Benefits of Silicon Nutrition on Growth, Physiological and Phytochemical Attributes of Basil upon Salinity Stress

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Abstract

In the present study, improvement of salt tolerance in basil (as a salt-sensitive plant) was investigated through silicon (Si) nutrition. Basil plants were subjected to silicon (0, 3 mM) and salinity (0, 50, 100, 150 and 200 mM NaCl) for a duration of one month. Salt stress significantly decreased the biomass of basil. Si supplement (3 mM) resulted in a considerable increase (averagely +135%) in the biomass of salinized plants. Salt stress significantly decreased photosynthetic pigments concentrations, but Si supplement improved total chlorophyll concentration (averagely up to +217% compared to salinized plants). This improvement in pigment concentrations also occurred for carotenoids content (+123%). Salinity increased lipid peroxidation and H_2O_2 level in the aerial parts of the basil plants, but Si decreased lipid peroxidation (-49.1%) and H_2O_2 content (-29%) under salinity condition. Results showed salinity (alone) or together with Si, increased the level of polyphenols and also the level of radical scavenging activities in the aerial parts of basil but this effect was much more in plants co-treated with Si and salinity. Si nutrition increased the activity of SOD, APX and GPX in response to salt stress, but it did not affect CAT activity. Overall, Si supplement could induce salt tolerance in basil plants by improving photosynthesis, membrane integrity, and detoxification of toxic radicals. Furthermore, silicon increased the medicinal properties of basil via elevating its antioxidant capacity under salt stress.

Keywords: *Ocimum basilicum;* Polyphenols; Radical scavenging activity; Salt tolerance.

Abbreviations: Ascorbate peroxidase (APX); Catalase (CAT); 1,1-diphenyl-2-picrylhydrozyl (DPPH); Gallic acid equivalent (GAE); Guaiacol peroxidase (GPX); Hydrogen peroxide (H_2O_2); Malondialdehyde (MDA); Nitroblue tetrazolium (NBT); Hydroxyl radical ($^\bullet OH$), Reactive oxygen species (ROS); Silicon (Si); Superoxide dismutase (SOD).

Introduction

Common basil (*Ocimum basilicum* L.), a medicinal plant belonging to Lamiaceae family, is a medicinal plant cultured all over the world. For basil cultivation, warm climate, adequate sunlight and enough water is needed. In pharmacy, this species is helpful because of its diuretic and stimulating attributes. Its other medicinal properties are carminative, galactogogue, stomachic and antispasmodic tonic and vermifugem. Besides, hot basil tea is useful to heal flatulence and dysentery (Özcan and Chalchat, 2002; Sajjadi, 2006). Additionally, antiviral and antimicrobial

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activity of this species is well-documented (Chiang et al., 2005). On the other hand, the essential oil of *O. basilicum* is utilized to formulate several perfumes and cosmetics (Onofrei et al., 2015).

Salinity is considered as one of the most serious threats limiting plant growth and crop productivity (Hashemi et al., 2010). Globally, over 800 million hectares which is nearly a third of all cultivated land and half of all irrigated land is affected by salt stress. It means that production of a third of the world's food is under the risk of salinity (Flowers, 2004; Tavakkoli et al., 2010). Moreover, secondary salinization is increasing due to irrigation with salty water or disregarding the soil drainage (Zhu and Gong, 2014). It is well-documented that salt stress confines plant growth and development causing serious morphological, physiological and biochemical modifications in plants (Among many: Gupta and Huang, 2014; Abbasi et al., 2015; Flowers and Colmer, 2015; Singh and Flowers, 2016; Flowers et al., 2018). Osmotic stress and ion toxicity are two main detrimental effects of salt stress that result in imbalances cellular ions and increasing ROS, such as singlet oxygen, superoxide anion, H_2O_2 and $^{\bullet}OH$, which leads to oxidative stress. ROS accumulation causes abnormal cellular metabolism and injures fundamental lipids, proteins and nucleic acids of plant cells. On the contrary, cellular ROS scavenging systems including enzymatic (e.g. SOD, CAT and peroxidases) and non-enzymatic antioxidant compounds (e.g. phenolic compounds and flavonoids) are components used to alleviate oxidative stress (Gill and Tuteja, 2010). Moreover, some ROS scavenging enzymes (such as cell wall bound peroxidases) involved in lignin biosynthesis (as an important polyphenol) are altered by salinity (Vaidyanathan et al., 2003; Gunes et al., 2007). It is revealed that plants with higher antioxidant properties have more protective activity against cancer development and chronic inflammatory diseases (Krishnaiah et al., 2010). Therefore,

finding approaches to improve salt tolerance as well as antioxidant capacity in crops or horticultural plants would be of great interest.

