



Effect of adjusting the salinity of irrigation water on *Catharanthus roseus* plant pigments and enzyme activity changes using jasmonic acid and salicylic acid

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ABSTRACT

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This research investigated the effect of foliar spraying of jasmonic acid and salicylic acid on the morphological traits, plant pigments, and enzyme activity of *Catharanthus roseus* under salt stress. The experiment used a factorial design based on a completely random statistical design with three repetitions. The treatments included sodium chloride salt and foliar spraying of jasmonic acid and salicylic acid in concentrations of zero, 50, and 100 mg/l and their interaction. Two weeks after transferring the seedlings to the pot, watering with sodium chloride was done 3 times every two days. Foliar spraying was done weekly, one week after applying salinity. The results showed that the highest fresh and dry weight of shoots and roots, root length, plant height, cell membrane stability index, total chlorophyll, protein content, catalase, superoxide dismutase, and peroxidase enzyme activity were observed in control and the highest flowers number and anthocyanin content were observed in 50 mg/l sodium chloride with 100 mg/l salicylic acid treatment. Also, the highest proline content was in the 50 mg/l sodium chloride treatment. According to the results, foliar application of 100 mg/l salicylic acid has an effective role in inhibiting the negative effects of salinity stress in *Catharanthus roseus*.



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1. Introduction

Catharanthus roseus L., commonly known as Madagascar periwinkle, is an ornamental and medicinal plant belonging to the Apocynaceae family, native to tropical regions. Depending on the regional climate, it is commonly used as a perennial flowering plant in landscaping (AlhverdiZadeh and Danaee, 2023). This plant is highly valuable in the treatment of various cancers, including breast, lung, uterine, skin, and bone cancers. The presence of alkaloids such as vincristine and vinblastine in its vegetative organs and roots contributes to its therapeutic properties. Both alkaloids exhibit anti-tumor properties by binding to microtubules and halting cell division during the metaphase of mitosis, which makes them useful in chemotherapy (Danaee, 2020).

Environmental stresses are limiting factors in the production of horticultural crops. By disrupting the plant's natural metabolism, they restrict growth and ultimately reduce yield. After drought stress, salinity stress is one of the most significant environmental stresses, causing reduced plant

growth, especially in arid and semi-arid regions of the world. Salinity negatively impacts the growth and development of crops through physiological and biochemical processes such as chlorophyll synthesis, photosynthesis, respiration, and ionic homeostasis. induced oxidative stress leads to the production of reactive oxygen species (ROS), which cause the oxidation of lipids, proteins, nucleic acids, and chlorophyll (Bin-Jumah et al., 2021). In one study, salinity stress in Damask rose (*Rosa damascena*) reduced the fresh and dry weight of leaves and shoots, the relative water content of leaves and shoots, total chlorophyll, carotenoids, and protein content. However, it increased the activity of superoxide dismutase, catalase, and guaiacol peroxidase enzymes, as well as the total phenol content in the leaves (Omidi et al., 2020). Similarly, in pot marigold (*Calendula officinalis* L.), salinity stress reduced the fresh and dry weight of roots, plant height, number of leaves and flowers, total chlorophyll and carotenoid content,. However, it increased proline levels and superoxide dismutase enzyme activity (Shabani Fard et al., 2024).

When exposed to stress conditions, plants send signals to various cellular metabolic pathways to activate the defence genes. Several molecules, including jasmonic acid and salicylic acid, act as signal transducers under stress conditions (Haghighi and Mansouri, 2019).

Jasmonic acid is a plant hormone that plays a key role in regulating plant growth processes, including leaf abscission, stomatal closure, β -carotene synthesis, and ethylene production. It also enhances plant resistance to stress by inducing the enzyme responsible for proline synthesis (Rezaei Nasab et al. 2016). In a study, the application of jasmonic acid in safflower (*Carthamus tinctorius* L.) increased leaf water content, catalase enzyme activity, and protein content (Khademian et al. 2019). Similarly, in peppermint (*Mentha piperita* L.) under salinity stress, jasmonic acid application increased superoxide dismutase and catalase activity, as well as essential oil production (Fathi et al. 2020).

Salicylic acid is a phenolic compound recognized as a hormone-like substance. It plays a crucial role in enhancing photosynthesis and inducing tolerance to both biotic and abiotic stresses. As a non-enzymatic antioxidant, it regulates physiological activities and enhances plant resistance. It also influences stomatal closure, transpiration, inhibition of ethylene biosynthesis, and nutrient uptake and translocation (Danaee and Abdossi, 2016). The effects of salicylic acid are variable, depending on concentration, timing, and plant species. At appropriate concentrations, it protects the photosynthetic apparatus by reducing chlorophyll degradation, enhancing cellular antioxidant capacity, and promoting protein synthesis (Haghighi and Mansouri, 2019). Application of salicylic acid in holy basil (*Ocimum sanctum* L.) improved the fresh and dry weight, relative water content, chlorophyll a, b, and total chlorophyll content. It also increased total phenol and flavonoid content, as well as total antioxidant activity under water stress (Mafakheri and Asghari, 2023). Furthermore, Abdolmohammadi et al. (2018) reported increases in plant height, fresh and dry weight of stems and roots, root length, relative water content in leaves, and protein content in stock (*Matthiola incana* L.) under salinity stress.

