



Morphological and physiological characteristics for evaluation of salicylic acid effects on *Celosia argentea* L. under salinity stress

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Abstract

Salinity stress is one of the most important abiotic stresses in agriculture, affecting about 20% of the total land cover. Salicylic acid (SA) plays an important role in plant tolerance to salinity stress and reducing its damages. In view of the importance of *Celosia argentea*, a factorial experiment was conducted based on a completely randomized design with three replications to investigate the mitigating role of SA on *C. argentea* plants under salinity. The factors included salinity in four levels (0, 40, 80, and 120 mM NaCl) and SA at three levels (0, 1, and 2 mM). Results showed that the effects of salinity stress on morphological and physiological characteristics were significant, in terms of a negative effect on growth and flowering of *C. argentea*. Foliar application of SA, especially at 1mM concentration, improved the morphological and physiological characteristics of the plants. Correlation between the measured traits showed that proline, malondialdehyde (MDA), and electrolyte leakage had a significant negative correlation with the other measured traits. As stress increased, stomatal conductance decreased, indicating closure of stomata during stress and plant resistance. In general, SA application increased photosynthesis rate through the improvement of plant pigments and proline contents under salinity stress. Therefore, because of its low costs and compatibility with the environment, SA is recommended as a simple solution to reduce salinity stress in this plant.

Keywords: chlorophyll, malondialdehyde, proline, photosynthesis rate, stomatal conductivity

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Introduction

Celosia argentea L. is a species of the Amaranthaceae family native to the warm regions of Asia and Africa. It is an annual flower with a height of 30-90 cm (Carter et al., 2005). This plant has flowers with different colors and therefore it

is used as a border flower and also as dried flowers due to its very high quality (Zuck, 2015). The flower has recently been reported to have the ability to adapt to, and grow in, soils with high copper (Wang et al., 2021). Studies have shown that several species in the Amaranthaceae family have secondary metabolites (Ali et al., 2021). The primary and secondary metabolites contents of the plant include carbohydrates, lipids, amino

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acids, peptides, phenols, phenolic acids, flavonoids, terpenes and alkaloids (Ali et al., 2021).

Salinity stress reduces plant growth in various ways, although the contribution of each of these ways is not well understood (Roozbahani et al., 2020; Yu et al., 2021). The effects of salinity stress on plants include a decrease in cell membrane stability, decrease in photosynthetic enzyme activity, reduction of photosynthesis, reduction of leaf development, decrease in ion uptake and especially the accumulation of sodium and chlorine ions in leaves, and finally decrease in vegetative growth and economic yield (Zhao et al., 2021; Yu et al., 2021). The reaction of plant species to soil or water salinity depends on their ability to eliminate the toxic effects of salinity (Hosseini et al., 2017; Salachna and Piechocki, 2021).

Salicylic acid (SA) or orthohydroxybenzoic acid, with the chemical formula $C_7H_6O_3$, is a plant phenol that plays an important role in physiological processes such as plant growth and development, photosynthesis, transpiration, and ion uptake (La et al., 2019; Safari et al., 2021). It is also known as an important signal molecule in plant responses to environmental stresses (Safari et al., 2021). Under conditions of severe stress, plant self-regulation is disrupted and the inefficiency of the antioxidant system causes an adverse effect on plant physiology and oxidative damage. Thus, the application of exogenous SA as a cellular messenger molecule can play an important role in inducing tolerance to biological and abiotic stresses (La et al., 2019).

SA affects many quantitative and qualitative characteristics of the plants under salinity stress conditions. Research has shown that SA has a positive effect on a wide range of characteristics such as seed germination, growth, yield, yield components, especially in saline conditions, but there is no general consensus about the optimal concentration for the use of SA (Jayakannan et al., 2015; Rasheed et al., 2020).

Considering the increasing salinity levels of soils, and the assessment of a solution to address this stress, the present study was conducted to investigate the effect of SA foliar

application on reducing the salinity effects of *C. argentea*.

Materials and Methods

Plant material and growth conditions

The experiment was done at the greenhouse of the Faculty of Agriculture, Lorestan University (Khorramabad, Iran; latitude 33° 26' N, longitude 48° 15' E) in a pot experiment. Seeds were planted in pots with 20 cm in diameter and 18 cm height containing a mixture of sand, soil, and composted manure by a ratio of 1:1:1. Seed germination was observed after seven days. Treatments were applied after the plants reached the four-leaf stage. The present study was carried out as a factorial experiment based on a completely randomized design with three replications. The factors included salinity in four levels (0, 40, 80, and 120 mM NaCl) and SA at three levels (0, 1, and 2 mM). The pots were placed in the greenhouse conditions ($450 \mu\text{mol m}^{-2} \text{s}^{-1}$ light intensity, 15 to 23 °C during the day and night and 70% relative humidity). The soil moisture of the pots was kept within the 90% field capacity. Salinity treatments were applied gradually to avoid sudden shock. Foliar application of SA was started one week before the onset of salinity stress and continued at intervals of seven days until the end of the experiment.

