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***Original Research Article***

# Effect of Silicon on Some Physiological Properties of Maize (*Zea mays*) under Salt Stress

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## ABSTRACT

**Salt stress is an abiotic stress that can effect the plant growth and physiological and biochemical activities such as photosynthesis activity and chlorophyll content. This study was conducted in order to evaluate effects of silicon (Si) on some physiological properties of maize (*Zea mays*) under salt stress. Selected seedlings were transplanted to plastic pots contained sterilized and non-saline sandy soil that continuously aerated full-strength Hoagland nutrient solution. The Si was added at four levels (0, 2, 4 and 6 mmol.L$^{-1}$) from source potassium silicate ($K_2SiO_3$) and salt stress was applied at four levels (0, 3, 6 and 9 dS.m$^{-1}$) from source sodium chloride (NaCl). A factorial experiment in a completely randomized design (CRD) with sixteen treatments and three replications was applied. Results indicated that salinity significantly decreased the fresh and dry weights of shoot and root, stem length, leaf area, chlorophyll content and relative water content (RWC) of maize plant and application of Si significantly increased them. Remarkable decrease observed in treatments with EC > 3 dS.m$^{-1}$, while, gradual increasing of Si increased physiological properties of maize. Therefore, proper Si nutrition can increase salt resistance in maize plant.**

**Key Words: Chlorophyll, leaf area, plant yield, relative water content, saline soils**

## INTRODUCTION

It is estimated that about 20% of the irrigated land in the present world is affected by salinity that is exclusively classified as arid and desert lands comprising 25% of the total land of our planet (Rasool *et al.* 2013). Salt stress creates both ionic as well as osmotic stress on plants (Parvaiz and Satyawati 2008). These stresses can be distinguished at several levels such as shoot, root and tissues (Tester and Davenport 2003). Salt stress is an abiotic stress that can effect the plant growth and physiological and biochemical activities such as photosynthesis activity and chlorophyll content (Hajer *et al.* 2006; Saleh 2012). Leaf chlorophyll under salt stress damage and it cause of decreasing of photosynthesis (Turan *et al.* 2009). Hung and Redman have mentioned to reduction of chlorophyll content in leaves of barely plants in salinity conditions (Hung and Redmann 1995). Osmotic stress is caused due to the excess of $Na^+$ and $Cl^-$ in the environment that decrease the osmotic potential of the soil solution and hence water uptake by plant root (Rasool *et al.* 2013).

Salinity can be minimized with water and drainage, but the cost of engineering and management is very high (Tuna *et al.* 2008). Soil salinity and sodicity can also be alleviated using chemical amendments. Silicon (Si) is the second most abundant element in the lithosphere, accounting for about 26% by weight (Snyder *et al.* 2007). Si is generally considered a beneficial element for the growth of higher plants and most of the Si taken up by plants is deposited on cell walls (Epstein 1999; Liang *et al.* 2005). Si accumulates in the leaf, forming a doubled layer (Pereira *et al.* 2013). This accumulation promotes a reduction in transpiration and decrease water loss by the maize plant (Freitas *et al.* 2011).

In this study, we hypothesized that Si nutrition reduces the destructive effects of salinity and improves the growth of maize plant. Therefore, this study aimed to investigate the effects of Si, salinity and interaction between Si nutrition and salinity in maize, and it evaluated the physiological properties such as yield, stem length, leaf area, chlorophyll content and relative water content (RWC).