Si is the second most abundant element participates in about 28% of the earth's crust (Sommer et al., 2006). However, it does not contribute in normal plant growth and development as an essential element (Epstein, 1994; Hodson et al., 2005). It is revealed that Si has a mitigating effect on the deleterious effects of biotic (Bélanger et al., 2003; Côté-Beaulieu et al., 2009) and abiotic (for example: Ali et al., 2013; Zhu and Gong, 2004; Keller et al., 2015; Ahmed et al., 2016) stresses. There are many reports that showed the beneficial effects of Si on salt tolerance in many plant species such as: *Triticum aestivum* (Saqib et al., 2008; Tuna et al., 2008), *Hordeum vulgare* (Liang et al., 2005), Lycopersicon *esculentum* (Al-aghabary et al., 2004; Romero-Aranda et al. 2006), *Cucumis sativus* (Zhu et al., 2004), *Zea mays* (Moussa, 2006), *Medicago sativa* (Wang and Han, 2007), *Saccharum* officinarum (Ashraf et al., 2010), Brassica napus (Hashemi et al., 2010), *Glycine max* (Lee et al., 2010), *Portulaca oleracea* (Kafi and Rahimi, 2011), *Spartina densiflora* (Mateos-Naranjo et al., 2013), *Oryza sativa* (Shi et al., 2013) and *Vicia faba* (Shahzad et al., 2013). Si involves in salinity tolerance through different strategies such as: 1) limiting transpiration by accumulation in leaves (Matoh et al., 1986), 2) confining of Na^+ in the roots tissues via complex formation (Ahmad et al., 1992), 3) maintenance of membrane stability and chloroplasts ultrastructure (Moussa, 2006; Gill and Tuteja, 2010; Liang et al., 2015), 4) mounting of H^+-ATPase activity (Liang et al., 2007), 5) lessening of oxidative stress through stimulation of cellular antioxidant system (Eraslan et al., 2008; Abbas et al., 2015; Shi et al., 2016; Kim et al., 2017), 6) reducing the entrance of Na^+ by restraining apoplastic pathway in the roots (Yeo et al., 1999), and 7) alleviation of salt induced osmotic stress (Kafi and Rahimi, 2011; Coskun et al., 2016). For instance, it is

detected that application of supplementary Si in the irrigation water is frequently of interest of researchers as a low cost and not detrimental way to improve salinity impacts on plants. Therefore, the current research was carried out to inspect the function of Si in alleviating salt stress in basil and also to find out the related physiological mechanism, with special spectacle to alteration of its total phenolic content and antioxidant capacity as a base of its medicinal properties.

Materials and methods

Plant material and growth conditions

Seeds of common basil (*Ocimum basilicum* L. var. green) (Pakan-Bazr company, Esfahan, Iran) were sterilized with 70% ethanol for 2 min followed by washing with sterilized distilled water for several times. The polystyrene boxes composed of 50% perlite and 50% fine sand were used to sow basil seeds. Thirty-day-old plants of basil were subjected to the treatments consisted of 1) control (Hoagland's solution without Si and NaCl), 2) Hoagland's solution with four levels of NaCl added (50, 100, 150 and 200 mM), 3) Hoagland's solution with 3 mM Si, and 4) Hoagland's solution containing 3 mM Si along with 50, 100, 150 or 200 mM NaCl. The treatments (in three replicates) were applied for duration of 30 days under greenhouse conditions (33 °C-14 L/20 °C-10 D and 50-60% of relative humidity) at Shahrekord University, Shahrekord, Iran. At the end of experiments (when plants were 60-day-old), fresh and dry weights of basil plants were assessed. Dry weights were measured after drying plant parts at 70 °C until the materials reached a constant weight based on the method described by Gong et al. (2005).

Measurement of photosynthetic pigments

The concentrations of photosynthetic pigments (total chlorophyll and carotenoids) were detected according to the method of Lichtenthaler and Buschmann (2001) with 80% acetone as the solvent.

Chlorophyll a ($\mu g\ ml^{-1}$) $= 12.25A_{663} - 2.79A_{646}$

Chlorophyll b ($\mu g\ ml^{-1}$) $= 21.5A_{646} - 5.1A_{663}$

Total chlorophyll ($\mu g\ ml^{-1}$) $=$ chlorophyll (a + b)

Carotenoids $= [1000A_{470} - (1.82 \times$ Chlorophyll a) $- 85.02 \times$ Chlorophyll b)/198

where, A_{663}, A_{645}, and A_{470} represent absorbance values read at 663, 645 and 470 nm wavelengths, respectively.

Estimation of Lipid peroxidation

Lipid peroxidation was estimated according to the concentration of MDA (Ksouri et al., 2007). 250 mg fresh samples of shoots were homogenized in 0.1% trichlroacetic acid (TCA) followed by centrifuging at 10000×g for 10 min at 4°C. Then, 1 mL of the supernatant was mixed with 0.5% thiobarbituric acid (TBA) prepared in TCA 20%, and incubated at 95°C for 30 min. Subsequently, the tubes were placed in an ice bath to stop the reaction and then centrifuged at 10000×g for 5 min. The absorbance of supernatant was recorded at 532 nm and after subtracting the non-specific absorbance at 600 nm, MDA content was evaluated using the extinction coefficient of 155 $mM^{-1}\ cm^{-1}$.

Estimation of H_2O_2 content in the shoots

The concentration of H_2O_2 in the shoots was assessed by measuring the absorbance of titanium-hydroperoxide complex (Nag et al., 2000). The absorbance of orange-yellow H_2O_2-Ti complex was determined at 410 nm against blank. The content of H_2O_2 was detected via standard curve plotted with known concentrations (a range of 10-100 μM) of H_2O_2.

