



Silicon Stimulates Physiochemical Properties of Coriander (*Coriandrum sativum* L.) to Improve Growth and Yield under Salt Stress

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Abstract

Silicon (Si) has a principal role in mitigating the adverse effects of salt stress on plant growth; however, its effects on physiochemical properties of medicinal plants under salinity is still unknown. This study investigated the effect of Si and salinity on growth, chlorophyll and water content, the antioxidant capacity of coriander (*Coriandrum sativum* L.) leaves as factorial in a randomized complete block design (RCBD). Foliar application of 50, 100, and 200 mg/L Si and 50, 100, and 200 mM NaCl were applied on 4-leaf plants. The results revealed reduced growth by representing lower shoot and root dry weight through progressing the salinity. 100 and 200 mg/L Si represented a greater role in alleviating the salinity stress on growth properties. Salinity significantly reduced chlorophyll (Chl) and relative water content (RWC), but they were improved by Si spraying. Total phenol content (TPC) and total flavonoid content (TFC) significantly increased up to 100 mM NaCl and then decreased at 200 mM NaCl. Salinity led to increases in catalase (CAT) and superoxide dismutase (SOD) activity, but higher Si concentration reduced them. 100 mg/L Si and 100 mM NaCl was the best treatment for obtaining the optimum essential oil (EO) percentage and yield. Finally, 100 mg/L Si can be suggested to promote the plant growth and yield by changing physiochemical characteristics under medium or severe salinity.

Keywords: Essential oil yield, Foliar-applied silicon, Phenolic content, Salinity

Introduction

Coriander (*Coriandrum sativum* L.) is a medicinal annual plant of Apiaceae, which is widely used because of its high nutritional and medicinal values [1]. Coriander is native to the European-Mediterranean area and was cultivated in China in the 1st century BC. Recently, it has been widely cultivated as a valuable herb all over the world [2]. In the food industry, coriander is confirmed in food and pharmaceutical industries by the US Food and Drug Administration. It has a long history as a culinary herb is the source of aroma compounds and essential oils (EOs) with biologically active components inducing antifungal, antibacterial, and antioxidant activities [3]. Coriander has various medicinal uses like treatment of disorders in skin inflammation, digestive, respiratory, and urinary systems [2,4].

Salinity is a principal abiotic factor that adversely affects the soil and limits crop productivity worldwide [1]. The

saline condition occurs when the electrical conductivity (EC) of the saturation soil is 4 dS m⁻¹ or more [1, 5]. Although the threshold level of saline stress is different among plant species, their growth and development are adversely affected at this level (1). Salt stress induces osmotic and ionic stresses that can lead to secondary stresses, for example, nutritional imbalances, and oxidative stress resulted because of the generation of reactive oxygen species (ROS) in plants [6]. Free and non-free radicals of ROS can detrimentally affect physiological and biochemical processes including water relations, transpiration, photosynthesis, cellular homeostasis, enzymatic and non-enzymatic activities in plants [1]. However, plant use mechanisms such as active Na⁺ efflux, inhibition of Na⁺ influx, up-regulation of antioxidant defense systems, accumulation of compatible osmolytes to alleviate salt stress [7].

Silicon (Si) as the main element in Earth's crust is addressed to increase plant growth under abiotic

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conditions [8,9]. Si has a positive effect on the uptake of other nutrients by plants. In saline conditions, Si can inhibit the excessive accumulation of Na and Cl in plant roots and leaves [10].

Despite the effective role of Si in mitigating salt stress conditions that have been reported in the growth and yield of many plants [1,11,12,14,15], little is known about the effect of Si on EO yield, antioxidant capacity, and water content of coriander leaves. Therefore, this work assessed the effects of salinity and Si foliar application on growth, photosynthesis pigments, phenolic content, water content, and EO percentage and yield of coriander leaves under salt stress. We hypothesized that Si can stimulate the biochemical properties to improve the plant growth and EO yield under salinity. By evaluating the possibility of the utilization of Si in alleviating salinity stress with the change in plant growth and yield, the findings of this study help to suggest the best treatment in improving the growth and EO yield of coriander plants.