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## MATERIALS AND METHODS

***Greenhouse condition, pot and laboratory experiments***

This study was carried out at greenhouse condition in Shahid Chamran university of Ahvaz (natural light, daily photoperiod 12 h, day average temperature 24°C, night average temperature 18°C and mean relative humidity 70 ± 5% in experiment duration). Seeds of maize (*Zea mays* cultivar 704) were obtained from Seed and Plant Improvement Institute (SPII), Karaj, Iran. The seeds were sterilized in hypochlorite solution (1%) for 5 min and washed with distilled water. Seeds of maize were germinated for 5 day in the dark at 24°C on sheets of filter paper wetted with 0.4 mmol.$L^{-1}$ $CaC1_2$. Afterwards, selected seedlings of equal size and vigor were transplanted to plastic pots contained sterilized and non-saline sandy soil (in depth of 2 cm). Plastic pots having 22cm height and 20 cm opening mouth diameter were selected. The pot contained 1000 mL of continuously aerated full-strength Hoagland nutrient solution (Hoagland and Arnon, 1950) which was renewed every other day. The pH of the nutrient solution was adjusted to 5.8 daily using 0.01 mol.$L^{-1}$ $H_2SO_4$ and / or KOH. Salinity and Si treatments were applied by adding sodium chloride (NaCl) and potassium silicate ($K_2SiO_3$) to the nutrient solution immediately after 10 day old seedling were transplanted to the nutrient solution. The initial pH of the nutrient solution after the addition of potassium silicate was 7.6, which was adjusted to 5.8 using 0.01 mol.$L^{-1}$ $H_2SO_4$ before transplanting. All plants were harvested two month (8 weeks) after treatments and separated into shoot and root after sterilized using deionized water and were immediately recorded. Afterwards, samples at 70 °C for 48 h, in a forced-air oven were dried and dry weight was determined. Fresh and dry weights of shoot and root recorded using electronic precision balance (0.001g).

Leaf area was measured with the help of area meter (model CI-202) by averaging the value taken from three plant samples. Leaf chlorophyll content was measured (according to SPAD units) using a hand-held chlorophyll content meter (SPAD-502, Konika Minolta, Japan). At each evaluation the content was repeated 5 times from leaf tip to base and the average was used for analysis.

The RWC of leaves was determined in the fully expanded topmost leaf of the main shoot (Pirzad *et al.* 2011). The fresh weight of the sample leaves was recorded and the leaves were immersed in distilled water in a Petri dish. After 2 h, the leaves were removed, the surface water was blotted-off and the turgid weight recorded. Samples were then dried in an oven at 70°C to constant weight. Leaf RWC was calculated using the following formula (Turner, 1981):

$$RWC (\%) = [(F.W – D.W) / (T.W – D.W)] \times 100$$

Where: F.W., Fresh weight; D.W., Dry weight; T.W., Turgid weight.

***Statistical analysis***

The experiment was performed as a 4×4 factorial experiment in a completely randomized design (CRD), with three replications. Sixteen treatments were in this study and one treatment had three replications. The Si was added at four levels (0, 2, 4 and 6 mmol.$L^{-1}$) and salinity was applied at four levels (0, 3, 6 and 9 dS.$m^{-1}$). Statistical analysis of data including normality test, analysis of variance (ANOVA), and comparisons of means was performed using SAS program (SAS institute, 2002). The comparison of means was carried out using Tukey's tests at $P < 0.05$.

## RESULTS

***Fresh and dry weights of shoot and root of maize plant***

The result of analysis of variance (ANOVA) for the major effect of salinity and Si on fresh and dry weights of shoot and root of maize plant showed that the salt stress had significant effect ($p<0.01$) on all fresh and dry weights of shoot and root (Table 1). The Si had significant effect ($p<0.01$) on fresh weight of shoot, dry weights of shoot and root, and significant effect ($p<0.05$) on fresh weight of root (Table 1). Furthermore, interaction effect (salinity × Si) significantly ($p<0.01$) influenced fresh and dry weights of shoot and root (Table 1). Salinity significantly decreased all fresh and dry weights of shoot and root, while The Si significantly increased fresh and dry weights of shoot and root (Table 1). The simultaneous application of salinity and Si showed that maximum and minimum contents for fresh weight of shoot (Figure 1a), fresh weight of root (Figure 1b), dry weight of shoot (Figure 1c) and dry weight of root (Figure 1d) observed in $E_1S_4$ (EC= 0 $dS.m^{-1}$ and Si= 6 $mmol.L^{-1}$) and $E_4S_1$ (EC= 9 $dS.m^{-1}$ and Si= 0 $mmol.L^{-1}$) treatments, respectively. As shown in Figure 1, a remarkable decrease observed in treatments with EC > 3 $dS.m^{-1}$, while, gradual increasing of Si increased fresh and dry weights of shoot and root. Therefore, there are not significant different in EC= 0 $dS.m^{-1}$ and 3 $dS.m^{-1}$ treatments. Positive effect of Si on plant yield in conditions that plant grew under salt stress was more observable in comparison to conditions that plant grew under normal conditions.