According to the water deficiency and the expansion of saline areas in Iran, employing suitable methods to increase plant tolerance to stressful conditions is very important. Therefore, this research aimed to evaluate the morphophysiological traits and enzymatic activity of Madagascar periwinkle (*Catharanthus roseus*) under salinity stress following foliar applications of jasmonic acid and salicylic acid.

2. Materials and Methods

2.1 Experimental design and treatments

To investigate the effects of jasmonic acid and salicylic acid on photosynthetic pigments and enzymatic activity of *Catharanthus roseus* under salinity stress, a greenhouse experiment was conducted with a 16-hour light/8-hour dark photoperiod, a temperature of 23 ± 2 °C, and a relative humidity of 50-60%. Treatments included sodium chloride (NaCl) at 0, 50, and 100 mg/L, foliar applications of jasmonic acid and salicylic acid at 0, 50, and 100 mg/L, and their interactions. Cuttings (8-10 cm long) were prepared in late winter, their lower leaves removed, and planted in a light, moist substrate of peat moss and leaf mold. Two weeks after

transplanting to the main pots, NaCl solution (150 ml) was applied three times at two-day intervals. Jasmonic acid and salicylic acid solutions (Sigma-Aldrich, Germany) were prepared in distilled water, and foliar application was performed weekly, starting one week after the salinity stress was applied. Sampling and trait assessment were performed approximately 45 days after transplanting the cuttings.

2.2 Evaluated traits

2.2.1 Morphological traits

The fresh weight of shoots and roots was measured immediately after harvest, and the dry weight was recorded after 72 hours at 60 °C with an accuracy of 0.01 g. Plant height and root length were measured using a metal ruler. Additionally, the number of flowers was counted, and the average was recorded (Soroori and Danaee, 2023).

2.2.2 Cell membrane stability index

To evaluate the cell membrane stability index (MSI), the petals of *Catharanthus roseus* were placed in a water bath at 30 °C for one hour, and the electrical conductivity (EC_1) was measured using a conductivity meter. The samples were then autoclaved at 121 °C for 20 minutes to determine EC_2 . Finally, Ion leakage was calculated using Eq. 1 (Roumani et al., 2022):

$$MSI = [1 - (EC_1/EC_2)] \times 100 \quad (1)$$

where *MSI* is the cell membrane stability index.

2.2.3 Anthocyanin content

The anthocyanin content of the petals was measured using a methanol extraction solution with 1 N hydrochloric acid. The absorbance of the obtained extract was read at wavelengths of 530 nm and 657 nm using a spectrophotometer, and the anthocyanin content was calculated using the following formula (Danaee and Abdossi, 2016):

$$ACN = A_{530\text{ nm}} - 1/4 A_{657\text{ nm}} \quad (2)$$

where *ACN* represents the anthocyanin content and *A* denotes the absorbance.

2.2.4 Total chlorophyll content in leaves

The total chlorophyll content in leaves was extracted using dimethyl sulfoxide (DMSO). The absorbance of the obtained solution was measured with a UV-visible spectrophotometer (Model Spectro Flex 6600) at wavelengths of 663 nm for chlorophyll a and 645 nm for chlorophyll b. The total chlorophyll was ultimately calculated using the following formula (Rahmanifard et al., 2024):

$$Chl = 20.2 (A_{645}) + 8.02 (A_{663}) \times V / (1000 \times W) \quad (3)$$

where *V* is sample volume (ml), *W* is sample weight (mg), and *A* is absorbance at the specified wavelengths (nm).

2.2.5 Leaf proline content

To evaluate the leaf proline content, sulfuric acid was added to the leaf sample, which was then filtered through filter paper. The resulting solution was mixed with ninhydrin reagent and sulfosalicylic acid. After heating in a warm water bath, the reaction was stopped in the presence of ice, and toluene was added to the mixture. The absorbance of the upper phase was read at a wavelength of 520 nm using a spectrophotometer, and

the results were expressed in mg/g dry weight (DW) (Mirakhorli et al. 2022).

2.2.6 Petal protein content

The petals of *Catharanthus roseus* were initially blended with phosphate buffer, and after centrifugation, the supernatant was mixed with Bradford reagent. The absorbance was then measured at a wavelength of 595 nm using a spectrophotometer. The protein content was calculated in g/mg fresh weight (FW) (Danaee et al. 2010).

