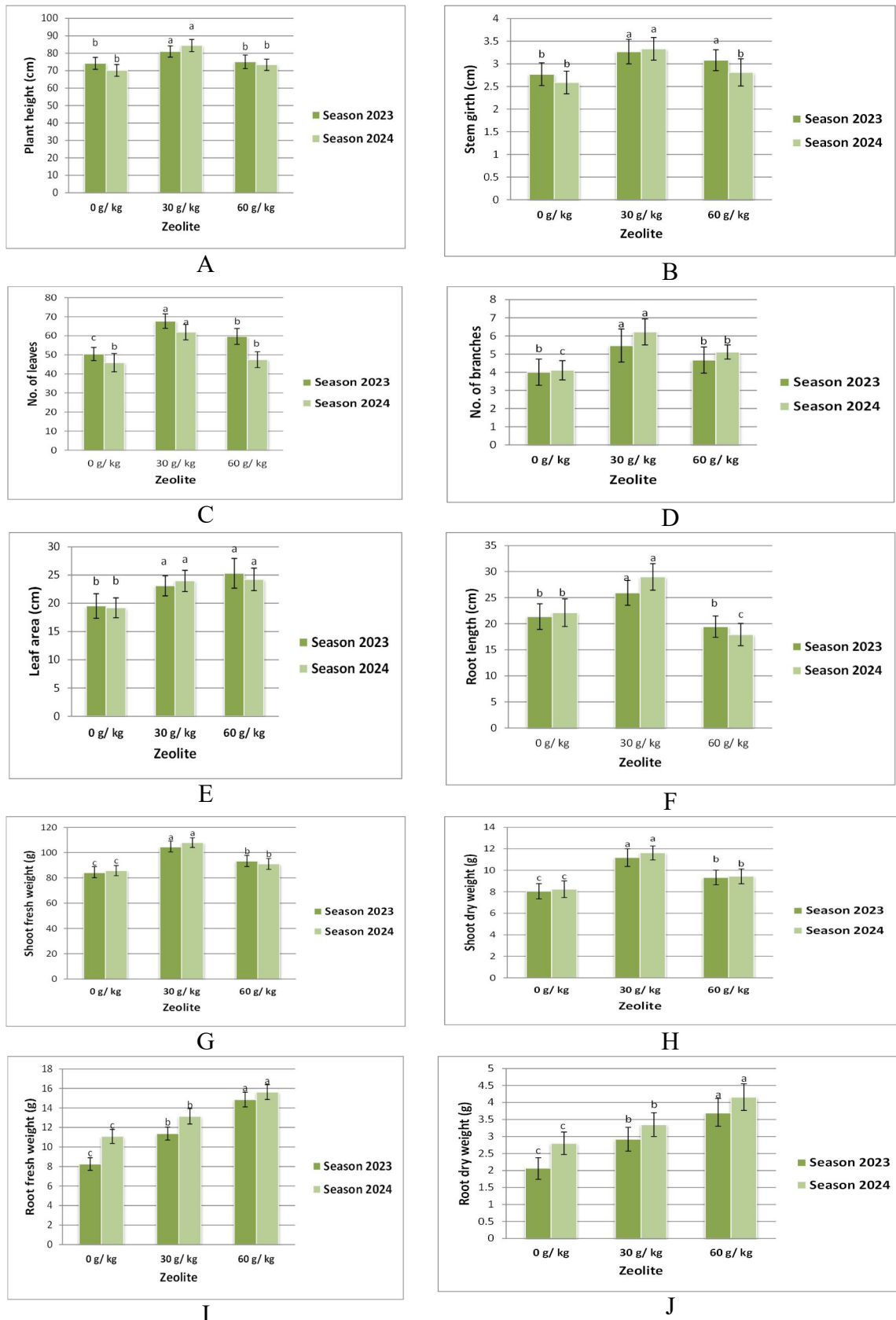


Fig. 2: Effects of zeolite application (g/kg soil) on the morphological traits of *Plectranthus scutellarioides* during the 2023 and 2024 season

3.2. Membrane stability index (MSI %) and relative water content (RWC %)

The data in Figure (3 A & B) showed that *P. scutellarioides* plants irrigated every 3 days presented the highest MSI% (64.25±4.06 and 62.72±4.17) and RWC% (90.68±3.66 and 86.80±4.15), respectively in both seasons. On the other hand, extending the dry period between irrigations to 7 days caused the highest decrement in both MSI% and RWC% as compared to other irrigation treatment.

The attached data in Figures (3 C & D) show that the application of zeolite at 30 g/kg soil significantly increased MSI%, with values 61.67±4.85 and 59.70±3.53; furthermore, RWC%, which significantly increased with the same treatment, with values of 89.50 ± 3.28 and 86.55±3.69, respectively, in 2023 and 2024 seasons.

The data in Fig. (3 E&F) illustrated that membrane stability index (MSI%) and relative water content (RWC%) were significantly affected by the interaction treatments. The irrigation every 3 days + zeolite 30g/ kg soil followed by irrigation every 3 days + zeolite 60g/ kg soil produced the highest MSI% without any significant differences between both treatments. RWC% significantly affected in plants irrigation every 3 days + zeolite 30 g/kg soil treatment followed by irrigation every 5 days + zeolite 30 g/kg soil, without any significant differences between both treatments. MSI% and RWC% negatively affected in plants irrigated every 7 days + zeolite 0 g/kg soil.