Morphological traits and growth parameters

Stem diameter, flower diameter, inflorescence length, number of leaves, flowers, and branches, as well as stem fresh weight, stem dry weight, flower fresh weight, flower dry weight, leaf fresh weight, leaf dry weight, root fresh weight, and root dry weight were measured at the flowering stage. The lengths of organs were measured using calipers or a ruler. After measuring the fresh weight of each plant tissue, the samples were oven-dried at 80 °C for 48 hours, and their dry weights were measured.

Proline measurement

Proline content was measured based on Bates method (Bates et al., 1973). Briefly, 10 ml of 3% sulfosalicylic acid was added to 0.2 g of powdered fresh leaf sample and centrifuged at 1000 rpm for

15 min. Next, 2 ml of the supernatant and 2 ml of ninhydrin and 2 ml of glacial acetic acid were added to a new falcon and heated at 100 °C in a water bath for 1 hour. Then, the falcons containing the extract were placed in ice rapidly (to stop the reactions) for 1 to 2 minutes, and 4 ml of toluene was added to the tubes and mixed with a vortex. The upper phase was separated and the amount of light absorption was measured at 520 nm with a spectrophotometer. Using the standard curve, the amount of proline was calculated in $\mu\text{mol g}^{-1}$ FW using the equation below.

Proline ($\mu\text{mol g}^{-1}$ FW) =

$$[(\mu\text{g proline/ml}) \times \text{ml toluene}] / 115.5 \mu\text{g}/\mu\text{mol} / [(\text{g sample})/5]$$

Measurement of malondialdehyde (MDA)

Membrane lipids peroxidation based on malondialdehyde (MDA) produced by membrane damage and its reaction with thiobarbituric acid, which forms the dye of thiobarbituric acid-malondialdehyde, was measured according to Buege and Aust (1978). Based on this method, 0.5 g of powdered fresh leaf tissue was added to a micro-tube containing 5 ml of 20% trichloroacetic acid solution (containing 0.5% thiobarbituric acid) and centrifuged for 15 minutes at 6000 rpm. The supernatant was placed in a hot water bath at 80 °C for 25 minutes and centrifuged for 5 minutes at 6000 rpm. The adsorption of the samples was observed at a wavelength of 600 and 532 nm. Finally, the amount of MDA was calculated through the following equation:

$$\text{MDA } (\mu\text{mol g}^{-1} \text{FW}) = [(A_{532} - A_{600}) / 155.5] \times 1000$$

Chlorophyll and carotenoid content

Lichtenthaler (1987) method was used to measure chlorophyll and carotenoids. In this method, 0.1 g of the powdered leaf sample was added to 10 ml of pure acetone and centrifuged for 10 minutes at 4000 rpm. Using a spectrophotometer, the absorbance was read at 470, 645, and 662. The amounts of chlorophyll a, chlorophyll b, and carotenoids were calculated through the following equations:

$$\text{Chl a } (\text{mg g}^{-1}\text{FW}) = 11.24 \times A_{662} - 2.04 \times A_{645}$$

$$\text{Chl b } (\text{mg g}^{-1}\text{FW}) = 20.13 \times A_{645} - 2.04 \times A_{662}$$

$$\text{Carotenoids } (\text{mg g}^{-1}\text{FW}) = [(1000 \times A_{470}) - (1.90 \times \text{Chl a}) - (63.14 \times \text{Chl b})] / 214$$

Electrolyte leakage measurement

The membrane stability index was evaluated by measuring the electrolytes leakage of leaf. According to the method of Huo et al. (1996), fresh leaf samples were washed three times with distilled water to remove contaminants. Leaf specimens were cut to a one cm^2 pieces and transferred to closed glass tubes containing distilled water with a volume of 10 ml and placed at room temperature (25 °C) for 2 hours. Then, the electrical conductivity (EC_1) of the distilled water along with the sample was measured as a primary leak. After autoclaving the samples at 120 °C for 20 minutes and cooling, secondary electrical conductivity (EC_2) was measured. The percentage of electrical conductivity was calculated through the following equation:

$$\text{Electrical conductivity } (\%) = (\text{EC}_1 / \text{EC}_2) \times 100$$

Determination of relative water content

To determine plant water status, relative water content (RWC) was measured according to the method of Yamasaki and Dillenburg (1999):

$$\text{RWC } (\%) = (\text{FW} - \text{DW}) / (\text{TW} - \text{DW}) \times 100$$

where FW is the fresh weight, DW is the dry weight, and TW is the turgid weight.