Polyphenol extraction and estimation

Sixty-day-old basil plants were shade dried and ground to fine powder. One g dry powder was extracted with 80% methanol with stirring for 30 min. The extract was then filtered through a Whatman filter paper and evaporated under vacuum. The Folin-Ciocalteu reagent was used to assay phenolic compound following Singleton's

method that slightly modified (Ksouri et al., 2007). Total phenolic concentration of plants (three replicates per treatment) was expressed as mg gallic acid equivalents (GAE) g^{-1} DW through a calibration curve with gallic acid: viz. $T = (C \times V)/M$ T is the total phenolic concentration in mg g^{-1} of the extracts as GAE, C is the concentration of gallic acid established from the calibration curve in mg ml^{-1}, V is the volume of the extract solution in ml and M is the weight of the extract in g.

DPPH radical-scavenging activity
The antioxidant activity of extracts, based on the scavenging activity of the stable 1,1-diphenyl-2-picrylhydrozyl (DPPH) free radical, was determined by the method described by Hanato et al. (1988). The antiradical activity was expressed as EC_{50} (µg/ml). The ability to scavenge the DPPH radical was calculated as: % Inhibition= $[(A_0-A_1)/A_0] \times 100$, where A_0 was the absorbance of the control and A_1 was the absorbance of extract/standard.

Assay of H_2O_2-scavenging activity
Measurement of hydrogen peroxide scavenging activity was based on the method described by Narwal et al. (2009). Evaluating the antioxidant activity of the extracts was based on EC_{50} (µg/ml). The ability to scavenge the superoxide anion radicals was calculated using the following formula: % Inhibition= $[(A_0- A_1)/A_0] \times 100$, where A_0 was the absorbance of the control and A_1 was the absorbance of the extract/standard.

Antioxidant Enzymes extraction and assay
Enzyme extraction procedure was accomplished according to the method of Chen et al. (2000) with some modification. All of the following operations were performed at 4°C. Fresh leaf samples (1 g) were ground in a mortar with liquid nitrogen and extracted in 100 mM Na-phosphate buffer (pH 6), containing 0.1 mM EDTA. The homogenate was centrifuged at 12000×g for 20 min. The supernatant was

transferred to Eppendorf tubes and kept in the -20 °C freezer. Total SOD activity was assayed in 100 mM potassium phosphate buffer, pH 7.5, 150 mM methionine L-methionine, 840 mM NBT, and 24 µM riboflavin by using the photochemical NBT method in terms of SOD's ability to inhibit reduction of NBT to form formazan by superoxide (Sairam et al. 2002). The photoreduction of NBT was measured at 560 nm. CAT activity was evaluated spectrophotometrically by determining the consumption of H_2O_2 ($\varepsilon= 39.4$ mM^{-1} cm^{-1}) at 240 nm in 50 mM phosphate buffer, pH 7.5 and 200 mM H_2O_2. Total APX activity was evaluated spectrophotometrically according to the method of Kato and Shimizu (1985) at 280 nm in 0.2 mM potassium phosphate buffer, pH 7.5, 15 mM ascorbic acid and 50 mM H_2O_2, as ascorbate ($\varepsilon= 2.8$ mM^{-1} cm^{-1}) was oxidized. GPX activity was assayed in 44 mM H_2O_2, and 45 mM guaiacol. The absorption at 470 nm was recorded and the activity was calculated using the extinction coefficient of 26.6 mM^{-1} cm^{-1} (Buchanan and Balmer, 2005). All enzyme activities were expressed as units per mg of protein. Protein content in all enzyme extracts was determined according to the method of Bradford (1976).

Statistical analysis and experimental design
The experiments were conducted in factorial and completely randomized design. The data was analyzed using the software SAS (V. 9.0) and the least significant difference (LSD) among treatments for each trait was calculated. P values less than 0.05 were considered to be statistically significant.

Results
The results of variance analysis for the studied physiological and biochemical characteristics of basil are shown in Table 1 and 2.

Results indicated that salt stress (at all applied degrees) significantly decreased the biomass of basil compared to control (p<0.05) (Fig. 1A). Increase in the level of

NaCl caused more decrease in the biomass. Si supplement (3 mM) resulted in a considerable increase in the biomass of salinized plants of basil (averagely 135% compared to saline condition alone). Si nutrition at 3 mM increased the dry weight of basil (+115%) at the absence of salinity stress (Fig. 1A). Salt stress significantly reduced the fresh weight of basil ($p<0.05$) (Fig. 1B), but Si nutrition significantly increased this parameter at all applied salinity levels.

Si addition increased total chlorophyll concentration significantly ($p<0.05$) in both normal and saline conditions (except at 200mM NaCl) (Fig. 2A). In non-stressed basil plants, Si (3 mM) increased total chlorophyll level by 130% compared to control. Under 50 mM NaCl, Si nutrition increased total chlorophyll level by 150% compared to exclusively salt stress at 50 mM. Nevertheless, the beneficial effect of Si was more remarkable at 100 and 150 mM NaCl, respectively 230% and 270%, compared to exclusively salt-stressed plants (at the mentioned saline conditions). But, Si did not improve the total chlorophyll level at 200 mM NaCl.