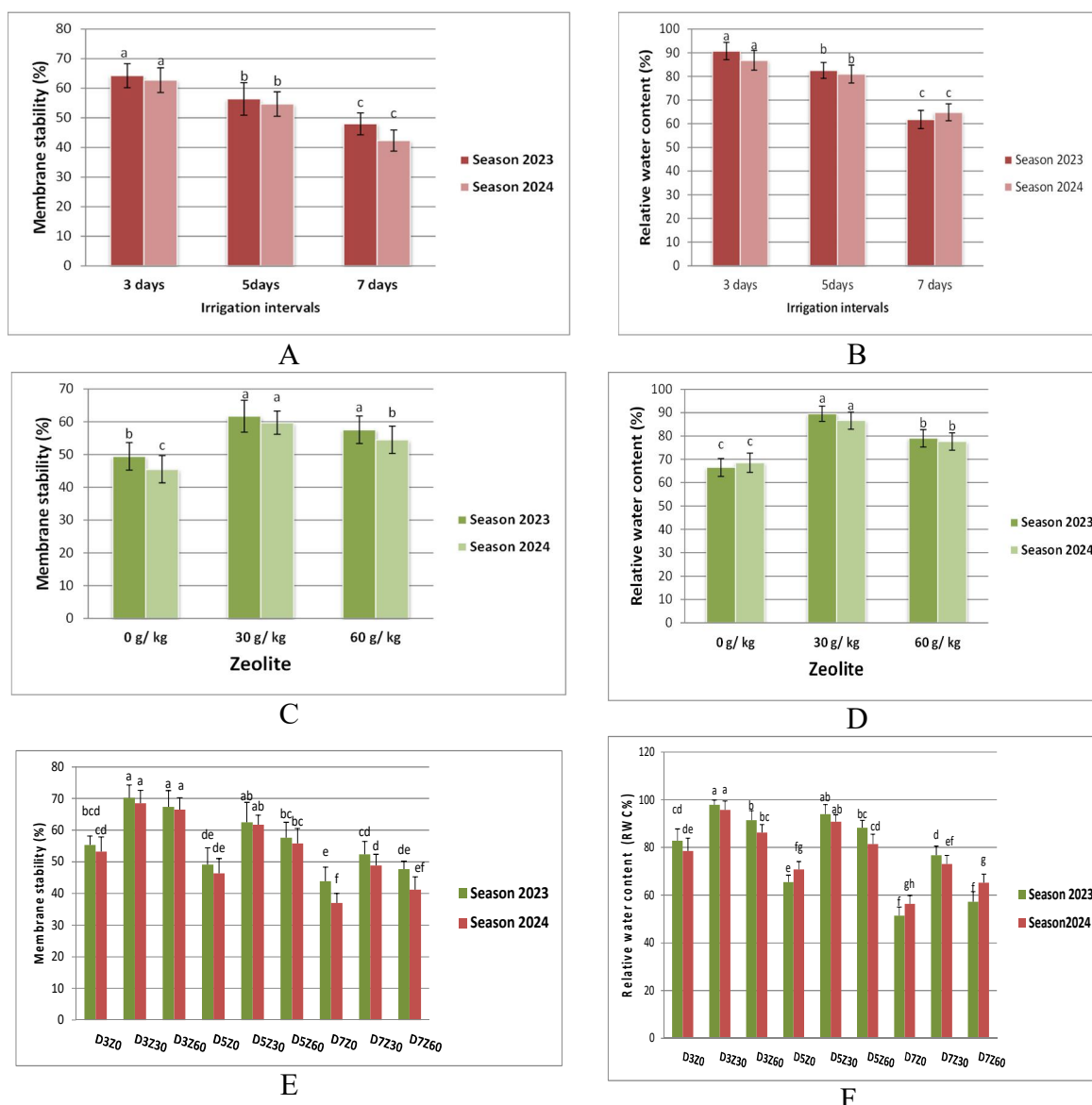


Fig. 3: Effects of different irrigation intervals, zeolite applications and their interactions on membrane stability% (A, C&E) and relative water content% (B, D&F).

3.3. Chemical constituents

3.3.1. Photosynthetic pigments (mg/g FW)

The data presented in Figures (4 A, B&C) indicate that irrigation every 5 days was the most effective irrigation treatment for promoting the synthesis and accumulation of the three photosynthetic pigments, in contrast to plants irrigated every 7 days which showed the highest decrement of photosynthetic pigments accumulation.

With respect to the effects of zeolites on photosynthetic pigments, as shown in Figures (5 A, B&C), the data revealed positive and active effects of zeolites at a level of 30 g/kg soil on photosynthetic pigments in *P. scutellarioides* leaves, followed by 60 g/kg soil during 2023 and 2024 seasons.

The data presented in Table (5) cleared that the irrigation every 5 days plus zeolite at 30 g/kg soil was the most effective for promoting the synthesis and accumulation of the photosynthetic pigments (chlorophyll a, b and carotenoid) during both seasons.

3.3.2. Total flavonoids (mg of RE/g FW), phenols (mg of GAE/g FW) and total antioxidants (%)

The recorded data in Figure (4 D, E & F) illustrate that the presence of total flavonoids, phenols and antioxidants activity increased with increasing irrigation interval in both seasons. Irrigation every 7 days increased the contents of total flavonoids (2.77 ± 0.28 and 2.86 ± 2.86 mg of RE/g FW), total phenols (0.78 ± 0.05 and 0.75 ± 0.073 mg of GAE/g FW) and total antioxidants (75.75 ± 3.83 and $74.85 \pm 4.40\%$) in 2023 and 2024 seasons, respectively.

In this context, the data in Figure (5 D, E & F) illustrate that the total flavonoid, phenol and antioxidant contents in plants treated with zeolite fluctuated with respect to the rate of application, where the total flavonoid content was affected by zeolite at 30 g/kg soil producing the highest values (2.79 ± 0.26 and 2.77 ± 0.25 mg of RE/g FW), whereas the highest values of phenols in plants without zeolite occurred (0.78 ± 0.06 and 0.75 ± 0.06 mg of GAE/g FW). Moreover, the highest activity of total antioxidants were produced by both zeolite treatment at 60 g/kg soil and control in the first and second seasons, respectively.