Leaf gas exchange traits

Net photosynthetic rate (Pn), transpiration rate (Tr), and stomatal conductance (gs) measurements were performed on five attached leaves. Measurements were taken by using a portable photosynthesis system (CI-340; CID, Inc., Camas, WA, USA). Leaf chamber (6.25 cm^2) conditions were 34 ± 1 °C air temperature, 55 ± 2 % relative air humidity, and an incoming air CO_2 concentration of $376 \pm 2 \mu\text{mol mol}^{-1}$. Light intensity was set at $300 \mu\text{mol m}^{-2} \text{ s}^{-1}$.

Statistical Analysis

Table 1

Analysis of variance (mean squares) of the effects of salinity and SA treatments on morphological, physiological, and biochemical properties of *Celosia argentea*

Source of variations	Plant height	Stem diameter	Flower diameter	Inflorescence length	Number of leaves	Number of flowers	Number of branches	Leaf fresh weight	Stem fresh weight
Salinity	72.14 **	7.17 **	61.1 **	738.55 **	4285.4 **	158.85**	5.29 ^{ns}	67.45 **	119.6 **
SA	9.84 **	2.00 *	6.86 **	70.03 **	207.86 **	16.69 *	4.78 ^{ns}	2.77 *	5.10 **
Salinity * SA	0.20 ^{ns}	0.16 ^{ns}	0.50 ^{ns}	0.21 ^{ns}	10.27 ^{ns}	1.44 ^{ns}	1.70 ^{ns}	1.52 ^{ns}	0.68 ^{ns}
Error	0.88	0.43	0.72	6.31	24.86	3.33	3.75	0.69	0.43

Source of variations	Flower fresh weight	Root fresh weight	Total fresh weight	Leaf dry weight	Stem dry weight	Root dry weight	Flower dry weight	Total dry weight	Proline
Salinity	30.66 **	65.69 **	1049.5**	5.91 **	7.47 **	7.15 **	4.43 **	97.48 **	0.443 **
SA	0.93 *	4.12 **	40.47 **	0.37 **	0.32 **	0.45 **	0.13 *	4.59 **	0.188 **
Salinity * SA	1.23 **	0.49 ^{ns}	11.43 **	0.04 ^{ns}	0.04 ^{ns}	0.05 ^{ns}	0.18 **	0.91 *	0.023 **
Error	0.27	0.34	2.52	0.03	0.03	0.04	0.04	0.30	0.0007

Source of variations	MDA	Chl a	Chl b	Carotenoid	Electrolyte leakage	RWC	Tr	Pn	gs
Salinity	0.808 **	5.34 **	0.942 **	0.854 **	442.95 **	758.9 **	11.81 **	13.86 **	0.1076 **
SA	0.212 **	0.495 **	0.087 **	0.079 **	31.47 **	75.76 **	0.503 **	2.835 **	0.0146 **
Salinity * SA	0.041 **	0.133 **	0.023 **	0.0213 **	5.94 **	21.82 **	0.11 **	0.142 ^{ns}	0.0032 **
Error	0.007	0.0201	0.003	0.0032	0.996	4.28	0.024	0.177	0.0034

Ns: not significant; *: significant at 0.05 probability level; **: significant at 0.01 probability level

All the data regarding morpho-physiological and biochemical attributes were analyzed by analysis of variance (ANOVA). Means were separated by LSD ($P \leq 0.05$) (Sokal and Rohlf, 1997) using Minitab software version 20.

Results

Plant height, stem diameter, flower diameter, and inflorescence length

Analysis of variance showed that salinity stress and foliar application of SA had a significant effect ($p < 0.01$) on plant height, stem diameter, flower diameter, and inflorescence length (Table 1). Mean comparison showed plant height (18.78–25.3 cm), stem diameter (6.64–7.71 cm), as well as flower diameter (22.59 –28.67 cm), and inflorescence length (57–77.98 cm) varied in control plants and plants under salinity (120 mM) stress. (Table 2). Application of SA was associated with increased plant height (21.46–23.17 cm), stem diameter (7.26–8.01 cm), flower diameter (25.23 –26.68 cm), and inflorescence length (67.42–70.83 cm) as compared to non-spray

plants (Table 2). Mean comparison of interaction effect showed the maximum plant height, stem diameter, flower diameter, and inflorescence length observed in control plants under foliar application of SA.

Number of leaves, flowers, and branches

Analysis of variance showed that salinity stress and foliar application of SA had a significant effect ($p < 0.01$) on the number of leaves and flowers, but they had no significant effect on the number of branches (Table 1). Mean comparison showed that the number of leaves (72.11–121 cm) and the number of flowers (11.78–21.56 cm) varied in control plants and the plants under salinity (120 mM) stress (Table 2). Application of SA was associated with an increased number of leaves (98.5–103.58 cm) and flowers (15.75–17.75 cm) as compared to control plants (Table 2). Mean comparison of the interaction effects showed the highest number of leaves and flowers were observed in the control plants under foliar application of 1mM SA.