Table 1. Analysis of variance for the studied physiological and biochemical parameters in basil plants irrigated for 30 days with NaCl and/or Silicon

S.O.V	MS df	Dry weight	Fresh weight	Total chlorophyll	carotenoids	H_2O_2 content	MAD content	Total phenolic content
Salinity	4	0.24**	22.24**	0.06**	0.005**	7731723**	0.183**	106188574**
Silicon	1	0.11**	8.31**	0.15**	0.002**	4485334**	0.050**	65090319**
Silicon × Salinity	4	0.005**	0.35**	0.03**	0.0001*	216535**	0.004**	20863976**
Error	20	0.001	0.1	0.0001	0.00005	15148	0.0003	181285
CV (%)	-	5.47	6.9	4.53	8.76	2.69	5.24	2.02

* and **: Significance at 0.05 and 0.01 probability

Table 2. Analysis of variance for studied characteristics of antioxidative responses in basil plants irrigated for 30 days with NaCl and/or Silicon

S.O.V	MS df	DPPH scavenging	H_2O_2 scavenging	SOD activity	CAT activity	APX activity	GPX activity
Salinity	4	7346**	10596**	0.361**	0.777**	0.307**	0.012**
Silicon	1	2000**	5712**	0.197**	0.01[ns]	0.105**	0.002**
Silicon × Salinity	4	647**	325**	0.021**	0.006[ns]	0.009**	0.0006**
Error	20	4.9	40	0.0009	0.006	0.002	0.00006
CV (%)	-	3.15	4.50	2.9	4.70	6	4.1

ns, **: not significance and Significance at 0.01 probability
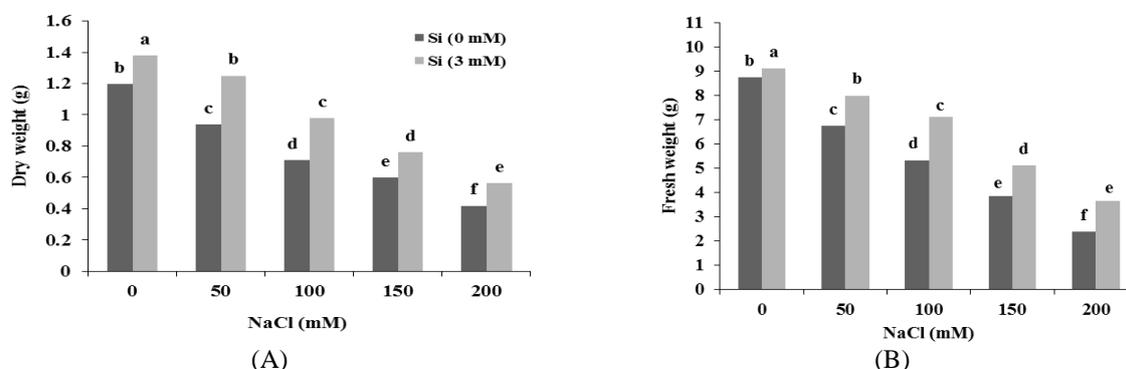

Fig. 1. Effects of different concentrations of NaCl (50, 100, 150 and 200 mM) and silicon (0 and 3 mM) on basil plants. (A) Dry weight, (B) Fresh weight. Means (three replicates) with the same letter are not significantly different at $p<0.05$.

At the absence of salinity, Si nutrition induced an increase of 310% in the carotenoids content of basil (Fig. 2B). Applied levels of NaCl (50, 100, 150 and 200 mM) resulted in decreasing carotenoids to less than 22.4, 38.3, 50.5 and 56.1%, respectively. Si nutrition (3 mM) increased significantly the carotenoids content at all utilized NaCl levels (p<0.05). This increase was 127, 125 and 118% at 50, 100, and 150 mM, respectively. At 200 mM NaCl, Si had no positive effect on the levels of carotenoids. The concentration of H_2O_2 in the salinized shoots increased in a NaCl concentration dependent manner. Accordingly, concentration of H_2O_2 increased by 15.7, 45, 60.5 and 73.7% at 50, 100, 150 and 200 mM NaCl, respectively (Fig. 2C). Si nutrition (3mM) brought about lessening of H_2O_2 in the salinized shoots at all utilized levels of NaCl. This decrease was recorded as 27.3, 29 and 19.7% at 50, 100 and 150 mM NaCl in comparison to the salinized shoots at the absent of Si. Si nutrition had no affirmative function to reduce H_2O_2 concentration at 200 mM NaCl.