With respect to the interaction treatment, the plants were watered every 7 days and treated with 30 g/kg zeolite markedly increased the content of total flavonoids. However, the seedlings irrigated every 7 days without zeolite treatment provided the highest content of phenols and total antioxidants activity in the both seasons (Tables 5 & 6).

3.3.3. Total sugars content (mg/g FW)

As shown in Figure (4 G), plants irrigated every 5 days resulted in the highest total sugars content, which was 6.03 ± 0.36 and 6.13 ± 0.38 mg/g FW, respectively, in the first and second seasons in comparison with the other treatments.

With respect to the effect of zeolite on the total sugars content, the results revealed that zeolite at 30 g/kg soil produced the highest value of total sugars, which were 5.92 ± 0.34 and 6.13 ± 0.37 mg/g FW, respectively, in both seasons (Figure 5 G).

With respect to the effect of the interaction (Table 6), irrigating water every 5 day interval plus zeolite at 30 g/kg soil resulted in the highest total sugars content, with values of 6.64 ± 0.36 and 6.76 ± 0.40 mg/g F.W. in the two seasons, respectively. while, the lowest values were obtained when the seedlings were irrigated every 3-day interval, and zeolite at 60 g/kg soil presented values of 4.17 ± 0.30 and 3.72 ± 0.34 mg/g F.W. in both seasons, respectively.

3.3.4. Proline ($\mu\text{g/g FW}$) and lipid peroxidation (MDA $\mu\text{mol/g FW}$)

The results shown in Figure (4 H& I) indicate that *P. scutellarioides* plants irrigated every 7 days increased the production of proline giving values 302.30 ± 7.31 and $298.34 \pm 7.49 \mu\text{g/g FW}$, and the highest production of MDA content with values 2.39 ± 0.059 and $2.31 \pm 0.066 \mu\text{mol/g FW}$, respectively, in both seasons as compared with those of plants irrigated every 3 days, which presented the lowest values (262.04 ± 7.25 and $260.05 \pm 6.12 \mu\text{g/g FW}$) for proline and (1.84 ± 0.053 and $1.82 \pm 0.056 \mu\text{mol/g FW}$) for MDA in the first and second seasons, respectively.

The lowest content of proline (268.78 ± 7.13 and $260.63 \pm 6.87 \mu\text{g/g FW}$) and the lowest MDA content (2.01 ± 0.060 and $1.92 \pm 0.062 \mu\text{g/g FW}$) detected in the plants exposed to zeolite treatment at 30g/ kg soil, respectively, in the first and second seasons (Figure 5 H&I).

Table 5: Effects of different irrigation intervals (D) and zeolite (Z) application rates on photosynthetic pigments (chl. a, b and carotenoids), total flavonoids and total phenols of *Plectranthus scutellarioides* during the 2023 and 2024 seasons

Treatments	Chl. a (mg/g FW)		Chl. b (mg/g FW)		Carotenoids (mg/g FW)		Total flavonoids (mg of RE/g FW)		Total phenols (mg of GAE/-g FW)	
	2023	2024	2023	2024	2023	2024	2023	2024	2023	2024
<i>D₃+Z₀</i>	0.45±0.04def	0.42±0.07def	0.20±0.04e	0.20±0.04de	0.34±0.04cde	0.35±0.05de	1.78±0.22e	1.72±0.21e	0.66±0.05def	0.63±0.05cde
<i>D₃+Z₃₀</i>	0.56±0.04bc	0.60±0.05ab	0.28±0.03c	0.30±0.04ab	0.44±0.03ab	0.51±0.05ab	2.55±0.22bcd	2.41±0.17bcd	0.62±0.06efg	0.59±0.07de
<i>D₃+Z₆₀</i>	0.53±0.04bcd	0.49±0.04bcde	0.25±0.03d	0.22±0.03cde	0.40±0.05bc	0.40±0.07cd	2.22±0.25cd	2.06±0.34de	0.55±0.05 g	0.52±0.07e
<i>D₅+Z₀</i>	0.49±0.05cde	0.52±0.07bcd	0.21±0.03e	0.24±0.03bcd	0.37±0.04bcd	0.43±0.07bcd	2.13±0.22de	2.23±0.30cd	0.82±0.06ab	0.80±0.05a
<i>D₅+Z₃₀</i>	0.68±0.04a	0.65±0.08a	0.32±0.03a	0.33±0.04a	0.50±0.04a	0.53±0.05a	2.61±0.24bc	2.66±0.34bc	0.72±0.06cd	0.72±0.07abc
<i>D₅+Z₆₀</i>	0.61±0.05ab	0.57±0.06abc	0.30±0.03b	0.28±0.04abc	0.48±0.05a	0.48±0.03abc	2.36±0.25bcd	2.61±0.20bc	0.58±0.05 fg	0.56±0.05e
<i>D₇+Z₀</i>	0.38±0.05f	0.35±0.09f	0.15±0.02g	0.17±0.03e	0.25±0.04ef	0.23±0.04f	2.35±0.30bcd	2.49±0.23bcd	0.87±0.06a	0.83±0.08a
<i>D₇+Z₃₀</i>	0.43±0.04ef	0.46±0.06cdef	0.18±0.02f	0.20±0.04de	0.31±0.05def	0.37±0.04d	3.20±0.32a	3.24±0.25a	0.76±0.05bc	0.75±0.07ab
<i>D₇+Z₆₀</i>	0.40±0.07f	0.38±0.07ef	0.17±0.02f	0.18±0.03de	0.28±0.04f	0.27±0.04ef	2.76±0.23b	2.85±0.24ab	0.70±0.04cde	0.68±0.07bcd

The means ± standard deviations were compared via Duncan's multiple range test ($p < 0.05$). The different letters in each column indicate a substantial effect, whereas values with the same letters do not.