2.2.7 Activity of catalase, superoxide dismutase, and peroxidase enzymes in petals

The activity of catalase enzyme was measured by calculating the decrease in absorbance of H₂O₂ at a wavelength of 240 nm. The reaction solution included the enzyme extract, phosphate buffer, and hydrogen peroxide, following the method outlined by Dareini et al. (2014). The activity of superoxide dismutase (SOD) was assessed based on its ability to inhibit the photochemical reduction of nitroblue tetrazolium (NBT) to formazan by superoxide radicals. The reaction solution included the enzyme extract, methionine, riboflavin, NBT, and peroxidase, analyzed according to the guaiacol method with the presence of hydrogen peroxide and guaiacol buffer. Absorbance readings were taken at wavelengths of 560 nm and 530 nm using the spectrophotometer (Soroori et al. 2021), and enzyme activities were calculated as units per gram of fresh weight.

2.3 Experimental design and statistical data analysis

The experiment was designed as a factorial arrangement in a completely randomized design with three replications and two factors: salinity stress and foliar application of jasmonic acid

and salicylic acid, including their interactions. Data analysis was performed using SAS software (version 9.1), and mean comparisons were conducted using Duncan's multiple range test at 5% levels. Graphs were created using Excel 2016 software.

3. Results and Discussion

3.1 Analysis of variance of data

The results of the analysis of variance presented in Table 1 indicated that the effect of salinity stress on fresh and dry weight of aerial parts and roots, root length, plant height, cell membrane stability index, petal anthocyanin content, total chlorophyll content in leaves, proline and protein content, and the activity of catalase, superoxide dismutase, and peroxidase enzymes was significant at the 1% level. The number of flowers was significant at the 5% level.

The effect of foliar application treatment on fresh weight of aerial parts and roots, dry weight of roots, cell membrane stability index, petal anthocyanin content, proline content, and the activity of catalase, superoxide dismutase, and peroxidase enzymes was significant at the 1% level, while the dry weight of aerial parts, root length, plant height, number of flowers, total chlorophyll content, and protein content were significant at the 5% level.

The interaction effect of salinity stress and foliar application was also significant at the 1% level for fresh and dry weight of aerial parts and roots, root length, cell membrane stability index, petal anthocyanin content, total chlorophyll content in leaves, proline and protein content, and the activity of catalase, superoxide dismutase, and peroxidase enzymes. Additionally, it was significant at the 5% level for plant height and number of flowers.

Table 1 Analysis of variance in the effect of salt stress and jasmonic acid and salicylic acid spraying on the morphological and physiological characteristics of *Catharanthus roseus*

S.O.V	Stress	Treatment	Stress× Treatment	Error	CV (%)
DF	2	4	8	30	---
Shoots fresh weight	123.43**	38.26**	75.18**	1.24	9.36
Shoots dry weight	41.91**	17.45*	28.17**	0.53	10.23
Roots fresh weight	21.64**	11.37**	16.42**	0.32	11.57
Roots dry weight	7.38**	2.69**	5.26**	0.71	10.65
Length of longest root	61.32**	22.56*	37.28**	0.82	10.76
Plant height	113.64**	31.27*	58.42*	1.06	9.42
Flower number	85.39*	24.57*	49.32*	0.95	10.21
Cell membrane stability index	248.31**	83.26*	15.43*	2.61	9.45
Anthocyanin	7.15**	2.83*	4.38**	0.059	11.52
Total chlorophyll	52.38**	19.16*	38.14**	0.72	10.86
Proline	18.25**	5.37*	11.42**	0.068	10.43
Protein	341.97**	62.38*	206.45**	3.26	9.72
Catalase	19.42**	7.11**	13.64**	0.075	11.26
Superoxide dismutase	12.83**	6.32**	9.85**	0.049	11.45
Peroxidase	38.76**	21.45**	29.37**	0.49	10.81

*, ** and ns: Significant at P<0.05, P<0.01 and insignificant, respectively.

3.2 Morphological Traits

The comparison of mean data in Table 2, showed that the highest fresh and dry weights of shoots were 41.25 and 9.26 g in control, while the lowest were 25.47 and 6.17 g in sodium chloride 100 mg/l treatment. The highest root fresh weight was

7.82 g and the dry root weight was 2.63 g in the control group, whereas the lowest root fresh and dry weights were 5.06 and 1.56 g in sodium chloride 100 mg/l treatment. Additionally, the longest root length was observed at 18.12 cm in the control group, while the shortest was 13.53 cm in sodium chloride 100

mg/l treatment. The highest plant height was 28.31 cm in control, and the lowest was 19.87 cm in sodium chloride 100 mg/l treatment. The highest and lowest flower numbers were in control (18) and sodium chloride 100 mg/l treatment (13).