Fresh and dry weights of leaf, stem, flower, root, and total

SA could improve them under salinity stress (Table 2). One (1) mM SA showed a pronounced effect on

Table 2
Mean comparison of the effects of salinity and SA treatments on morphological, physiological, and biochemical properties of *Celosia argentea*

Treatments	Plant height (cm)	Stem diameter (mm)	Flower diameter (mm)	Inflorescence length (mm)	Number of leaves	Number of flowers	Number of branches	Leaf fresh weight (g)	Stem fresh weight (g)
Salinity (NaCl)									
Control	25.30 a*	8.71 a	28.67 a	77.89 a	121 a	21.56 a	14.67 a	14.35 a	13.29 a
40 mM	23.38 b	7.72 b	26.94 b	72.44 b	111.89 b	17.67 b	15.11 a	13.75 a	11.26 b
80 mM	21.03 c	7.12 bc	25.11 c	65.22 c	91.56 c	14.56 c	14.11 a	10.83 b	7.402 c
120 mM	18.78 d	6.64 c	22.59 d	57.00 d	72.11 d	11.78 d	13.33 a	8.44 c	5.242 d
SA									
control	21.64 b	7.26 b	25.23 b	67.42 b	98.5 b	15.75 b	13.583 a	11.33 b	9.192 b
1 mM	23.17 a	8.01 a	26.68 a	70.83 a	103.58 a	17.75 a	14.583 a	12.27 a	9.999 a
2 mM	21.56 b	7.36 b	25.57 b	66.17 b	95.333 b	15.67 b	14.75 a	11.93 ab	8.709 b

Treatments	Flower fresh weight (g)	Root fresh weight (g)	Total plant fresh weight (g)	Leaf dry weight (g)	Stem dry weight (g)	Root dry weight (g)	Flower dry weight (g)	Total plant dry weight (g)
Salinity (NaCl)								
Control	10.28 a	10.94	48.86 a	3.28	3.32 a	3.61 a	3.91 a	14.12 a
40 mM	8.53 b	8.35	41.90 b	2.51	2.82 b	2.76 b	3.24 b	11.32 b
80 mM	6.65 c	6.60	31.49 c	1.98	1.85 c	2.18 c	2.53 c	8.54 c
120 mM	6.29 c	4.57	24.54 d	1.37	1.31 d	1.51 d	2.39 cd	6.58 d
SA								
control	7.79 b	7.17 b	35.47 b	2.15 b	2.30 b	2.37 a	2.96 b	9.77 b
1 mM	8.26 a	8.28 a	38.81 a	2.48 a	2.50 a	2.73 b	3.14 a	10.85 a
2 mM	7.76 b	7.40 b	35.81 b	2.22 b	2.18 b	2.44 b	2.95 b	9.79 b
Salinity * SA								
Control control	10.56 a	10.92 a	49.89 a	3.28 a	3.45 a	3.60 a	4.01 a	14.34 a
Control 1 mM	9.56 b	11.07 a	47.79 ab	3.32 a	3.39 ab	3.65 a	3.63 b	13.99 a
Control 2 mM	10.72 a	10.82 a	48.90 a	3.25 a	3.13 bc	3.57 a	4.07 a	14.02 a
40 mM control	8.51 cd	7.63 c	40.33 c	2.29 c	2.77 de	1.25 g	3.23 cd	10.81 c
40 mM 1 mM	9.26 bc	9.40 b	45.20 b	2.82 b	3.03 cd	1.75 ef	3.52 bc	12.47 b
40 mM 2 mM	7.83 de	8.03 c	40.16 c	2.41 c	2.66 e	1.52 fg	2.97 de	10.69 c
80 mM control	6.22 gh	6.32 d	29.70 e	1.90 d	1.84 fg	2.52 c	2.36 gh	8.18 e
80 mM 1 mM	7.30 ef	7.36 c	34.67 d	2.21 c	2.06 f	3.10 b	2.78 ef	9.47 d
80 mM 2 mM	6.42 gh	6.13 de	30.10 e	1.84 de	1.65 gh	2.65 c	2.44 gh	7.95 e
120 mM control	5.87 h	3.80 g	21.98 f	1.14 g	1.13 j	2.08 d	2.23 h	5.75 f
120 mM 1 mM	6.90 fg	5.29 ef	27.58 e	1.59 ef	1.52 hi	2.43 c	2.62 fg	7.48 e
120 mM 2 mM	6.09 gh	4.61 fg	24.07 f	1.38 fg	1.28 ij	2.02 de	2.31 gh	6.50 f

*Comparison of the means was done using the LSD test at $p < 0.05$. Means with the same letter are not significantly different.