Salinity caused a significant increase in lipid peroxidation in the aerial parts of basil in comparison to control (Fig. 2D). With increasing the level of NaCl (from 50 to 200 mM) in the medium of basil, lipid peroxidation increased by 53.2, 200 and 300%, respectively. Under salt stress, lipid peroxidation decreased by Si nutrition. This decrease was 49.1, 35 and 10.7 at 50, 100 and 150 mM NaCl, respectively. Furthermore, Si nutrition caused a decrease in lipid peroxidation of the non-stressed shoots by 42.7% compared to the control.
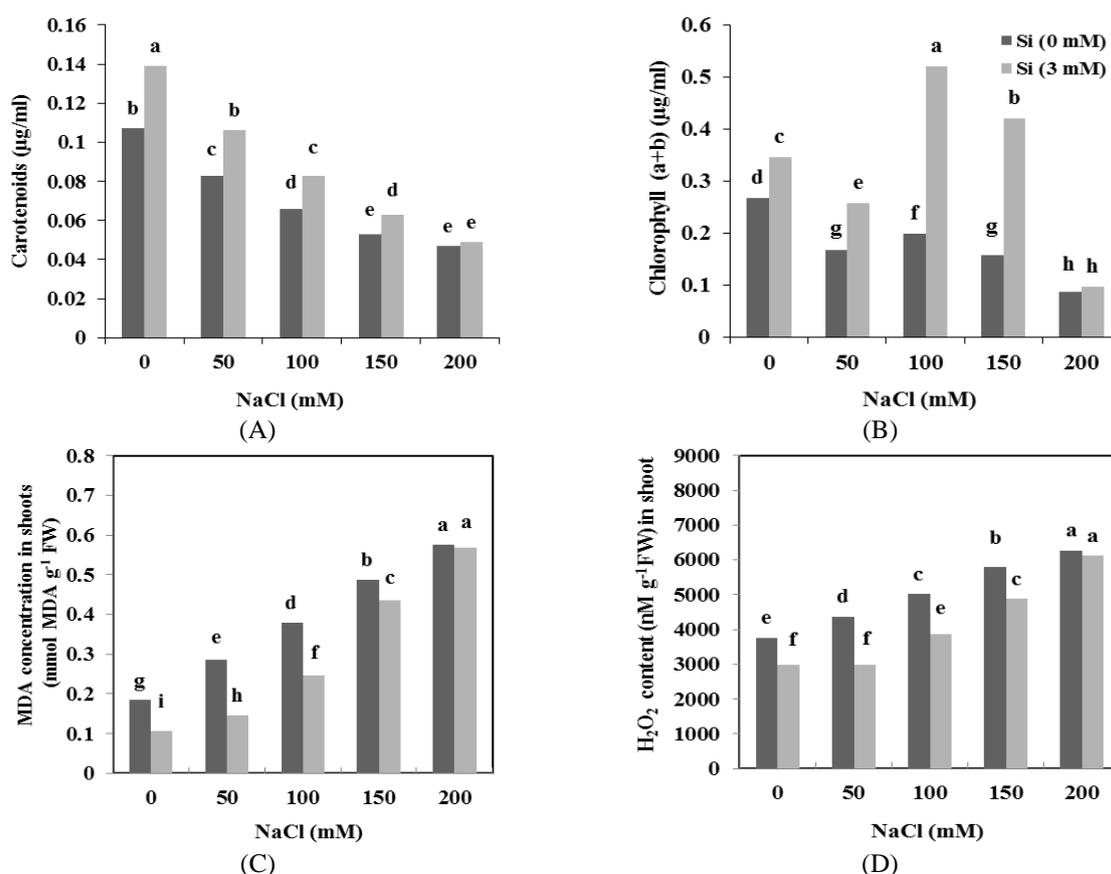

Fig. 2. Effects of different concentrations of NaCl (50, 100, 150 and 200 mM) and silicon (0 and 3 mM) on basil plants. (A) Total chlorophyll concentration, (B) Carotenoids concentration, (C) H_2O_2 level of the shoots, (D) MDA concentration. Means (three replicates) with the same letter are not significantly different at p<0.05.

Under salt stress and at the absence of Si, the highest amount of polyphenols (+66.2% compared to control) was obtained at 200 mM NaCl (Fig. 3A). Interestingly, Si nutrition caused a second buildup of polyphenols in the salinized shoots in comparison to salt stress alone. At this case, the highest increase (+40.2% compared to salinity alone) attributed to the salinized shoots of basil at 100 mM NaCl. Si nutrition (alone) had no significant effect on the concentration of polyphenols under neither 200 mM NaCl nor normal condition.

With increasing in the degree of salinity from 50 to 200 mM NaCl, the amount of EC_{50} for DPPH scavenging activity decreased significantly (Fig. 3B). Under salt stress, the most reduction (less than 66% compared to control) for this parameter was recorded for the salinized shoots of basil at 200 mM NaCl. Si nutrition caused to reduce EC_{50} for DPPH scavenging activity more than the case of salinity alone. The maximum level of DPPH scavenging ability was gained at Si nutrition under 150 mM NaCl.