Table 6: Effects of different irrigation intervals (D) and zeolite (Z) application rates on the total antioxidant activity, total sugars content, proline content and lipid peroxidation of *Plectranthus scutellarioides* during the 2023 and 2024 seasons

Treatments	Total antioxidant activity (%)		Total sugars (mg/g FW)		Proline content (µg/g FW)		Lipid peroxidation (MDA µmol/g FW)	
	2023	2024	2023	2024	2023	2024	2023	2024
<i>D₃+Z₀</i>	46.31±4.02ef	43.56±5.40 fg	4.50±0.29ef	4.21±0.33ef	262.64±7.21c	268.53±5.97de	1.92±0.070f	1.93±0.046ef
<i>D₃+Z₃₀</i>	39.57±4.12f	36.12±4.66 g	5.28±0.28cd	5.36±0.34cd	259.63±6.87c	250.41±6.43f	1.76±0.043 g	1.69±0.053 g
<i>D₃+Z₆₀</i>	51.43±4.45de	50.71±4.86ef	4.17±0.30f	3.72±0.34f	263.84±7.66c	261.22±5.96ef	1.83±0.046 fg	1.85±0.070f
<i>D₅+Z₀</i>	68.96±4.20c	65.47±3.20bc	6.44±0.37a	6.41±0.37ab	287.01±6.71b	280.46±6.68c	2.27±0.062c	2.22±0.056b
<i>D₅+Z₃₀</i>	57.63±5.46d	55.84±4.89de	6.67±0.36a	6.79±0.40a	261.07±6.66c	255.38±6.39f	2.06±0.082e	1.96±0.061e
<i>D₅+Z₆₀</i>	67.48±4.75c	62.93±5.00cd	5.02±0.36de	5.19±0.37d	269.80±7.18c	271.85±7.73cde	2.14±0.044de	2.04±0.062cd
<i>D₇+Z₀</i>	79.65±3.45a	77.79±3.34a	5.63±0.26bc	5.89±0.29bc	328.47±8.17a	316.82±6.18a	2.56±0.061a	2.44±0.056a
<i>D₇+Z₃₀</i>	71.02±3.93bc	71.58±4.34ab	5.85±0.37b	6.23±0.36ab	285.63±7.87b	276.09±7.79cd	2.20±0.053cd	2.12±0.072bc
<i>D₇+Z₆₀</i>	76.58±4.12ab	74.37±5.51a	4.79±0.38de	4.53±0.22e	292.79±5.89b	302.11±8.49b	2.40±0.062b	2.36±0.069a

The means ± standard deviations were compared via Duncan's multiple range test (p< 0.05). The different letters in each column indicate a substantial effect, whereas values with the same letters do not.