Material and Methods

Growth Conditions

The coriander seeds were obtained from the Medicinal Plants Research Center, Institute of Medicinal Plants, ACECR, Karaj, Iran. The pot experiment was conducted in a greenhouse with a photoperiod of 16/8 (lightness/darkness) and relative humidity of 65%-80% in the University of Tehran, Karaj (1312 m asl, 35°48'45" N, 51°01'30" E). The pots had a top diameter of 19 cm, a base diameter of 13 cm base, and a height of 10 cm. The pots were filled with cocopeat and perlite (at a ratio of 2:1).

Experimental Design and Treatments

The factorial experiment was carried out in a completely randomized design (RCD) with three replications. All plants were normally irrigated up to 4 leaf stage, then the uniformly sized plants were selected for salinity treatments. Four different levels of salt stress were 50, 100, 200 mM NaCl, and non-saline conditions (control). The irrigation with saline solution (250 mL in each pot) occurred every three days for 2 months. To avoid the accumulation of salt at the bottom of the pots, the pots were completely watered (non-saline water) after 10 days.

Dry Weight

To measure dry weight, plants were cut from the bottom of the stem, and they immediately were transferred in the envelopes and dried at 40 °C in the dark condition till got a constant weight (16).

Chlorophyll (Chl) Assay

The contents of Chl a and b were extracted according to the method proposed by Arnon [17]. 200 mg of fresh samples were homogenized in 8 ml 80% acetone. After that, the mixture was centrifuged at 4 °C for 15 min (3000 rpm). Supernatants were used for analyzing chlorophyll content. Absorbance was determined at 645 and 663 nm by the spectrophotometer.

Relative Water Content (RWC) Measurement

The RWC of leaves was calculated as a percentage according to the method of Dhopte and Manuel [18] as follows:

$$RWC = \frac{(FW - DW)}{(SW - DW)} \times 100$$

Where FW is fresh weight, SW is leaf weight after soaking for 24 hours at room temperature and DW is leaf dry weight after drying for 24 h at 75 °C.

Antioxidant Enzyme Assays

300 mg of leaf samples were used to extract protein for enzyme activity. Extraction was made with 2 mL phosphate buffer (100 mM, pH = 6.8) containing EDTA (1 mM), PMSF (100 Mm) and 2% PVP. The extract was centrifuged for 15 minutes at 4 °C and 15000 g, the supernatant was used for the enzymatic assays and stored at -80 °C for further use.

The catalase (CAT) activity was determined by using the protocol described by Aebi [19]. 300 µl enzyme extract and 200 H₂O₂ (1%) was added in phosphate buffer 50 mM (pH 7). Enzymatic activity was defined by measuring the total oxygen produced from enzymatic dissociation of H₂O₂ in darkness for 1 min. The evolution of oxygen was defined by measuring the decrease in H₂O₂ absorption at 240 nm ($\epsilon = 40 \text{ mM}^{-1} \text{ cm}^{-1}$). Results were expressed as absorbance *enzyme unit*/mg of protein. Superoxide dismutase (SOD) was evaluated via inhibiting the photochemical decrease of nitroblue tetrazolium (NBT) by the enzyme at 560 nm according to the method of Giannopolitis and Ries [20]. Reaction solution included 50 µL of enzyme extract, 50 mM potassium phosphate buffer (pH 7.8), 13 mM, methionine, 0.075 mM NBT, 0.1 mM EDTA, 150 µL of deionized water and 0.002 mM riboflavin. The tubes were irradiated for 10 min.