The level of EC_{50} for H_2O_2-scavenging activity decreased through increasing the level of NaCl from 50 to 150 mM (Fig. 3C). The lowest level of this parameter was observed at 150 and 200 mM NaCl (Fig. 3C). Si nutrition significantly decreased the level of H_2O_2-scavenging activity in the salt-stressed plants in comparison to the saline condition alone. At this situation, the lowest level of EC_{50} was recorded at 150 mM NaCl. Under Si + NaCl condition, the level of H_2O_2-scavenging activity was relatively similar to those at 100 and 200 mM NaCl.
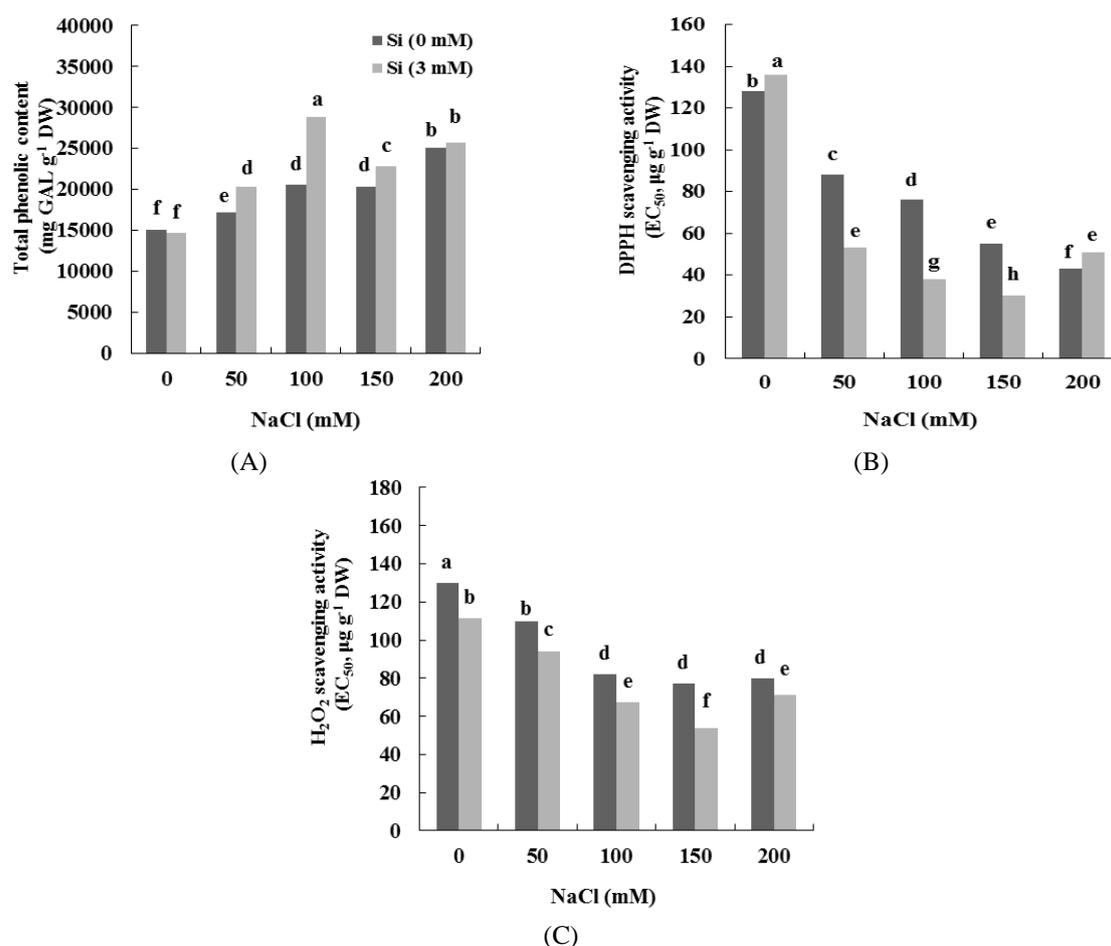

(A)

(B)

(C)

Fig. 3. Effects of different concentrations of NaCl (50, 100, 150 and 200 mM) and silicon (0 and 3 mM) on basil plants. (A) Total phenolic content, (B) DPPH-scavenging activity, (C) H_2O_2-scavenging activity. Means (three replicates) with the same letter are not significantly different at p<0.05.

Salt stress successively and significantly decreased the activity of SOD enzyme at all saline levels (50, 100, 150 and 200 mM NaCl) (Fig. 4A). Si nutrition (alone) increased the activity of this enzyme compared to the control. Also, the presence of Si (3 mM) in the saline medium (50, 100 and 150 mM NaCl) significantly decreased SOD activity in comparison to the simply salt-stressed plants. The lowest level of this activity was observed at 200 mM NaCl regardless of Si presence (Fig. 4A).

Results showed that Si nutrition did not alter CAT activity in the aerial parts of basil plants at non-stressed situation (Fig. 4B). Nevertheless, CAT activity decreased at 50, 100, 150 and 200 mM NaCl consecutively and this lessening was significant compared to control ($p<0.05$). Si nutrition did not affect the activity of this enzyme under salt stress condition (Fig. 4B).

Data analysis revealed that APX activity successively increased at 50, 100, 150 and 200 mM NaCl (Fig. 4C) ($p<0.05$). The uppermost level of APX activity was recorded at 200 mM NaCl. Si nutrition increased APX activity at both normal and saline culture solutions ($p<0.05$). Yet, at Si + NaCl condition, APX activity was significantly superior compared to the saline condition alone. Under Si nutrition, the maximum level of this activity was distinguished at 100 and 200 mM NaCl. At 200 mM NaCl along with Si, the activity of this enzyme was alike (Fig. 4C).