Analysis of variance showed that salinity stress and foliar application of SA had a significant effect ($p < 0.01$) on the fresh and dry weights of leaves, stems, flowers, roots and total (Table 1). Mean comparison showed all these traits varied in control plants and plants under salinity stress (Table 2). Besides, the traits decreased with increasing salinity stress, and foliar application of

the biomass traits of the flowers under salinity.

Proline

Analysis of variance showed that salinity stress, foliar application of SA, and interaction had a significant effect ($p < 0.01$) on proline content (Table 1). Mean comparison showed proline

content varied in control plants and the plants under salinity stress (Fig. 1). Application of SA was associated with increased proline content as compared to control plants (Figs. I & II). Mean comparison of interaction effect showed the trend of proline content increased rapidly with increasing salinity. However, foliar application of SA was also effective in increasing the proline level (Fig. 1).

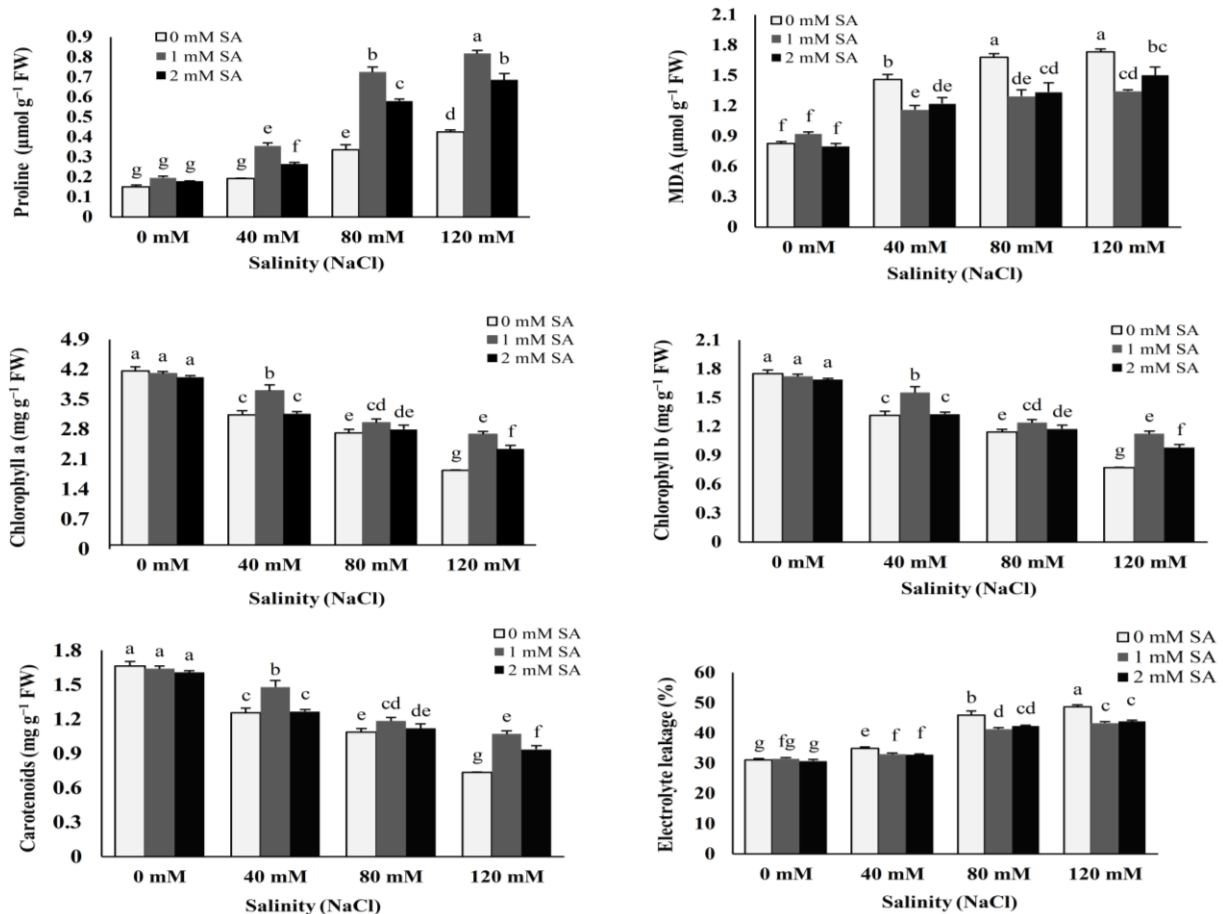


Fig 1. Proline, MDA, electrolyte leakage, chlorophyll a and b, and carotenoids in *Celosia argentea* under salinity stress and foliar applications of salicylic acid; data represent means \pm standard error (SE). Comparison of the means was done using the LSD test at $p < 0.05$. Means with the same letter are not significantly different.