The findings of this study indicated that increasing the concentration of sodium chloride resulted in a decrease in the fresh and dry weight of aerial parts and roots, the length of the longest root, plant height, and flower number. This reduction is attributed to the effects of salinity stress, which leads to decreased photosynthesis, reduced water availability, nutrient imbalance, osmotic regulation issues, and toxicity from chloride and sodium ions. These factors adversely affect various physiological processes in plants, such as photosynthesis, changes in chloroplast absorption spectra, and reduced stomatal conductance, ultimately leading to decreased growth indices (Fazeli et al. 2017). Salinity stress limits root growth and elongation due to reduced water uptake by the plant (Salimi and Shekari, 2012). Additionally, inadequate cell turgor and the plant's increased use of synthesized materials to

cope with stress can hinder normal cell development, resulting in reduced plant height (Safari Mohamadiyeh et al., 2015). Furthermore, decreased photosynthesis reduces the carbohydrates necessary for cell division and elongation, causing leaves to act as consumers and inhibiting the plant's reproductive growth (Kalhor et al. 2018).

In this study, the application of salicylic acid and jasmonic acid improved the growth indices of the plant under salinity stress conditions. Salicylic acid plays a protective role in response to environmental stresses by enhancing CO₂ assimilation, photosynthesis, mineral uptake, influencing plant hormones, and maintaining tissue water, all of which promote plant growth and flowering (Aghaei Joubani et al. 2015). Similarly, jasmonic acid positively influenced growth indices by affecting cell division and elongation (Mazarie et al. 2019). The results of this study are consistent with the findings of Moradian et al. (2023) in marigolds (*Tagetes patula*) and Sharifian Jazi et al. (2023) in basil (*Ocimum basilicum* L.).

Table 2 The effect of salt stress and jasmonic acid and salicylic acid spray on the morphological characteristics of *Catharanthus roseous*

Salt stress (mg/l)	Spraying (mg/l)	Shoot fresh weight (g)	Shoot dry weight (g)	Root fresh weight (g)	Root dry weight (g)	Root height (cm)	Plant height (cm)	Flower number
Control	0	41.25 ^a	9.26 ^a	7.82 ^a	2.63 ^a	18.12 ^a	28.31 ^a	18.00 ^a
	0	33.73 ^f	7.26 ^f	5.73 ^g	2.04 ^e	15.82 ^f	23.61 ^e	15.50 ^d
50	Jasmonic acid 50	36.58 ^d	8.52 ^c	7.32 ^c	2.46 ^b	16.81 ^d	26.39 ^c	17.50 ^b
	Jasmonic acid 100	35.12 ^e	7.65 ^e	6.42 ^e	2.21 ^d	16.47 ^e	25.12 ^d	15.67 ^d
	Salicylic acid 50	38.65 ^c	8.01 ^d	6.87 ^d	2.31 ^c	17.32 ^c	26.34 ^c	17.00 ^c
	Salicylic acid 100	40.16 ^b	8.98 ^b	7.51 ^b	2.59 ^a	17.75 ^b	27.45 ^b	18.33 ^a
	0	25.47 ^j	6.17 ^j	5.06 ^j	1.56 ⁱ	13.53 ^j	19.87 ⁱ	13.00 ^h
100	Jasmonic acid 50	32.11 ^g	6.62 ^{hi}	5.43 ^h	1.75 ^j	15.26 ^h	22.46 ^g	14.67 ^{ef}
	Jasmonic acid 100	27.23 ⁱ	6.43 ⁱ	5.21 ⁱ	1.67 ^h	14.72 ⁱ	20.86 ^h	13.67 ^g
	Salicylic acid 50	29.07 ^h	6.76 ^h	5.51 ^h	1.78 ^g	15.29 ^h	22.75 ^{fg}	14.33 ^f
	Salicylic acid 100	33.9 ^f	7.04 ^g	6.14 ^f	1.95 ^f	15.82 ^g	23.17 ^f	15.00 ^e

The same letters indicate no significant difference at the level of $P \leq 0.05$

3.3 Cell membrane stability index

The results of the mean comparison indicated that the highest and lowest cell membrane stability index was 78.64 and 52.41% in control and sodium chloride 100 mg/l treatment (Fig.1). The increase in lipid peroxidation and ion leakage under stress conditions occurs due to the rise in free radicals, which affects the plasma membrane's functionality and can lead to cell death under severe stress (Jouyban 2012). In miniature rose (*Rosa chinensis*), salinity stress also resulted in increased ion leakage, consequently reducing the cell membrane stability index (Shahbani et al. 2018).

The application of salicylic acid and jasmonic acid improved the cell membrane stability index under salinity stress, indicating the positive impact of these compounds on membrane integrity and the enhancement of cellular membrane conditions against oxidative damage (Saheri et al., 2023). Additionally, in psyllium (*Plantago ovata* Forssk), the

application of salicylic acid also increased the cell membrane stability index (Roumani et al., 2022).