Determination of Total Phenolic Content (TPC)

Folin–Ciocalteu reagent was selected to measure TPC spectrophotometrically [21]. 100 µl of the MeOH solution of the precisely measured weight of investigated plant 1–10 (2.54, 2.58, 2.25, 4.03, 4.80, 2.13, 4.62, 1.47, 1.58, 15.05 mg/mL respectively) were mixed with 0.75 mL of Folin–Ciocalteu reagent and allowed to stay at 22° C for 5 min. The mixture was supplied with 0.75 mL of NaHCO₃. Absorbance was measured at 725 nm by UV–VIS spectrophotometer (Varian Cary 50) after 90 min at

22 °C. A standard curve was calibrated by Gallic acid (0–100 mg/mL; $r > 0.99$). The results were represented as mg Gallic acid/(GA) g Dry weight.

Determination of total flavonoid content (TFC)

The flavonoid levels were measured by the aluminum chloride colorimetric method [22]. Briefly, 0.5 mL of extract solution with 1.5 mL of 95% ethanol, 0.1 mL of aluminum chloride 10%, 0.1 mL of 1 M potassium acetate were mixed with 2.8 mL of distilled water. The mixture vortexed for 10 s and left to stand at 25 °C for 30 min. The absorbance of the mixture was read at 415 nm. Quercetin concentrations (0 to 1200 µg/mL) were prepared and the linear fit was used for calibration of the standard curve.

EO Content

EO content of flowering branches was quantified using the method described by the European Pharmacopoeia for oil production [23]. Briefly, 20 g of dried aerial plant parts were subjected to hydro-distillation for 3 h using a Clevenger-type apparatus.

Data Analysis

The SAS software package for Windows (SAS, version 9.3, SAS Institute, Cary, NC) was used for data analysis. The mean values were compared with Duncan's multiple range tests. The data were statistically analysed at a 5% probability level ($P \leq 0.05$).

Results

Shoot and Root Dry Weight

Salinity and Si significantly ($P \leq 0.0.1$) affected shoot and root dry weight (Table 1). Shoot weight decreased by progressing the salinity. Under no foliar application of Si, severe salinity (200 mM NaCl) decreased shoot weight by 43% compared to non-stressed conditions. However, Si mitigated the salinity effects by improving the shoot weight. In severe salt stress, 200 mg/L Si increased shoot dry weight by 53% as compared to control plants (Fig. 1a). Root weight showed a little difference from shoot weight under salt stress. It was enhanced by slight salinity (50 mM NaCl) but decreased under 100 and 200 mM NaCl. In highly stressed and non-stresses plants, foliar application of Si significantly increased root weight (Fig. 1b).

Chlorophyll (Chl) and Relative Water Content (RWC)

Total Chl (Fig. 2a) and RWC (Fig. 2b) decreased by progressing the salinity; however, they were improved by advancing Si spraying particularly at severe stress conditions. Reduced Chl concentration in coriander leaves was observed in plants experiencing the saline conditions. 100 and 200 mg/L Si induced the positive

effects only in severe salt stress. RWC differed from 55% in 200 mM NaCl without Si application to 84.6% in non-saline condition with 100 mg/L Si.

Total Phenolic Content (TPC) and Total Flavonoid Content (TFC)

Salinity and foliar-applied Si significantly ($P \leq 0.0.1$) influenced TPC and TFC (Table 1). Fig. 3 represents the variability of TPC and TFC under salinity and foliar application of Si. Both salinity and Si application increased TPC and TFC. However, TPC and TFC demonstrated a reduced trend under severe saline stress compared to moderate stress treatment (100 mM). Although the highest amounts of TPC and TFC were obtained in plants under 100 mM NaCl, Si did not affect these traits significantly. A significant increase of TPC and TFC was observed in control plants, in which 100 mg/L Si resulted in a greater improvement.

Catalase (CAT) and Superoxide Dismutase (SOD)

CAT and SOD activities were significantly ($P \leq 0.0.1$) changed under salinity and Si application (Table 1). Fig. 4 shows the variation CAT (a) and SOD (b) in the presence of salinity and Si foliar application. The activity of these antioxidant enzymes increased by advancing the salinity levels. Under non-Si application, severe salinity enhanced the activity of CAT and SOD by 89 and 57%, respectively, compared to non-saline treatment. The role of Si was observed in the 100 and 200 mM NaCl as it decreased the activity of CAT and SOD. Accordingly, under severe stress conditions, Si alleviated the activity of CAT and SOD by 20 and 18% as compared to non-Si treatment.