**Table 1.** Mean comparison and analysis of variance effects salinity and silicon on fresh and dry weights of shoot and root of maize plant.

| S.O.V | | Fresh weight of shoot (g) | Fresh weight of root (g) | Dry weight of shoot (g) | Dry weight of root (g) |
|---|---|---|---|---|---|
| Salinity ($dS.m^{-1}$) | | | | | |
| | 0 | 29.92 a | 14.86 a | 4.23 a | 2.26 a |
| | 3 | 29.69 a | 14.89 a | 4.29 a | 2.23 a |
| | 6 | 23.52 b | 12.39 b | 3.01 b | 1.93 b |
| | 9 | 18.55 b | 9.13 c | 1.78 c | 1.55 b |
| Silicon ($mmol.L^{-1}$) | | | | | |
| | 0 | 23.58 b | 11.93 c | 2.85 c | 1.69 c |
| | 2 | 25.25 a | 12.41 b | 3.36 b | 1.96 b |
| | 4 | 26.45 a | 13.48 a | 3.58 a | 2.16 a |
| | 6 | 26.40 a | 13.46 a | 3.52 a | 2.17 a |
| | df | Significance level | | | |
| Salinity | 3 | ** | ** | ** | ** |
| Silicon | 3 | ** | * | ** | ** |
| Salinity × Silicon | 9 | ** | ** | ** | ** |
| Error | 32 | 0.86 | 0.11 | 0.01 | 0.03 |

* and ** significant at level of 5 and 1%, respectively, Means, in each column, with similar letters are not significantly different at the 5% probability level using Tukey's test.