Salt stress enhanced GPX activity in the aerial parts of basil plants (Fig. 4D) ($p<0.05$). The highest level of GPX activity was monitored at 200 mM NaCl. Si nutrition did not modify GPX activity significantly at normal culture solution. Nonetheless, the activity of this enzyme increased in higher levels of NaCl (50, 100, 150 mM) under Si nutrition ($p<0.05$). The maximum activity of GPX was recorded at Si + 150 mM NaCl.
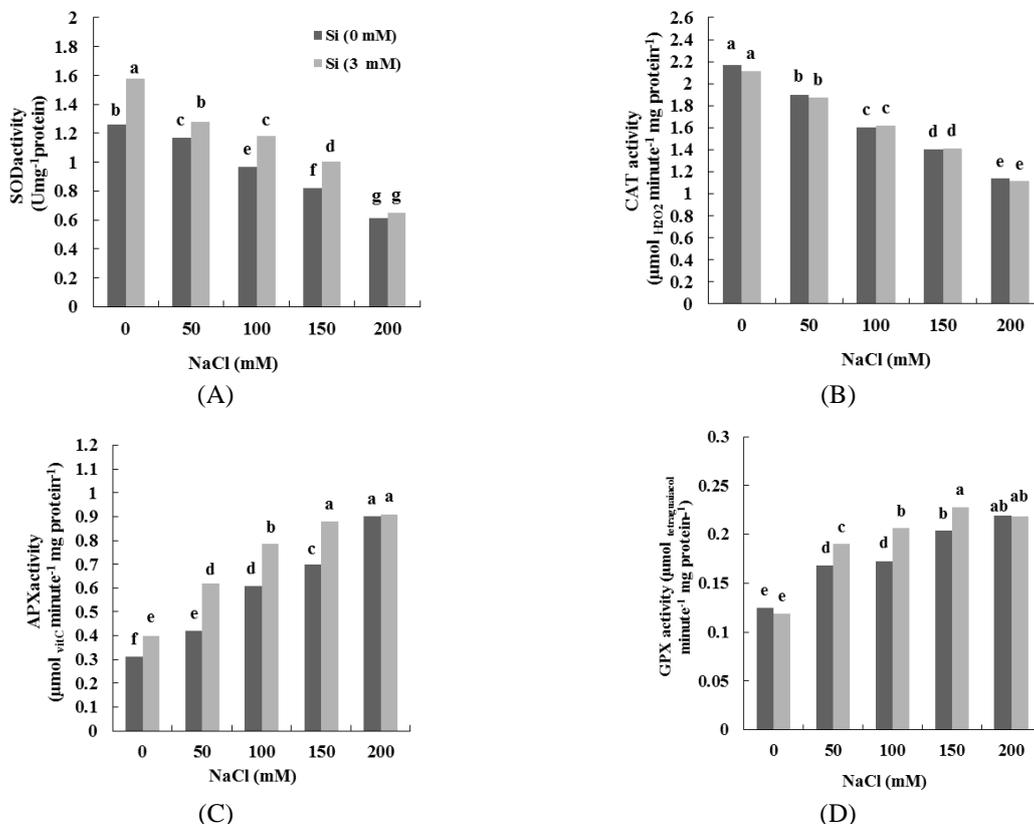

Fig. 4. Effects of different concentrations of NaCl (50, 100, 150 and 200 mM) and silicon (0 and 3 mM) on basil plants. (A) SOD activity, (B) CAT activity, (C) APX activity, and (D) GPX activity of the shoots. Means (three replicates) with the same letter are not significantly different at $p<0.05$.

Discussion

The buildup of NaCl in plant tissues negatively affects plant growth by inducing osmotic stress and ion toxicity (Munns and Tester, 2008). Additionally, the absorption and translocation of essential elements are susceptible by salinity causing oxidative stress in plants (Qiu et al., 2011; Abbasi et al., 2015). Therefore, reduction of deleterious effect of salt stress is crucial to reserve the quality and quantity of plant yield and production.

In the current work, Si utilization significantly alleviated the salt-induced biomass reduction in basil plants. This finding was in agreement with the previous studies (see Rizwan et al., 2015). In fact, various mechanisms could be involved to improve plant biomass by Si nutrition under salinity. As proposed, Si application can recover water status and water-use efficiency in plants (Coskun et al., 2016). Current data showed that the fresh weight decreased by salt stress in basil plants, whereas Si improved this trait in both stressed and non-stressed conditions. It is already reported (for example: in sorghum) that the root and whole-plant hydraulic conductance, transpiration, stomatal conductance, and leaf water content are improved by Si under osmotic stress. As suggested, the role of Si on augmentation of root hydraulic conductance is associated with increased expression and activity of plasma-membrane intrinsic protein (PIP) aquaporins, which are involved in water transport. Nevertheless, the related mechanisms have yet to be determined. Moreover, Si can adjust the osmotic potential of cells through increased osmolytes (e.g. proline, soluble sugars inorganic ions, etc.; Sonobe et al., 2010) accumulation and consequently affect water transport. On the other hand, Si-induced reductions in oxidative stress and membrane damage can positively improve root hydraulic conductance (Shi et al., 2016). Furthermore, the binding of Si with cell-wall hemicelluloses results in improved structural stability (He et al., 2015), which is obviously valuable under water deficit. As well- documented, Si can strengthen plant cell walls and contributes cell mechanical support through enhancing suberization, lignification, and silicification (Guerriero et al., 2016).