Essential Oil (EO) Percentage and Yield

Salinity and foliar application of Si significantly ($P \leq 0.0.1$) changed EO percentage and EO yield (Table 1). EO percentage increased by salinity up to 100 mM NaCl (Fig. 5a). Besides, 100 mg/L Si was the most effective treatment of foliar application to obtain the optimum EO content. The maximum EO percentage (0.61%) was observed in leaves of plants experiencing moderate saline stress (100 mM NaCl) and Si application. The optimum EO yield was also obtained by the slight and moderate salt stresses with 100 mg/L Si (Fig. 5b)

Discussion

The adverse effects of salinity on growth were alleviated by Si application. These improvements were more effective with the level of 100 or 200 mg/L Si than other 50 mg/L. Alzahrani *et al.* [12] showed a significant reduction of dry matter of wheat under salinity, but Si tried to improve it. The increment in growth by Si corresponds to an improved rate of photosynthesis that is

related to leaf ultra-structure, leaf content of chlorophyll, and activity of ribulose biphosphate carboxylase [24]. Salt stress decreases the activity of cell physiology including photosynthesis, extremely because of the osmotic stress, nutritional imbalance, and toxicity in addition to the oxidative stress [25].

The decline of dry matter with progressing salinity could be attributed to inhibition of cell elongation and expansion, reduced turgor pressure, alteration of energy from growth to the synthesis of compatible solutes to maintain cell turgor, which results in reduced tissue water contents and trimming down the photo-assimilation and metabolites required for cell division [26]. The increase of root weight in slight stress can be due to the strategy of coriander plants that feel the slight stress as a signal for expanding their roots to provide the required nutrients and water.

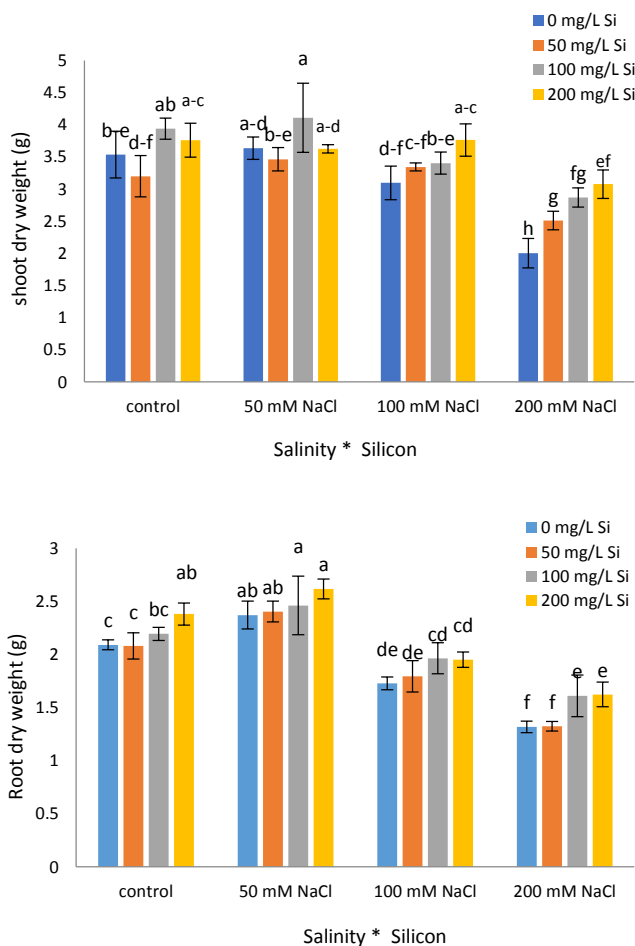


Fig. 1 The dry weight of shoot and root of coriander under salinity and silicon application. Values are means ± standard deviation (SD) of three replications (n= 3). Different letters show statistically significant differences among treatments at $P \leq 0.05$.