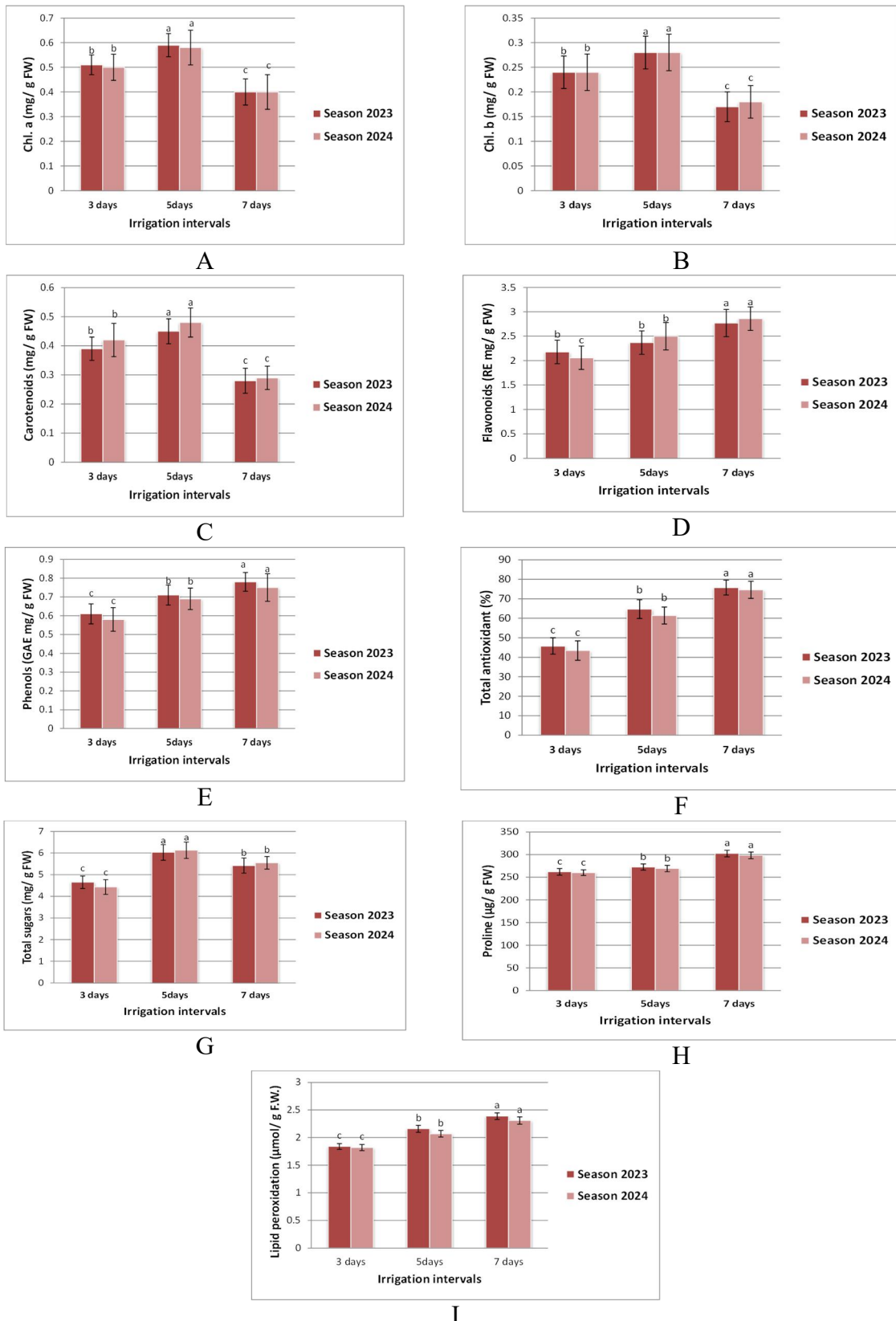


Fig. 4: Effects of different irrigation intervals (days) on the chemical constituents of *Plectranthus scutellarioides* during the 2023 and 2024 seasons

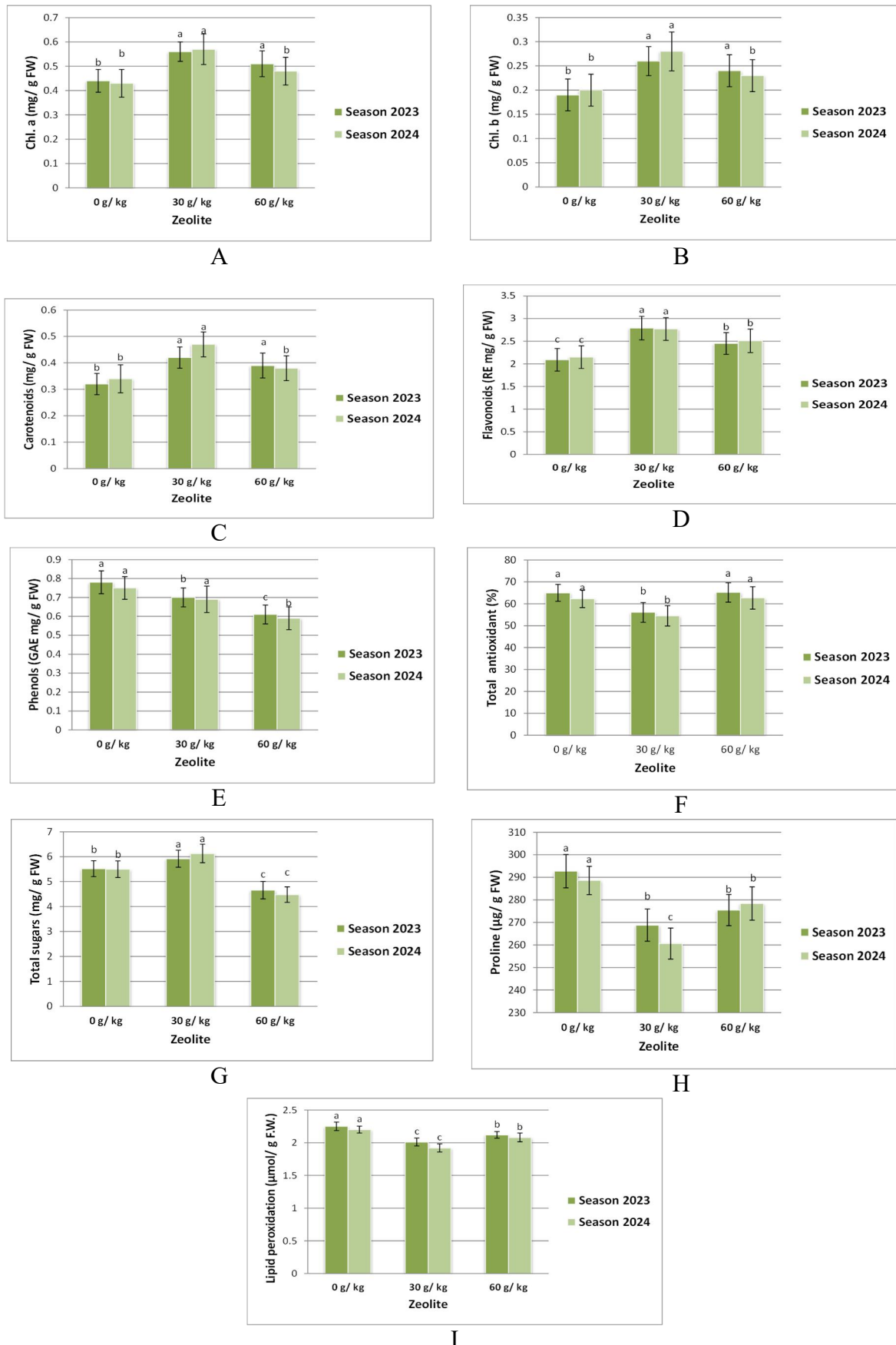


Fig. 5: Effect of zeolite application (g/ kg soil) on the chemical constituents of *Plectranthus scutellarioides* during the 2023 and 2024 seasons

In this context, the interaction between irrigation interval (3 days) and zeolite application at 30 g/kg soil revealed that the combination of both factors effectively decreased the production of proline and MDA. The lowest proline contents were 259.63 ± 6.87 and 250.41 ± 6.43 $\mu\text{g/g}$ FW, and the MDA contents were 1.76 ± 0.044 and 1.69 ± 0.053 $\mu\text{mol/g}$ FW in the first and second seasons, respectively. While irrigating every 7 days without any zeolite treatment caused signs of stress in the plants, namely an increase in the plant's proline and MDA content (Table 6).