Salt-stressed plants usually experience a water deficit that leads to oxidative stress, which is overproduction of ROS such as H_2O_2, $^{\bullet}OH$, superoxide anion and singlet oxygen. These toxic radicals disrupt normal metabolism and damage plant cell plasma membrane and endomembrane systems (Gill and Tuteja, 2010). However, plants have enzymatic or non-enzymatic constituents, known as antioxidant defense systems, to scavenge ROS. Reports have revealed that Si decreases ROS (such as H_2O_2) levels in plant tissues through enhancing the activities of antioxidant enzymes such as SOD, peroxidases, CAT as well as glutathione reductase activities (among many: Eraslan et al., 2008; Ali et al., 2013, Abbas et al., 2015; Muneer and Jeong, 2015; Shi et al., 2016). The current results were in agreement with the previous reports. Apparently, Si could lessen oxidative stress in salt-stressed basil plants via regulation of antioxidant enzymes activities and non-enzymatic defense constituents to scavenge ROS.

Lipid peroxidation generated by ROS is another major detrimental effect of salt toxicity in higher plants (Gill and Tuteja, 2010). As already reported, Si diminishes the concentration of MDA, the end-product of lipid peroxidation, in salt-stressed barley (Liang et al. 2005), maize (Moussa 2006), grapevine rootstock (Soylemezoglu et al., 2009) and pea plants (Shahid et al., 2015), and as expected it would help to maintain membrane integrity and decrease membrane permeability (Liang et al., 2015). Khoshgoftarmanesh et al. (2014) reported that MDA concentration was positively correlated with Na^+ uptake in salt-stressed cucumber, whereas its level reduced with Ca^{+2} and K^+ uptake, and also

with Si supply. The role of Si in this response is still indistinct, but one elucidation is that, Si supply improves stabilized membranes which consequently lead to symplastic $[Na^+]$ reductions and $[K^+]$ and $[Ca^{2+}]$ accumulation. However, Liang et al. (2007) realized that Si does not affect membrane fluidity and H^+-ATPase activity *in vitro* in non-salinized plants. Then, they proposed the effect of Si on membrane fluidity and enzyme activity could be indirectly or secondarily.

Photosynthesis, as one of the most fundamental biochemical processes to increase plant biomass, can be simply injured by salt stress. NaCl overdose in plant tissue damages chloroplast ultrastructure and reduces the level of photosynthetic pigments. As the current results showed and in coordinate with the prior reports (Tuna et al., 2008; Mateos-Naranjo et al., 2013; Yin et al., 2014), Si addition can improve photosynthetic machinery (for instance: increasing the level of chlorophylls and carotenoids) under saline condition. As suggested, the beneficial effects of Si on the photochemical apparatus and photosynthetic pigments is partly attributed to the Si-mediated decrease in Na^+ uptake and increase in K^+ uptake in plants tissues under salt stress (Liang, 2007). Obviously, Si application has additional benefits on gas exchange characteristics, water potential, and reduction in oxidative stress in plant, which all may positively affect the level of photosynthetic pigments.

In the present study, changes of total phenolic content and antioxidant capacity of the shoots of basil were assessed under salinity and Si nutrition. To evaluate antioxidant properties of basil, the assessment of DPPH-radical and H_2O_2 scavenging activities were employed. Si nutrition increased extensively radical scavenging activity of the salinized shoots of basil. As recognized, plants containing high levels of polyphenols and radical scavenging activity are strong in antioxidant properties and would be valuable for human health (Krishnaiah et al., 2010). Hence, finding treatments to increase plant tolerance to environmental stresses such as salinity as well as increase in antioxidant properties of plant tissues could be useful in the agriculture and food industry. The current results illustrated that Si noticeably increased total phenolic content in the salinized shoots of basil. As the hydroxyl groups of phenolic compounds make them effective hydrogen donors, thus a highly positive relationship between total phenols and antioxidant activity has been reported in many plant species (Vinson et al., 1998). Phenolic acids and flavonoids are major phenolics that possess antioxidant activity and are widely found in the plant kingdom particularly in fruits and vegetables. The use of vegetables with high antioxidant properties is associated with prevention and/or lessening of the risk of diseases associated to free radical reactions (Katalinic et al., 2010).

Conclusions

In general, it could be concluded that Si nutrition alleviates detrimental effects of NaCl and enhances the growth of basil plants. According to the current data, this ameliorative effect of Si might be due to different mechanisms including enhancement of plant antioxidative systems to reduce oxidative stress, maintenance of membrane integrity and decrease permeability. Moreover, Si elevated medicinal properties of basil under salinity.

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