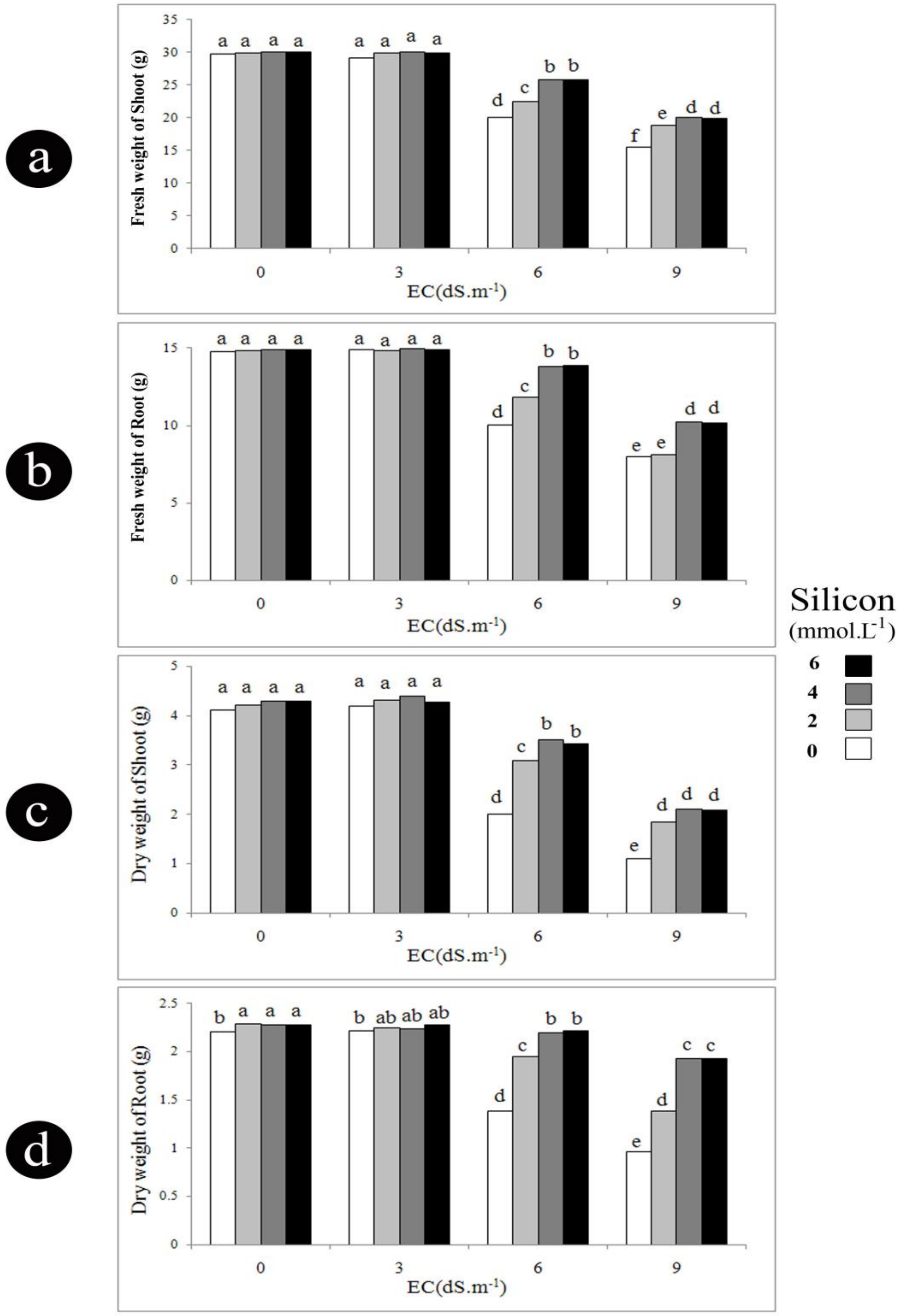


**Figure 1.** Effects of salt stress and silicon on **a**- fresh weight of shoot, **b**- fresh weight of root, **c**- dry weight of shoot, **d**- dry weight of root of maize plant. Means, in each box, with similar letters are not significantly different at the 5% probability level using Tukey's test.

***Stem length***

The result of ANOVA for the major effect of salinity and Si on stem length showed that the salinity had significant effect ($p<0.01$ and $p<0.05$) on stem length (Table 2). Interaction effect (salinity × Si) significantly ($p<0.01$) influenced stem length (Table 2). Salinity significantly decreased stem length, while the Si significantly increased stem length (Table 2). The simultaneous application of salinity and Si showed that maximum and minimum values for stem length (Figure 2a) observed in $E_2S_3$ (EC= 3 $dS.m^{-1}$ and Si= 4 $mmol.L^{-1}$) and $E_4S_1$ (EC= 9 $dS.m^{-1}$ and Si= 0 $mmol.L^{-1}$) treatments, respectively.

**Table 2.** Mean comparison and analysis of variance effects salinity and silicon on stem length, leaf area chlorophyll content and relative water content (RWC) of maize.

| S.O.V | | Stem length (cm) | Leaf area ($cm^2$) | Chlorophyll content (SPAD) | Relative Water Content (RWC) (%) |
|---|---|---|---|---|---|
| Salinity ($dS.m^{-1}$) | | | | | |
| | 0 | 58.58 a | 135.14 a | 39.37 a | 90.50 a |
| | 3 | 59.00 a | 136.78 a | 38.92 a | 84.75 b |
| | 6 | 51.17 b | 116.50 b | 36.17 a | 80.25 b |
| | 9 | 43.67 c | 100.67 c | 30.83 b | 73.25 c |
| Silicon ($mmol.L^{-1}$) | | | | | |
| | 0 | 50.08 b | 109.50 c | 34.12 b | 75.25 c |
| | 2 | 52.16 ab | 121.34 b | 35.79 b | 81.00 b |
| | 4 | 55.75 a | 129.78 a | 38.34 a | 86.00 a |
| | 6 | 54.42 a | 128.47 a | 37.04 a | 86.50 a |
| | df | Significance level | | | |
| Salinity | 3 | ** | ** | ** | ** |
| Silicon | 3 | * | ** | ** | * |
| Salinity × Silicon | 9 | ** | ** | ** | ** |
| Error | 32 | 0.33 | 1.73 | 0.10 | 2.35 |