3.4 Anthocyanin content

The highest anthocyanin content in the petals was recorded 3.12 mg/g FW in sodium chloride 50 mg/l with salicylic acid 100 mg/l treatment, and the lowest was 1.79 mg/g FW in sodium chloride 100 mg/l treatment (Fig. 2). The reduction in anthocyanin biosynthesis at high concentrations of sodium chloride can be attributed to the toxic effects of sodium and oxidative stress caused by salinity (Khosravi et al. 2022). Salicylic acid and jasmonic acid enhance the accumulation of plant pigments, including anthocyanins, by increasing photosynthesis, the synthesis of plant hormones, and enzymes (Aldesuquy and Ghanem, 2015). Rasoli and Gholipoor (2023) reported the positive effects of salicylic acid and jasmonic acid in echinacea (*Echinacea purpurea*).

Fig. 1 Effect of salt stress and jasmonic acid and salicylic acid spraying on cell membrane stability index of *Catharanthus roseous*

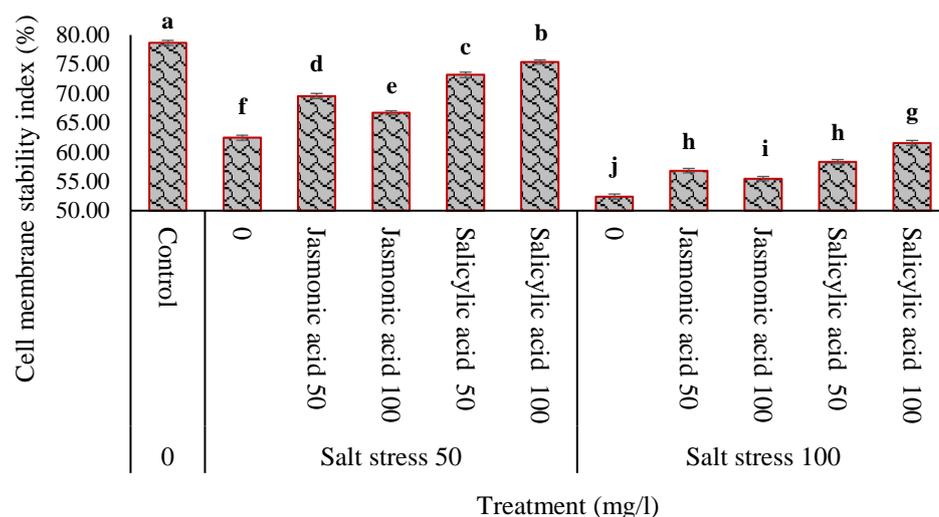
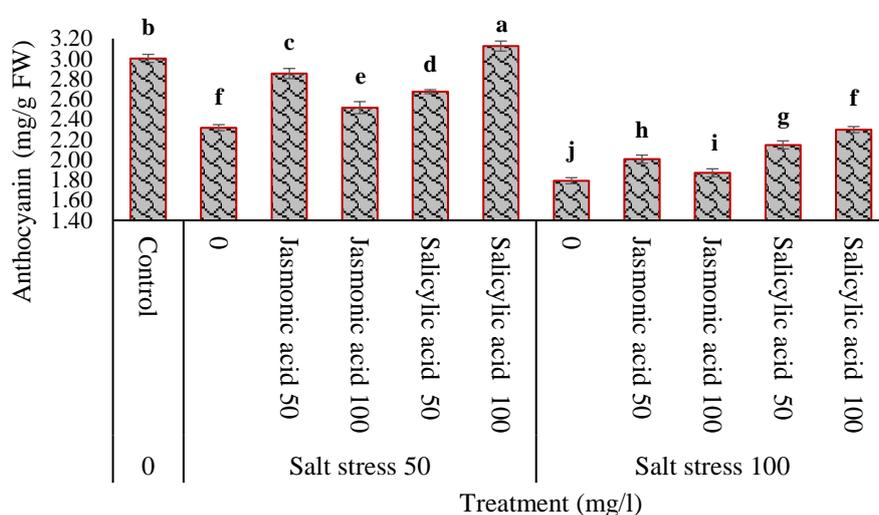


Fig. 2 Effect of salt stress and jasmonic acid and salicylic acid spraying on anthocyanin content of *Catharanthus roseous*



3.5 Total chlorophyll content

As illustrated in Fig. 3, the highest total chlorophyll content was found to be 15.82 mg/g FW in control, and the lowest was 9.63 mg/g FW in sodium chloride 100 mg/l treatment. Osmotic stress in saline environments leads to an increase in reactive oxygen species, which in turn activates the enzyme chlorophyllase. Additionally, salt stress limits the uptake of elements such as magnesium and iron, which are crucial for chlorophyll synthesis (Attarzadeh et al. 2014). The reduction in total chlorophyll content under salt stress conditions has been reported in lemon balm (*Melissa officinalis* L.) by Ghasemian et al. (2019).