There was observed the decline in Chl content, which directly resulted in growth prohibition. Bybordi *et al.*,

[27] recorded reduced Chl content and biomass of onion when exposed to salt stress. High accumulation of Na^+ ions in plants drastically affects the plant physiological attributes like photosynthetic rate, transpiration rate, and stomatal opening and closing [28]. Reduction in photosynthesis and hindrance in the uptake of essential nutrients results in the scarcity of important metabolites under certain stresses [29, 30]. RWC is a good tool to assess water balance in plant leaves exposed to biotic and abiotic stresses [12].

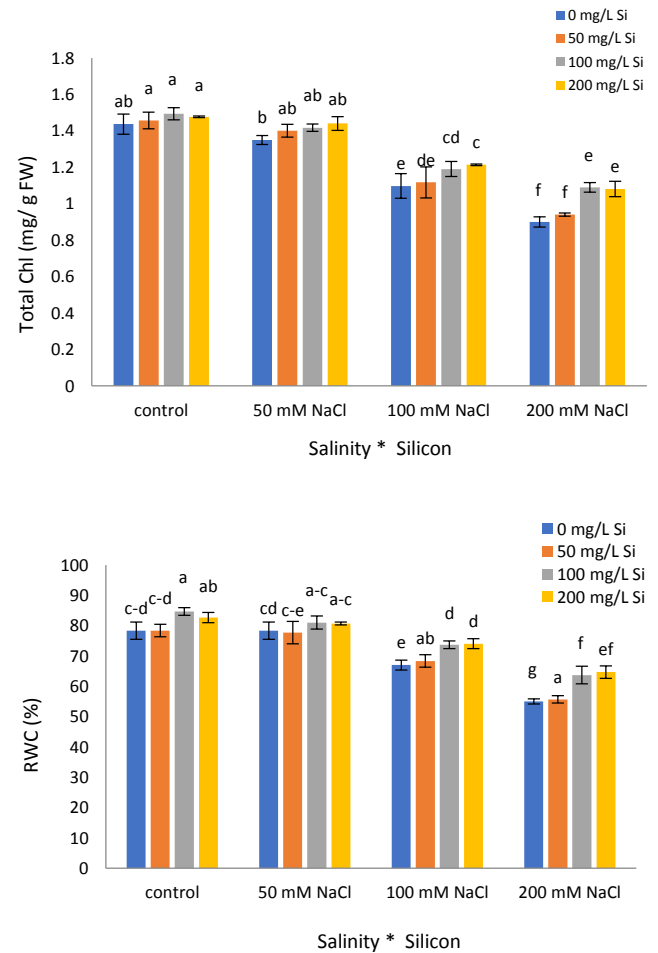


Fig. 2 Total chlorophyll (Chl) and relative water content (RWC) and of coriander under salinity and silicon application. Values are means ± standard deviation (SD) of three replications (n= 3). Different letters show statistically significant differences among treatments at $P \leq 0.05$. It also evaluates the water percentage present in a leaf as a fraction of the total volumetric water that can be held by a leaf at its full turgor. To maintain RWC in cells and tissues at a good status confers the metabolic activity to be continued by osmotic adjustments and other adaptations to salinity [1]. The reduced RWC under saline conditions have been reported in alfalfa [31] and olive plants [32].

Table 1 Analysis of variance for physiological and biochemical traits of coriander under salinity and silicon application

S.O.V.	df	MS									
		Shoot dry weight	Root dry weight	Total Chl.	RWC	CAT	SOD	TPC	TFC	EO content	EO yield
Salinity (S)	3	8.81**	2.21**	0.55**	1142**	0.07**	16.1**	225.4*	34.8**	0.06**	106**
Silicon (Si)	3	2.62**	0.19**	0.03**	134.5**	0.004**	1.14**	14.1**	0.98**	0.01**	77**
S* Si	9	1.69*	0.009 ^{ns}	0.003*	7.3 ^{ns}	0.001*	0.42**	1.94 ^{ns}	1*	0.003**	5.9*
Error	30	0.08	0.23	0.002	6.67	0.0007	0.07	2.42	0.4	0.007	2.6
CV.	-	8.41	7.7	4.06	3.54	8.79	5.1	4.9	6.3	5.68	10.1*