* and ** significant at level of 5 and 1%, respectively, Means, in each column, with similar letters are not significantly different at the 5% probability level using Tukey's test.

***Leaf area***

The result of ANOVA for the major effect of salinity and Si on leaf area showed that the salinity had significant effect ($p<0.01$) on stem length (Table 2). Interaction effect (salinity × Si) significantly ($p<0.01$) influenced stem length (Table 2). Salinity significantly decreased leaf area, while the Si significantly increased leaf area (Table 2). The simultaneous application of salinity and Si showed that maximum and minimum values for leaf area (Figure 2b) observed in $E_2S_3$ (EC= 3 $dS.m^{-1}$ and Si= 4 $mmol.L^{-1}$) and $E_4S_1$ (EC= 9 $dS.m^{-1}$ and Si= 0 $mmol.L^{-1}$) treatments, respectively. Overall, in whole salinity levels, the best of performance among these treatments observed in treatments that it had received 4 $mmol.L^{-1}$ of Si.

***Chlorophyll content***

The result of ANOVA for the major effect of salinity and Si on leaf area showed that the salinity had significant effect ($p<0.01$) on chlorophyll content (Table 2). Interaction effect (salinity × Si) significantly ($p<0.01$) influenced chlorophyll content (Table 2). Salinity significantly decreased chlorophyll content, while the Si significantly increased chlorophyll content (Table 2). The simultaneous application of salinity and Si showed that maximum and minimum contents for chlorophyll content (Figure 2c) observed in $E_2S_3$ (EC= 3 $dS.m^{-1}$ and Si= 4 $mmol.L^{-1}$) and $E_4S_1$ (EC= 9 $dS.m^{-1}$ and Si= 0 $mmol.L^{-1}$) treatments, respectively.

***Relative water content (RWC)***

The result of ANOVA for the major effect of salinity and Si on leaf area showed that the salinity had significant effect ($p<0.01$ and $p<0.05$) on RWC (Table 2). Interaction effect (salinity × Si) significantly ($p<0.01$) influenced RWC (Table 2). Salinity significantly decreased RWC, while the Si significantly increased RWC (Table 2). The simultaneous application of salinity and Si showed that maximum and minimum values for RWC (Figure 2d) observed in $E_1S_4$ (EC= 0 $dS.m^{-1}$ and Si= 6 $mmol.L^{-1}$) and $E_4S_1$ (EC= 9 $dS.m^{-1}$ and Si= 0 $mmol.L^{-1}$) treatments, respectively.