In this study, the foliar application of salicylic acid resulted in an increase in chlorophyll levels in plants under stress, indicating this compound's ability to mitigate stress effects. It appears that the foliar application of salicylic acid and jasmonic acid enhances the antioxidant capacity of cells, which reduces lipid peroxidation and provides greater protection for cellular membranes and photosynthetic

pigments, thereby preventing chlorophyll catabolism (Farahbakhsh and Pasandipour, 2017; Kheiry et al. 2017). According to the results of this experiment, Gorni et al. (2020) also reported the positive effect of salicylic acid on total chlorophyll content in yarrow (*Achillea millefolium* L.).

3.6 Proline content

The comparison of mean data revealed that the highest and lowest proline content were 4.16 and 2.51 mg/g FW in sodium chloride 100 mg/l and control treatment, respectively (Fig. 4). The increase in proline under stress conditions acts as a defensive mechanism and enhances plant tolerance to stress by maintaining cellular pH, regulating water within plant cells, and stabilizing enzymes and proteins (Iqbal et al. 2014). Salicylic acid and jasmonic acid have been shown to increase proline levels by enhancing the activity of the enzymes involved in proline synthesis (Salimi et al. 2012). The results of this experiment are consistent with the findings of Yeganehpour et al. (2017) in coriander (*Coriandrum sativum* L.).

Fig. 3 Effect of salt stress and jasmonic acid and salicylic acid spraying on total chlorophyll content of *Catharanthus roseous*

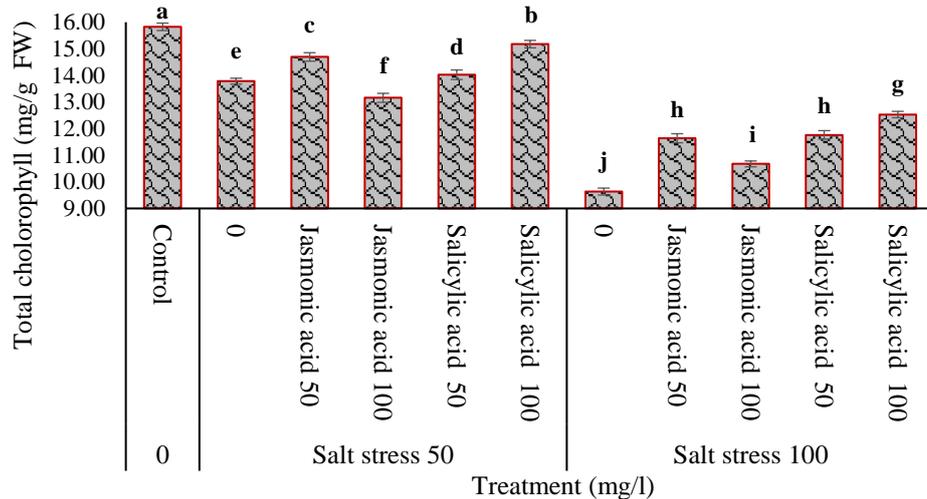


Fig. 4 Effect of salt stress and jasmonic acid and salicylic acid spraying on proline content of *Catharanthus roseous*

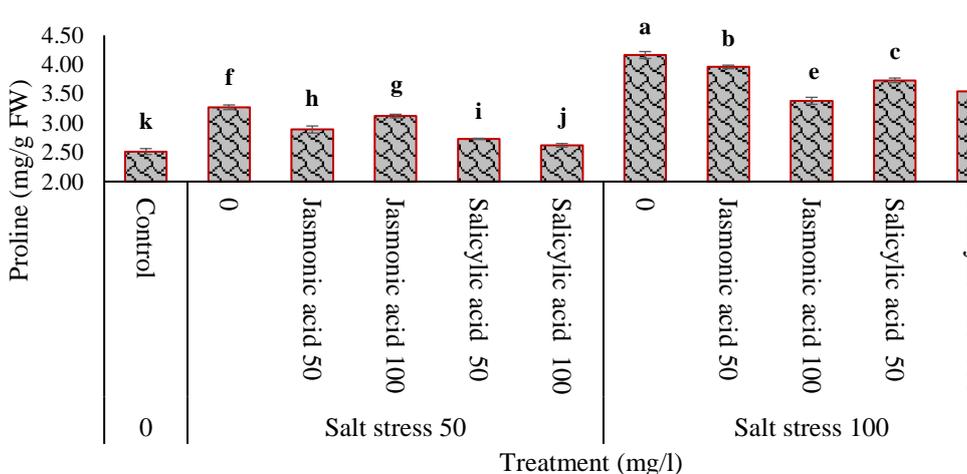
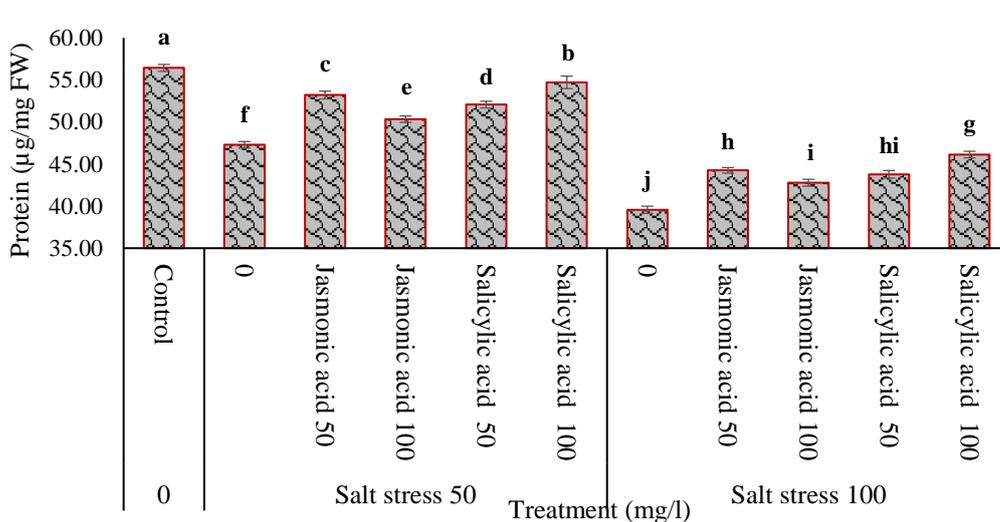