** , * , and ^{ns} show the significance at 1%, 5%, and no significance, respectively.

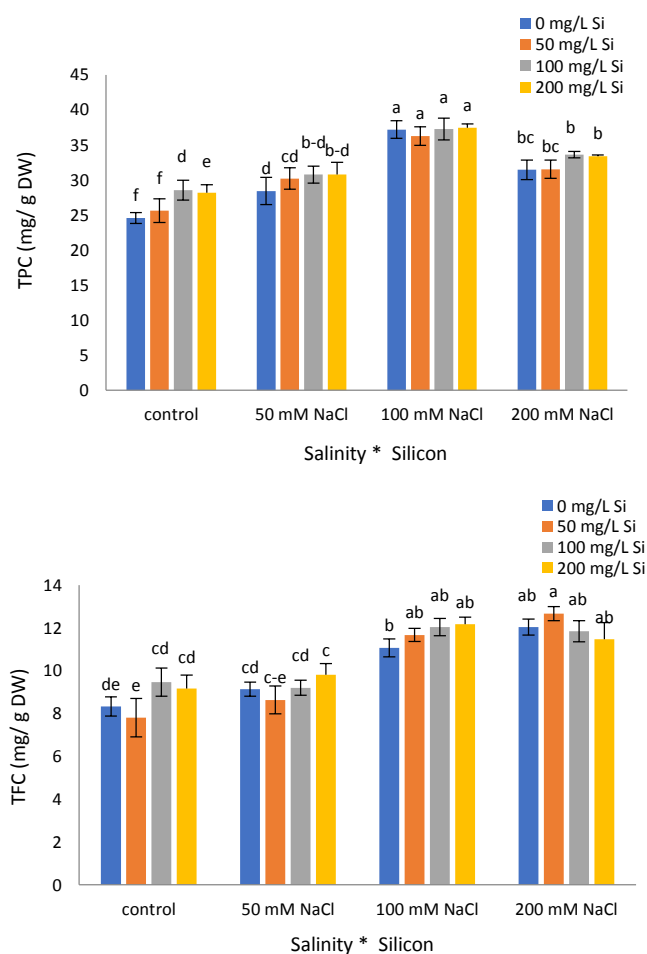


Fig. 3 Total phenolic content (TPC) and total flavonoid content (TFC) of coriander under salinity and silicon application. Values are means \pm standard deviation (SD) of three replications ($n=3$). Different letters show statistically significant differences among treatments at $P \leq 0.05$.

Here, salt stress led to reduced RWC, but Si supplementation could alleviate salt stress by a higher value of RWC. 100 or 200 mg/L Si had a positive effect on improving RWC. Si spraying significantly improved RWC by improving the growth and ability of plant roots to absorb more water from the growth medium and flow from roots to leaves adjusts the transpiration rate with

stomatal conductance and regulates plant water potential [33].

Coriander plants improved antioxidant capacity by enhancing enzyme activity. Under stress conditions, reactive oxygen species (ROS) increase in plants, in which its overproduction is toxic and causes damage to subcellular components and DNA which ultimately leads