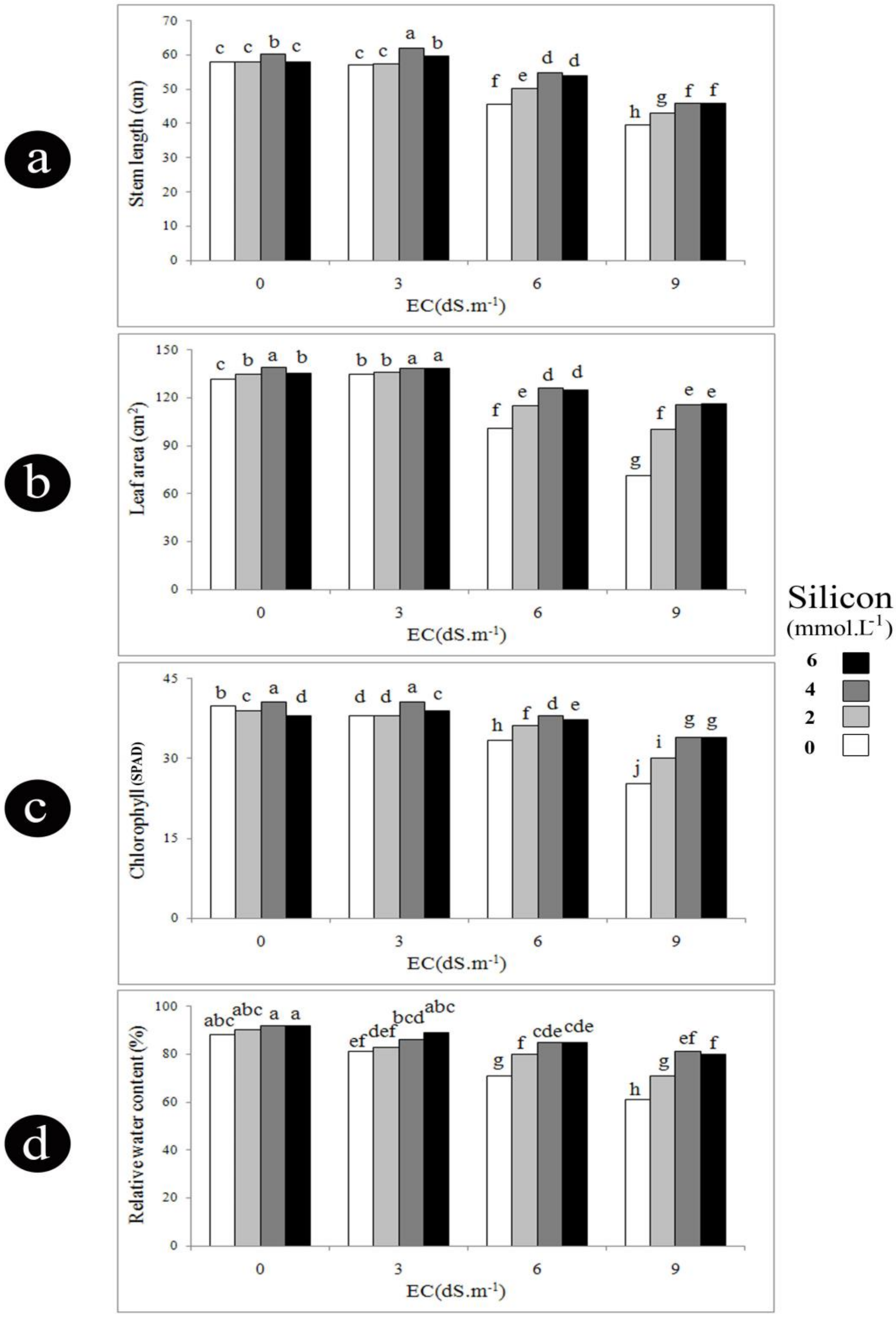


**Figure 2.** Effects of salt stress and silicon on **a-** stem length, **b-** leaf area, **c-** chlorophyll content, **d-** relative water content (RWC) of maize plant. Means, in each box, with similar letters are not significantly different at the 5% probability level using Tukey's test.