Fig. 5 Effect of salt stress and jasmonic acid and salicylic acid spraying on protein of *Catharanthus roseous*



3.7 Protein content

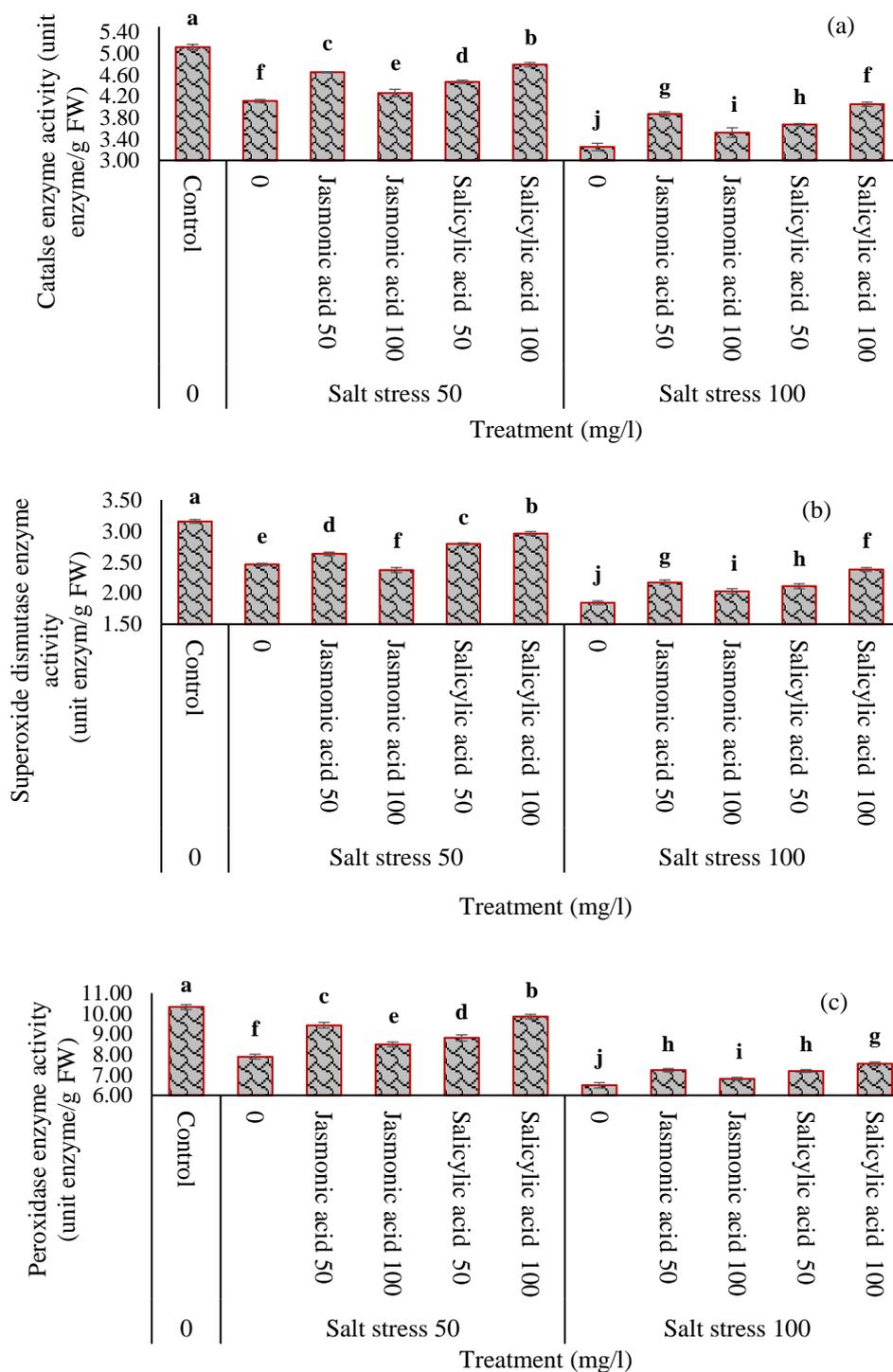
The results of the comparison of mean data showed that the highest protein content was 56.43 µg/mg FW in the control, and the lowest was 39.56 µg/mg FW in sodium chloride 100 mg/l treatment (Fig. 5). High sodium concentrations hinder