MDA and electrolyte leakage

Analysis of variance showed that salinity stress, foliar application of SA, and their interaction had a significant effect ($p < 0.01$) on MDA content and electrolyte leakage (Table 1). Mean comparison showed MDA content and electrolyte leakage increased with increasing salinity (Fig. 1). Application of SA was associated with increased proline content as compared to the control plants (Fig. 1). Mean comparison of the interaction effect

showed the maximum MDA content and electrolyte leakage observed in 120 mM treatment without SA application (Fig. 1)

Plant pigments

Analysis of variance showed that salinity stress, foliar application of SA, and interaction had a significant effect ($p < 0.01$) on chlorophyll a, b, and carotenoid contents of the plants under study

(Table 1). Mean comparison showed chlorophyll a, b, and carotenoids decreased with increasing salinity (Fig. 1). Application of SA was associated with increased chlorophyll a, b, and carotenoids as compared to the control plants (Fig. 1). Mean comparison of the interaction effect showed the highest chlorophyll a, b, and carotenoid contents observed in control plants without foliar application of SA. However, in the absence of salinity stress, the application or non-application of SA did not show significant differences.

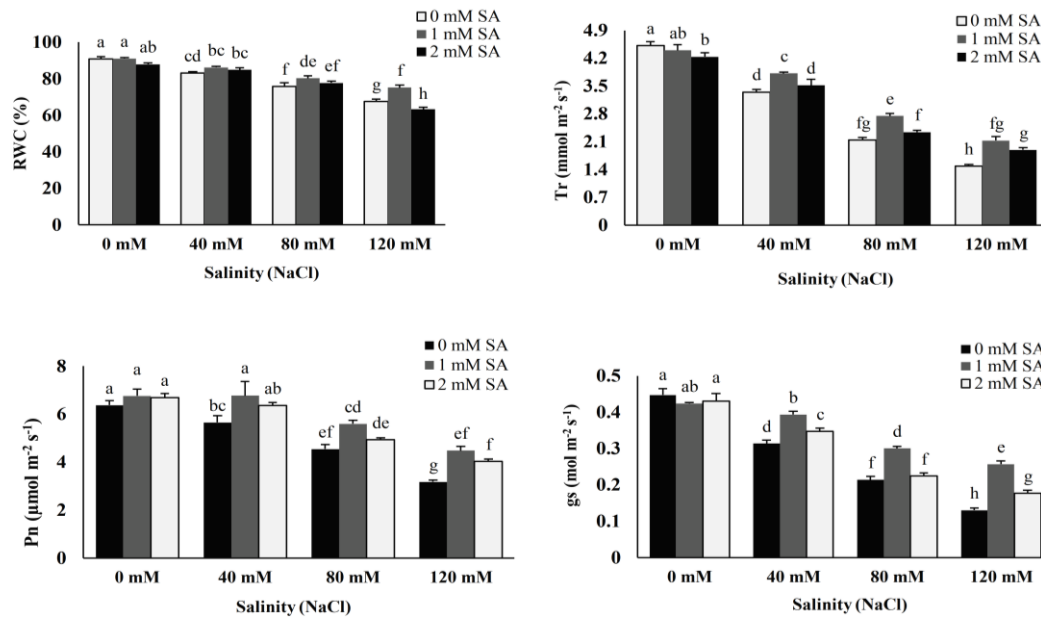


Fig. II. RWC, Tr, Pn, and gs in *Celosia argentea* under salinity stress and foliar applications of salicylic acid; data represent means \pm standard error (SE). Comparison of the means was done using the LSD test at $p < 0.05$. Means with the same letter are not significantly different.

Leaf gas exchange traits and hydration status

Salinity stress, foliar application of SA, and their interaction had a significant effect ($p < 0.01$) on leaf gas exchange traits (Pn, gs, and Tr) and hydration status (RWC) (Table 1). Foliar application of SA led to an increase in Pn, gs, Tr, and RWC as compared to controls (Fig. II). Foliar application of SA stimulated a higher increase in these traits especially in 1mM concentration as compared to control (Fig. II). Mean comparison of the interaction effect showed that foliar application of SA had no significant effect on salinity control treatment (0 mM salinity), although SA spraying was effective in increasing these traits by applying salinity stress (Fig. II).

In breeding programs, selection for one trait may affect other traits, so it is important to study the correlations between traits. Phenotypic correlation coefficient is a measure of the relationship between traits and can be used as an important selection indicator (Khorshid et al., 2003). Correlation between the traits (Fig. III) showed that proline, MDA, and electrolyte leakage had a significant negative correlation with other measured traits. The morph-physiological

and biochemical traits in this study had a significant positive correlation with each other except for the number of branches which had a low non-significant correlation with all measured trait (Fig. III).