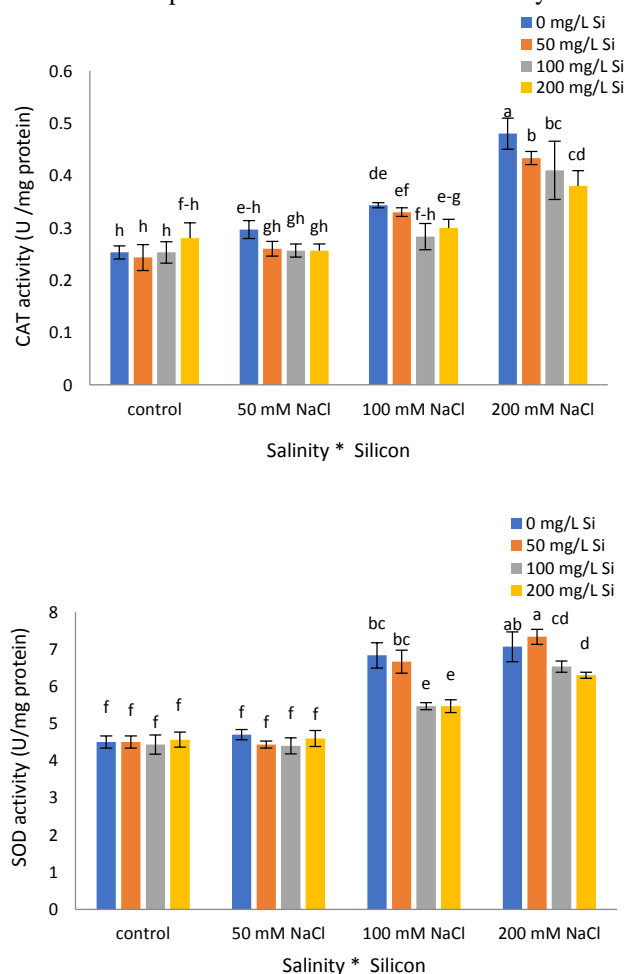


Fig. 4 Catalase (CAT) and Superoxide dismutase (SOD) of coriander under salinity and silicon application. Values are means \pm standard deviation (SD) of three replications ($n=3$). Different letters show statistically significant differences among treatments at $P \leq 0.05$.

to cell death [34]. Salinity altered the amount and activities of antioxidant enzymes in scavenging ROS [35] by efficient destruction caused by O^{2-} and H_2O_2 in chloroplast and cytoplasm of the plant cell [36]. Si has also been recognized to act as an antioxidant and alleviated the activities of CAT and SOD activities in the leaves of coriander plants. However, the increases of antioxidant enzymes under salinity stress have been reported in some studies [36,37].

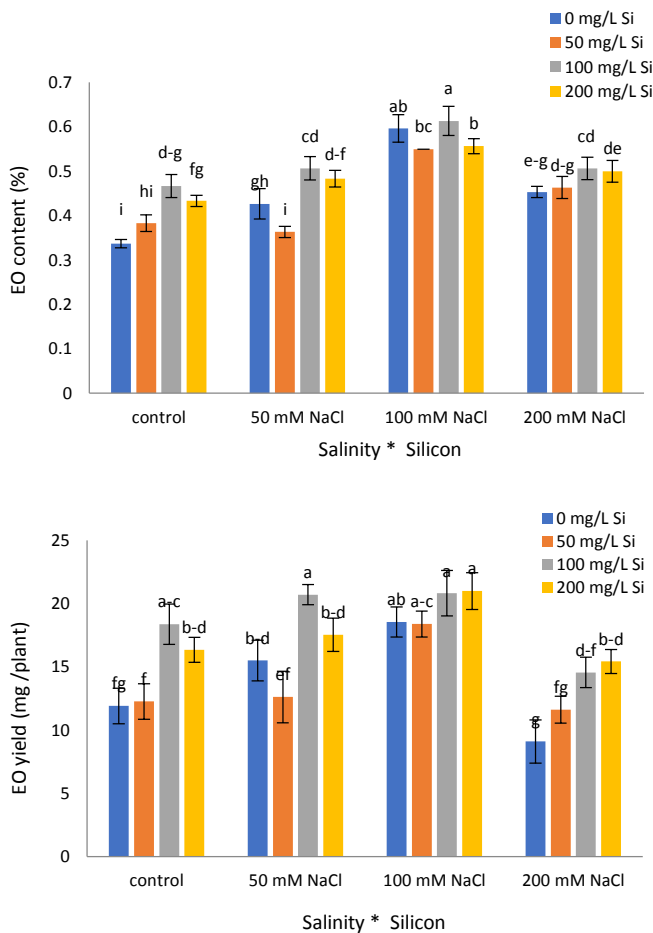


Fig. 5 Essential oil content and yield of coriander under salinity and silicon application. Values are means \pm standard deviation (SD) of three replications ($n=3$). Different letters show statistically significant differences among treatments at $P \leq 0.05$.