3.2. Correlation analysis

The strength of the correlation coefficient between some morphological characteristics and the chemical content of *P. scutellarioides* plants during the 2023 and 2024 seasons, whether positive or negative, is expressed in Figure (6), where it was found that certain vegetative traits, such as plant height, leaf area, and photosynthetic pigments (carotenoids, chlorophyll a, and b), were strongly positively correlated with membrane stability and relative water content. On the other hand, plant height, leaf area, photosynthetic pigments, membrane stability, and relative water content were strongly inversely correlated with proline content, antioxidant activity, and lipid peroxidation. Root length was strongly positively correlated with plant height and relative water content, moderately positively correlated with leaf area and membrane stability, and weakly positively correlated with total sugars content. Moreover, root length was strongly inversely correlated with proline content and antioxidant activity, and it was not significantly inversely correlated with lipid peroxidation.

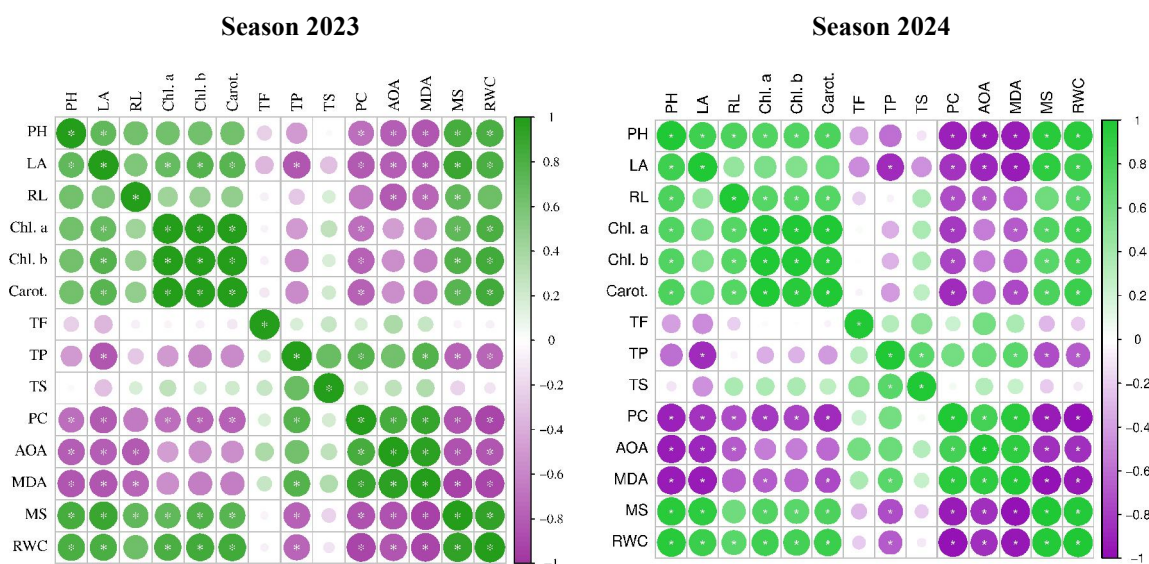


Fig. 6: The plot of Pearson's correlation coefficient analysis for some morphological parameters and chemical components of *P. scutellarioides* plants during the 2023 and 2024 seasons, where **PH:** plant height, **LA:** leaf area, **RL:** root length, **Chl. a:** chlorophyll a, **Chl. b:** Chlorophyll b, **Carot. :** carotenoids, **TF:** total flavonoids, **TP:** total phenols, **TS:** total sugars, **PC:** proline content, **AOA:** antioxidant activity, **MDA:** lipid peroxidation, **MS:** membrane stability, **RWC:** relative water content

4. Discussion

The findings of the present study demonstrated that the morphological traits of *Plectranthus scutellarioides* plants were negatively impacted by longer irrigation intervals. According to Farahani *et al.* (2025), the vegetative development properties of *Melissa officinalis* plants are adversely affected when they are exposed to low soil moisture contents. Khamis *et al.* (2025) compared the growth of C3 plants, represented by *Brassica oleracea*, and C4 plants, represented by *Echinochloa crusgallii*, under drought stress; the majority of the vegetative traits of both plants were shown to be adversely impacted by drought, despite variations in photosynthetic methods, whereas *B. oleracea* was more affected than *E. crusgallii*. Zhao *et al.* (2024) reported that *Begonia semperflorens* plants with acute water shortages

presented significantly lower morphological and ornamental values than did plants with normal irrigation. In another study, *Azadirachta indica* seedlings that were irrigated every eight days presented a substantial reduction in all the examined morphological parameters compared with those that were irrigated every four days (Mohamed *et al.*, 2023).