potassium absorption in plants, as potassium helps in protein synthesis by connecting tRNA to the ribosome. As a result, sodium disrupts protein synthesis (Askary et al. 2016). The protein content increased under the influence of salicylic acid and jasmonic acid, which play a significant role in protecting cellular membranes and organelles involved in protein

synthesis from oxidative stress effects, thus preventing protein degradation. Additionally, salicylic acid enhances protein synthesis through the activity of the nitrate reductase enzyme

(Rasouli et al. 2018). The results of this experiment align with the findings of Nisar et al. (2018) in *Petunia hybrida* Vilm.

Fig. 6 The effect of salt stress and jasmonic acid and salicylic acid spraying on catalase a), superoxide dismutase b) and peroxidase and c) enzyme activity of *Catharanthus roseus*



3.8 Activity of catalase, superoxide dismutase, and peroxidase enzymes

The comparison of mean data indicated that the highest catalase, superoxide dismutase, and peroxidase enzyme activities were 5.12, 3.15, and 10.32 units enzyme/g FW in control, and the lowest values were 3.25, 1.84, and 6.49 units enzyme/g FW in sodium chloride 100 mg/l treatment, respectively (Fig. 6). Salinity stress leads to the conversion of

superoxide radicals (O_2^-) into hydrogen peroxide (H_2O_2) within cells, and the detoxification of free radicals is carried out by catalase, superoxide dismutase, and peroxidase enzymes (Eskandari Zanjani et al. 2013). Changes in the activity levels of these antioxidant enzymes can significantly affect plant resistance, which varies depending on the plant species' sensitivity, growth stage, and the intensity and duration of the stress (Sirousmehr et al., 2015).

In this study, an increase in stress intensity resulted in a decrease in antioxidant enzyme activities, indicating a potential lack of tolerance of the studied plant species under saline stress conditions. Similarly, a reduction in the activities of superoxide dismutase and peroxidase enzymes under saline stress has been reported in peppermint (*Mentha piperita* L.) (Danaee and Abdossi, 2019). The application of salicylic acid and jasmonic acid moderates the production of reactive oxygen species and enhances plant resistance to stress through both direct and indirect regulatory roles on antioxidant enzyme activities (Singh and Gautam, 2013). The results of this experiment are consistent with the findings of Taghizadeh Tabaria et al. (2022) in borage (*Borago officinalis* L.).

4. Conclusion

In this study, the effects of jasmonic acid and salicylic acid on plant pigments and enzymatic activity of *Catharanthus roseus* under salinity stress conditions were investigated.

1. The highest fresh and dry weights of shoots and roots, length of longest root, plant height, cell membrane stability index, leaves total chlorophyll content, protein content, and catalase, superoxide dismutase, and peroxidase enzymes activities were observed in control.

2. The highest flowers number and petals anthocyanin content were obtained in sodium chloride 50 mg/l with salicylic acid 100 mg/l treatment.

3. The highest proline content was also found in the sodium chloride 100 mg/l treatment.

In this study, salinity stress led to a reduction in growth indices and enzymatic activity of *Catharanthus roseus*, while the application of jasmonic acid and salicylic acid helped improve quality and flowering which mitigated the negative effects of stress. Based on the results, the application of salicylic acid at a concentration of 100 mg/l showed the most significant impact on *Catharanthus roseus* under salinity stress conditions. Therefore, it is suggested that future studies investigate the foliar application of other compounds such as amino acids, polyamines, and nutrients to reduce the effects of salinity stress in *Catharanthus roseus*.

Statements and Declarations

Data availability

The data obtained from the experiments conducted in this study are presented in the text of the article.

Conflicts of interest

The author of this paper declared no conflict of interest regarding the authorship or publication of this paper.

Author contribution

S. Tarameshloo: Project execution, data collection, and drafting the article; E. Danaee: Project design and management, data analysis and results, review, and approval of the article.

AI Use Disclosure

During the preparation of this manuscript, the authors used ChatGPT for language translation. All content has been

carefully reviewed and revised by the authors, who take full responsibility for the final version of the manuscript.

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