Discussion

The adverse effects of salinity stress on growth and physiological characteristics of *C. argentea* were addressed together with the alleviating role of SA application. In this study, foliar application of SA (0, 1, and 2 mM) was used to reduce the harmful effects of salinity stress (0, 40, 80, and 120 mM NaCl). Morpho-physiological traits, stomatal conductance (gs), and photosynthesis were measured to evaluate the plant response to the studied treatments.

Morphological characteristics of *C. argentea* showed that plant height, stem diameter, inflorescence length, and also the fresh and dry weights of leaves, stems, flowers, roots, and total were significantly affected by salinity, and decreased under saline condition. Analysis of the data on the effect of SA on morphological characteristics of *C. argentea* showed that all

measured characteristics were significantly affected by foliar application of SA and improved under salinity stress.

stresses in plants (Rasheed et al., 2020). It also interacts with plant hormones under different conditions. This compound inhibits the reduction of auxin and cytokinin hormones under stress.

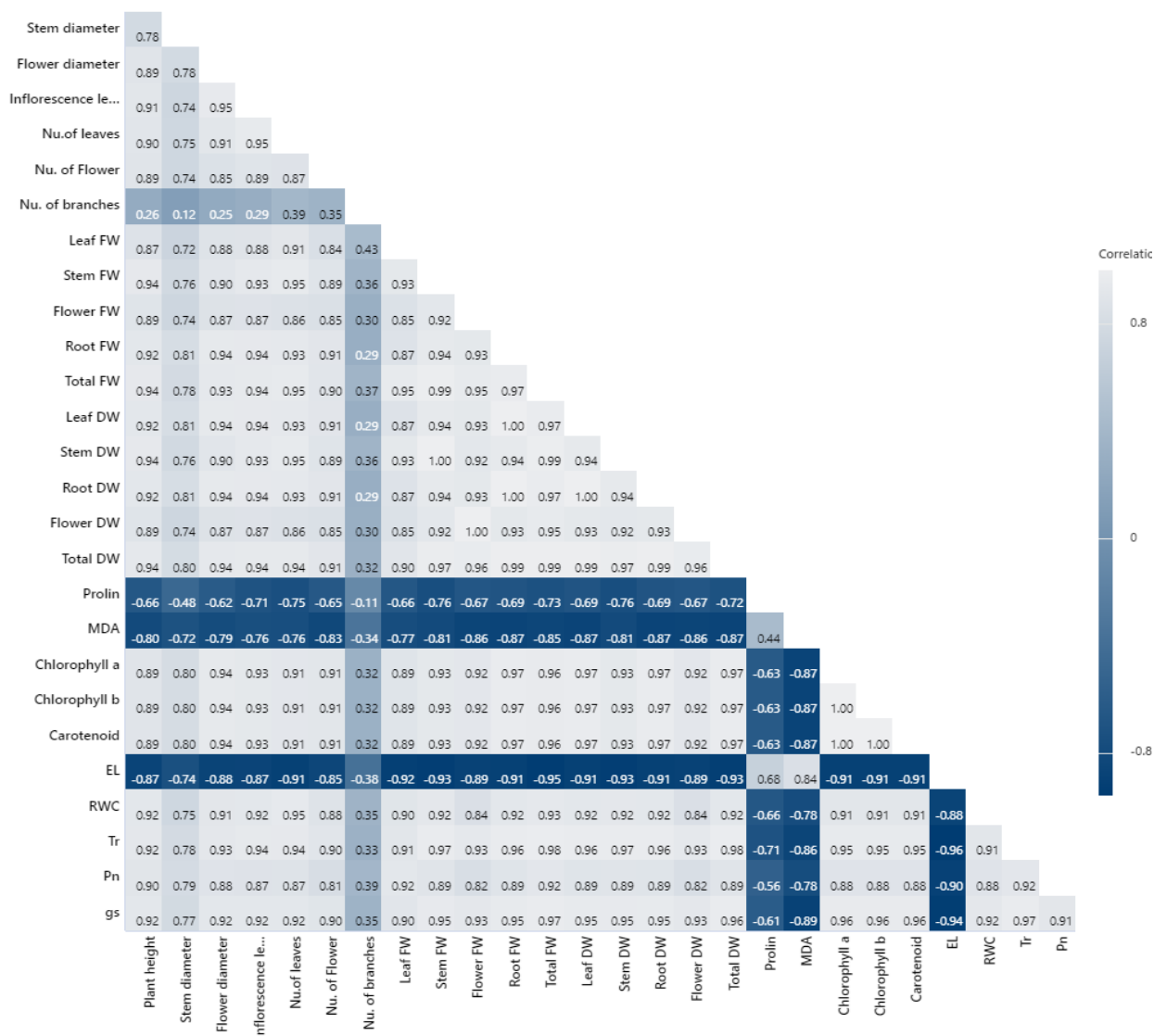


Fig. III. Pearson correlation coefficients of the measured traits in *Celosia argentea* under salinity stress and SA; correlation coefficients greater than 0.35 at the 5% level and greater than 0.45 at the 1% level are significant.