The ability of plants to adjust to variations in soil moisture content during drought by delaying development or altering their morphological traits is crucial for survival (Tian *et al.*, 2025). Kapoor *et al.* (2020) reported that growth is one of the physiological factors that is most sensitive to drought because of its cellular turgor, which in turn affects the cellular expansion, restricting the growth and development of plants. The reduction in growth might be due to a decrease in cell elongation caused by the inhibitory impact of water deficit on growth-stimulating hormones, which, in turn, leads to a reduction in cell turgor and volume and eventually growth and dry matter accumulation (Mahdi *et al.*, 2017; Coussement *et al.*, 2021). Water stress can restrict internode elongation and leaf expansion by inhibiting cell expansion (Namich and Emara, 2007). According to our research, irrigation every seven days decreased root growth, root length and fresh and dry weight measurements, which was consistent with the findings of El-Sayed *et al.* (2022) on *Swietenia mahogany*. Indole acetic acid (IAA), which is crucial for the division and elongation of root cells, may have decreased as a result of the exposure of the plant to dryness, according to Rauf and Sadaqat (2007). The results obtained in the present study are consistent with those of the study conducted by Elsayed *et al.* (2022) on *Eucalyptus citriodora* seedlings, which revealed that irrigating the plant every eight days resulted in a decrease in most of its morphological characteristics, with the exception of the values of root characteristics, which were related to development, contrary to the results of our current study, which increased when the irrigation period was extended (8 days). The root extension under water deficit conditions in line to the discussion put forth by Burman *et al.* (1991) that certain plants withstand drought stress by extending their roots and penetrating the lower soil layers in search of available moisture; this view has been supported by several studies (Hanafy, 2017).

The interactions between zeolite levels and watering intervals in the current study boost almost all growth metrics. The favorable effect of zeolite on plant growth may be because zeolite can improve soil characteristics and nutrient availability, promoting plant health (Aslan and Arslan, 2024). In a study on the coriander plant, Mahmoud *et al.* (2023) verified this finding, indicating that zeolite application enhanced the growth features under investigation; this may be related to increased photosynthetic rates, nutrient buildup, water use efficiency, and phytohormone concentrations of IAA and GA₃ and decreased concentrations of ABA. The increased absorption of nutrients and water, which are crucial for the biosynthesis of tryptophan, may be the cause of this outcome. This amino acid is thought to be essential for the production of IAA in plants (Zhang *et al.*, 2020). The increase in the GA₃ content in leaves is mostly associated with IAA upregulation in plants (Zhang *et al.*, 2021).

Owing to its numerous negative effects, drought stress is a complicated and catastrophic threat to plants. The osmotic imbalance that arises during drought disrupts several physiological and metabolic systems. The leaf relative water content, which is thought to be an important measure of a plant's water condition, reflects the balance between the amount of water the plant absorbs and its rate of transpiration (Prathyusha & Chaitanya, 2019). The RWC (%) of *P. scutellarioides* dramatically decreased during drought stress. Water distribution in plants is often determined by the metabolic capacity of plant tissues; consequently, plant tissues with high metabolic capacity may contain more water for survival (Sun *et al.*, 2020). Zhao *et al.* (2024) measured the relative water content in the leaves and stems of *B. semperflorens* plants under different drought conditions and reported that it decreased in both organs as the severity of drought increased. Our results also agree with those of Farahani *et al.* (2025); Luo *et al.* (2023); Tian *et al.* (2025); and Zhao *et al.* (2023) regarding the plants *Melissa officinalis*, *Chrysanthemum morifolium*, *Rhododendron delavayi*, and *Helleborus orientalis*. The results of this study revealed that zeolite improved the RWC under conditions of water deficiency stress, and the results were in accordance with those of Miranda-Rojas *et al.* (2025) for *Solanum lycopersicum* plants. The ability of zeolite to retain soil water, in turn, mitigates the negative impact of water stress on the soil, thus increasing the water retention capacity of plant tissue (González-Espíndola *et al.*, 2024).

Since the cell membrane is thought to be the first site of damage caused by continuous stress, a plant's ability to survive under environmental stress depends on its cell membrane integrity. Therefore, assessing the integrity of the cell membrane during drought stress is thought to be crucial for identifying tolerant plants (Prathyusha and Chaitanya, 2019). By measuring the MSI of leaf cells, the integrity of

the Coleus cell membrane during drought stress was investigated in this work. This study revealed that cell membranes were significantly damaged with increasing periods of drought. Water deficit stress is distinguished by the careless production of ROS due to limitations in stomatal gaseous exchange and electron diversion in electron transport chains and other energy-dissipating pathways. Increased ROS generation causes fast membrane degradation and leakage as a result of membrane lipid peroxidation (Guo *et al.*, 2018); moreover, destruction of the cell membrane system caused by drought stress induces membrane lipid peroxidation (MDA), damage to cytomembranes, and increased permeability (Liang *et al.*, 2023). This explains the parallel relationship between MSI% and MDA generation in drought-stressed plants in our current study. According to our research, zeolite treatment of *P. scutellarioides* plants decreased the negative impacts of dryness, which in turn decreased plant MDA production, which is consistent with the findings of Elawady (2024) and AbdEL-Azeiz and Elsonbaty (2024). This could be as a result of zeolite's enhanced soil qualities, which increase the capacity of the soil to hold water and decrease the sensitivity of the plant to drought stress (Shahsavari, 2019).

An insufficient water supply to plants leads to a decrease in their relative water content, as demonstrated in the current study. A reduced water content has an impact on photosynthesis, according to Lawlor & Cornic (2002). According to Meng *et al.* (2016), electron transport and PSII photochemical activity decrease with decreasing water content. Additionally, they discovered that the OEC (oxygen-evolving complex) content decreases as the duration of drought exposure increases, underscoring the harm that dryness does to photosynthetic system II. The reduction in chlorophyll content induced by water deficit may be attributed to the loss of chloroplast membranes, distortion of the lamellae and the appearance of lipid droplets (Fu and Huang, 2001). Chloroplasts also experience significant ultrastructural changes, such as shrinkage of the thylakoid membrane, disarray of granum stacks, and buildup of ROS (Wang *et al.*, 2025). These results are in accordance with those obtained by Elsayed *et al.* (2022) and Mohamed *et al.* (2023). In our study, the results revealed that, compared with untreated plants, the zeolite-treated plants presented a greater photosynthetic content under the same drought conditions, which is in line with the findings of Bahador and Tadayon (2020) on hemp plants. This may be because zeolite treatment clearly decreased leaf chlorophyll degradation and enhanced stomatal conductance, Fv/Fm, and the photosynthetic apparatus (Shahsavari, 2019). These results may be linked to the capacity of zeolite treatments to mitigate the detrimental effects of drought by increasing hydraulic conductivity, preserving higher rates of transpiration and photosynthesis and photosynthetic pigment concentrations, and lowering oxidative damage (Ghorbani *et al.*, 2022).