The phenolic compound is an eminent antioxidant factor for protecting the plants from dangerous radicals such as ROS. The higher TPC and TFC were observed under slight and moderate salt stress and with Si application, but they decreased in plants experiencing the severe drought. However high salinity decreased the phenolic contents. Therefore, it can be concluded that severe stress might inhibit the synthesis of TPC and TFC [38]. Plants use different mechanisms to distribute flavonoids among subcellular compartments [39]. The augmentation of TPC and TFC under moderate salinity and their reduction at severe stress conditions have been obtained in *Thymus vulgaris* L [40]. Phenolics and flavonoids are produced by shikimic acid or phenylpropanoid pathways, and hydroxyl groups can donate hydrogen and react with

ROS in the termination reaction, which ultimately breaks the cycle of producing new radicals. The shikimic acid pathway converts simple carbohydrate precursors into aromatic amino acids [39]. Si due to its role in improving the production of secondary metabolisms such as TPC and TFC can protect the stressful plants from the ROS. The positive role of Si on polyphenols has been observed on coriander plants grown in lead (Pb)-spiked soil [8].

Adverse impacts of severe salt stress were obtained on EO percentage and yield of coriander, but they were improved by Si. EO yield is calculated by dry matter and EO percentage. So the reduction of shoot biomass and EO percentage results in EO yield [41]. Low and medium concentrations of salinity (50 and 100 mM NaCl) increased EO yield but high saline reduced it (Fig.1). Si alleviated the undesirable effects of salt stress and promoted EO yield (Fig.1). Environmental stresses are important in EO production [42, 43]. Our results revealed that slight and moderate salt stress and Si enhanced EO yield, which confirmed the previous investigations with salinity [44] or Si application [43, 45]. The increased EO amount under saline conditions may be attributed to an enhancement of oil gland number and density [43]. Salt stress may affect EO production by changing the pathways of secondary metabolites production and their distribution [45]. This study shows the improvement of EO yield by Si application. Si plays an important role in elicitor-accelerated secondary metabolite production by inducing several transcriptional modifications [43]. Si may increase EO yield via improvement of cell development, ion uptake, and leaf oil gland density and size [46]. Different Si application methods proved to have positive impacts on EO production. EO yield with Si foliar spraying and significantly higher than that in salt-affected plants (Fig. 1). The improvement of EO yield under Si treatment in this work is in agreement with those reported on rosemary [44], geranium [41], and sweet basil [43]. Since carbon dioxide and glucose are the main precursors of EO production, there is a direct relationship between photosynthesis and EO biosynthesis [43]. Salt stress-induced injuries, such as oxidative stress and chlorophyll breakdown, result in reduced EO yield [44]. Si treatment increased EO production, and probably served as an effective stimulant to increase the production of secondary metabolites [13].

Conclusions

Soil or water salinity leads to desertification of large cultivated areas and significant economic losses in medicinal plants. When exposed to salt stress, coriander represented disrupted growth, reduced chlorophyll content, poor plant water relations, altered phenol and flavonoid, and high antioxidant enzyme activity. On the

other hand, a significant increase in plant biomass, chlorophyll and water contents, phenol and flavonoid concentrations was observed by exogenous application of Si, which alleviated the undesirable effects of salinity. 50 mg/L had no significant difference with control. Since both 100 and 200 mg/L Si showed no significant difference in growth and physiochemical attributes of coriander plants, 100 mg/L of Si spraying was selected to stimulate the plant growth and yield by changing physiochemical characteristics under medium and severe salinity.

Author Contribution

All authors contributed equally to this article.

Conflict of Interest

The authors declare that they have no conflict of interest.

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