Salinity on the one side reduces the water potential of the root environment due to the reduction of available water potential for the plant and on the other, increases the toxicity of some ions for physiological and biochemical processes of the plant, which ultimately leads to reduced plant growth (Waqas-ud-Din Khan et al., 2020; Yu et al., 2021)

SA increases plant growth by increasing the rate of cell division and cell growth in meristematic regions and reduces the damage of abiotic

Considering the role of these hormones in the growth of plant cells, SA improves the growth of plants under stress conditions (Sharma et al., 2020). Qureshi et al. (2015) showed that SA treatment (3 mM) in *Dianthus caryophyllus* increased plant height, number of leaves, leaf area, number of flowers, stem length, and total dry matter. Furthermore, while salinity stress was reported to reduce total dry weight and total soluble protein in strawberry plant, foliar

application of SA improved these properties in the stressed plant (Dedejani et al., 2021).

In this study, although salinity stress reduced the pigments and RWC, foliar application of SA increased these traits. The amounts of plant pigments (chlorophyll a, b, and carotenoids) and RWC indicate its capacity for growth and development (Müller et al., 2001; Najar et al., 2020). Salinity stress reduces the leaf chlorophyll and carotenoids contents by impairing the absorption of some elements involved in chlorophyll synthesis, such as iron and magnesium. This reduction might be related to the degradation of the chloroplast structure and photosynthetic apparatus, photo-oxidation of chlorophyll, degradation of precursors of chlorophyll, inhibition of biosynthesis of new chlorophylls, and activation of chlorophyll degrading enzymes such as chlorophyllase (Kwon et al., 2019; Siddiqui et al., 2020). SA improved plant metabolism and increased its tolerance to salinity stress by preventing the destruction of chloroplast structure under saline conditions (Khodary, 2004).

Plants exposed to salinity stress also suffered from high levels of electrolyte leakage and MDA content (Fig. 1). These results showed that salinity induced an increase in the relative ion content of the apoplastic space, which in turn indicates reduced membrane stability. Plants grown under salinity stress and simultaneously supplemented with SA showed reduced electrolyte leakage levels and MDA content (Fig. 1). Thus improved membrane stability and reduced lipid peroxidation contributed to the increased growth and improved physiological and biochemical characteristics of this plant.

In this study, proline content increased with salinity stress, although foliar application of SA had an aggravating effect on increasing proline content. In addition, with increasing salinity stress, MDA and electrolyte leakage enhanced, although spraying SA reduced electrolyte leakage and MDA content. Plants increase the amount of proline when they are exposed to salinity stress, which is actually a kind of plant defense mechanism (El Moukhtari et al.,

2020). Proline increases plant resistance to stress by several mechanisms such as osmotic regulation, prevention of enzyme degradation, and protein preservation. Foliar application of SA would increase proline levels in the face of stress, which in turn can help the plant to reduce the effects of stress (Safari et al. 2021).

Positive effect of SA on plant water status can be related to the improved adjustment of cell osmotic pressure (Guan, et al., 2009; Safari et al., 2021). Under improved RWC, plants express higher *gs*. Increased stomatal conductance (*gs*) of SA-treated plants is highly and positively associated with higher *Pn*. Increased photosynthesis rate and *gs* under SA application has been already shown in other species (Van et al., 2013; Malerba and Cerana, 2018). Therefore, enhanced assimilation rate contributed to the better *C. argentea* growth and productivity following SA application, and this effect is partly related to improved water relations.

Conclusion

In the current study, foliar application of SA was performed to reduce the negative effects of salinity stress in *C. argentea*. In general, salinity stress reduced plant growth and development by reducing most of the growth traits. However, foliar application of SA somewhat mitigated the salinity stress. Assessment of physiological traits showed that foliar application of SA was effective on the relative water content and improved the water status. In addition, SA application was effective on the amount of proline content, which could lead to the maintenance of osmotic potential under stress. As a result of the changes made by foliar application of SA, the amount of photosynthesis in the plant improved and therefore, plant growth increased under salinity stress.

SA is a relatively inexpensive, accessible, and environmentally friendly material. In this view, foliar application of SA (in particular with 1 mM concentration) presents a cost-effective and green approach to mitigate the negative effects of salinity stress.

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