In addition to their fundamental role in reducing or inhibiting lipid oxidation, eliminating oxygen-free radicals, quenching singlet oxygen or decomposing peroxides, phenolic compounds are essential antioxidants that are responsible for protection against proliferation and advancement of the oxidation chain and defense against reactive oxygen species (Sakihama and Yamasaki, 2002). Phenolic compounds, which include many compounds, including flavonoids and total phenols, are naturally synthesized in the cell under optimal conditions, but when there is biotic stress, the concentration of these products is significantly affected (Madhvi *et al.*, 2020), possibly because a crucial enzyme in the phenolic pathway, phenylalanine ammonia-lyase (PAL), is frequently upregulated to promote phenolic production (Hossain *et al.*, 2024). Under abiotic stressors such as water deficit, PAL activity usually increases, resulting in the accumulation of phenolic compounds that play roles in membrane stability and ROS scavenging (Rao and Zheng, 2025). This explanation may explain why the total flavonoid and phenol contents and total antioxidant activity of the plants increased when the irrigation interval was extended to 7 days. This result is consistent with the results obtained by Ruttanaprasert *et al.* (2025) when *Helianthus tuberosus* plants were subjected to drought stress. In drought-stressed coleus plants, zeolite treatment increased the total phenolic and flavonoid contents as well as the total antioxidant activity, leading to increased stress resistance. This result is in line with the studies conducted by Mahmoud *et al.* (2023) on coriander plants and Othman *et al.* (2023) on *Solidago canadensis*.

To survive the current drought stress conditions, plants accumulate osmotic solutes through a variety of adaptations (Obidiegwu *et al.*, 2015). The buildup of osmolytes causes a net rise in the concentration of cellular solutes, which promotes hydration and preserves cell turgor. This increase may be because, during the course of drought stress, active solute accumulation of total sugars is claimed to be an effective stress tolerance mechanism (McKersie and Lesheim, 2013). Total sugars play dual roles in plants; they are involved in metabolic processes and act as molecular signals regulating various genes, particularly those involved in photosynthesis, sucrose metabolism and osmolyte synthesis (Rosa

et al., 2009). Moreover, proline plays an important biological role in the stress response (Qinglan *et al.*, 2023), and osmotic accumulation in plants results in a decrease in the osmotic potential of plants and therefore in the maintenance of water absorption and cell turgor pressure, facilitating physiological processes such as stomatal opening, photosynthesis, and growth under dry conditions (Pamungkas and Farid, 2022). Proline accumulation can influence stress tolerance in various ways. As a radical scavenger and effective osmoprotectant, proline shields the cellular machinery from damage caused by reactive oxygen (Zulfiqar and Ashraf, 2023; Iftikhar *et al.*, 2022). Since proline helps stabilize membrane-associated macromolecules and acts as an antioxidant in drought-affected cellular environments, its accumulation is recognized as a sign of stress tolerance (Rugienius *et al.*, 2020).

Previous research has demonstrated that under stressful conditions, several enzymes are markedly activated in an effort to maintain the intracellular osmotic pressure by controlling solute accumulation and proline synthesis and accumulation within cells (Sharma *et al.*, 2019). ABA increases the transcript levels of genes encoding important enzymes of proline biosynthesis pathways, such as pyrroline-5-carboxylate synthetase P5CS (Amini *et al.*, 2015). Additionally, abscisic acid plays a significant role in regulating sugar metabolism during water deficiency conditions, increasing sugar levels and increasing vacuolar invertase activity and the gene expression of invertase enzyme (IVR2) (Wu *et al.*, 2025). Proline builds up in the cytosol and shields membranes and enzymes from damage. Proline synthesis is associated with nitrogen assimilation with the help of ethylene (Iqbal *et al.*, 2015). With the aid of P5C synthetase and P5C reductase, plants primarily produce proline from P5C (Chalecka *et al.*, 2021). The results revealed a slight decrease in proline and sugar contents with zeolite treatment, which may be due to the improvements in the soil properties and water retention caused by zeolite, thus reducing plant exposure to drought and consequently decreasing proline production for this purpose.

4. Conclusion

Plectranthus scutellarioides (L.) showed a substantial decline in growth characteristics with increasing irrigation interval, according to the statistical analysis of the features examined. On the other hand, adding 30 g/kg zeolite to the soil improved the plant's capacity to grow normally in situations where water was scarce. Thus, we may conclude that the plants performed well in terms of growth when they were irrigated every three days with zeolite applied at a rate of 30 g/kg soil. However, when the watering interval was extended to five days in addition to the zeolite treatment at a rate of 30 g/kg soil, the plants attained balanced growth.

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Authors' contributions

El-Sayed SM participated in proposing the work plan, participated in the experiment's carry out and follow-up, collected samples, logged data, performed chemical analyses, participated in performing the statistical analysis of the data, prepared the figures, and wrote and reviewed the manuscript.

Abd Elhafez AE participated in proposing the work plan, participated in the experiment's carry out and follow-up, collected samples, logged data and participated in writing and reviewing the manuscript.

Mazhar AAM participated in the experiment's carry out and follow-up, performed chemical analyses, participated in performing the statistical analysis of the data, and wrote and reviewed the manuscript.

All authors read and approved the final manuscript.

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