## DISCUSSION

The results of this study indicated that salt stress decreased all physiological properties of maize. Effects of salinity (decreasing) and Si (increasing) on fresh and dry weights of shoot and root of maize was similar. The root and shoot growth reduces abruptly in salt sensitive plants (Parvaiz and Satyawati 2008). Giaveno *et al.* (2007) reported that salt treatments affected root and shoot fresh weight. Growth processes are particularly sensitive to salinity; biomass yield and growth rate are considered reliable criteria for evaluating the degree of salt sensitivity (Larcher 1995). These phenomena were closely related with the extensibility of the cell wall, affecting cell growth and cell division process (Giaveno *et al.* 2007). Higher values of fresh and dry weights of shoot and root were recorded in plants exposed to higher Si levels (6 $mmol.L^{-1}$) as compared to lower levels of Si (2 and 4 $mmol.L^{-1}$). Numerous studies have shown that Si supply may influence positively plant growth and yield (Amirossadat *et al.* 2012; Turan *et al.* 2009). However, beneficial effects of Si usually expressed more clearly when plants were subjected to various stress conditions (Henriet *et al.* 2006). Kudinova (1974) reported that application of Si fertilizer increased the dry weight of barley by 21 and 54% over 20 and 30 days of growth, respectively, relative to plants receiving no supplemental Si. Snyder *et al.* (2007) reported that optimization of Si nutrition results in increased mass and volume of roots, giving increased total and adsorbing surfaces.

Application of salinity levels decreased stem length of maize. Results of Savvas et al. (2007) showed that the increase of the NaCl concentration in the root zone restricted stem length of roses in soilless culture in greenhouse. Salt stress significantly decreased chlorophyll content, while, Si nutrition increased them under salt stress. These results were in accordance with results of Al-aghabary *et al.* (2004) in tomato (*Lycopersicon esculentum* cultivar Mill), Amirossadat *et al.* (2012) in cucumber (*Cucumis sativ*us L.) and Moussa (2006) in maize. Si by increasing the activities of tonoplast $H^+$-ATPase and $H^+$-PPase (which decreased significantly in roots under salt stress), minimize arrived damage to chloroplast and hereby it increase leaf chlorophyll and photosynthesis activity (Liang *et al.* 2005). Tonoplast $H^+$-ATPaseand $H^+$-PPase play a crucial role in salt tolerance of higher plants (Liang *et al.* 2005).

Leaf area significantly contributed toward physiological indices, which boosted up crop growth and accumulation of more photoassimilates from source to sink and consequently, it led to higher grain yield (Ahmed *et al.* 2012). Water stress and turgor loss through inadequate osmotic adjustment slow cellular expensive growth and lead to a reduction in leaf cell size (Curtis and Lauchli 1987). Photosynthetic rate is lower in salt-treated plants and it is expressed with regard to chlorophyll or leaf area (Parida and Bandhu Das 2005). Decreases in photosynthetic rate are due to several factors: 1- dehydration of cell membranes which reduce their permeability to $CO_2$, 2- salt toxicity, 3- reduction of $CO_2$ supply because of hydroactive closure of stomata, 4- enhanced senescence induced by salinity, 5- changes of enzyme activity induced by changes in cytoplasmic structure, and 6- negative feedback by reduced sink activity (Iyengar and Reddy 1996; Parida and Bandhu Das 2005).

Water potential and osmotic potential of plants become more negative with an increase in salinity, whereas turgor pressure increases with increasing salinity (Parida and Bandhu Das 2005). Mali and Aery (2008) reported that Si nutrition (25-200 ppm) enhanced RWC. Probably, formation of double layer cuticle-Si in leaf cause of increasing of thickness of this layer and thus cuticular transpiration in leaf was decreased too much and RWC was increased (Romero-Aranda *et al.* 2006).

## CONCLUSIONS

For overcoming the negative effects of salinity on the plant growth and yield can be to attempt to new strategies. Benifitical effects of Si on yield and quality of maize as well as other crops observed in this study. Positive effect of Si on physiological properties was in conditions that plant grew under salt stress was more remarkable in comparison with conditions that plant grown under normal conditions. The results of this study showed that Si can be involved in the metabolic or physiological activity in higher plants exposed to abiotic stresses. Proper Si nutrition can increase salt resistance by plants. Therefore, it is necessary to investigate Si action, and optimal concentration to be used in this culture. Using of chemical materials such as sodium silicate or potassium silicate as source of Si for combating of salinity are not economical, while crop residues such as stalks of rice, sugarcane and bagasse (sugarcane pulp) can be used as source of Si.

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