INTERNATIONAL JOURNAL OF AGRICULTURE & BIOLOGY
ISSN Print: 1560–8530; ISSN Online: 1814–9596
16–0236/2017/19–6–1475–1482
DOI: 10.17957/IJAB/15.0446
http://www.fspublishers.org

Full Length Article

Silicon Effects on the Growth and Yield of Chickpea under Salinity Stress

Gholam Reza Zamani[1], Jalal Shaabani[2] and Ali Izanloo[1]
[1] *Department of Agronomy and Plant Breeding, University of Birjand, Iran*
[2]*Department of Agronomy & Plant Breeding, Faculty of Agricultural Sciences and Engineering, College of Agriculture and Natural Resources, University of Tehran, Karaj, Iran*
*For correspondence: jshaabani@ut.ac.ir

Abstract

Salinity is a major abiotic stress that limits growth and productivity of crop plants. Role of silicon (Si) in mitigating salt stress damages has gained an increasing attention in recent years. Chickpea is a sensitive crop to salinity and grown mainly in arid and semi-arid regions of the world. This study was conducted to investigate the protective effect of Si on a chickpea landrace from Iran. A factorial experiment was conducted in a completely randomized design with three replications. Salinity at four levels (0, 3, 5 and 7 dS m^{-1}) and silicon at three levels (0, 0.5 and 1 mM) were applied. Phenological, physiological and biochemical characteristics were studied to evaluate Si effects on chickpea yield and its components. Application of Si had no significant effect on phenological traits. Growth related traits, including number of leaves and branches, total dry matter, percent of fertile branches and leaf carbohydrate content were decreased by salinity stress. However, Si application was able to mitigate the effect of salinity in the measured traits. The amount of leaf proline content increased by intensifying salinity stress but using Si showed a significant decrease in proline production. The results showed positive and highly significant correlations between grain yield and total dry matter, leaf number and percent of fertile branches. While a negative and significant correlation was observed between yield and phenological traits as well as leaf proline. It seems that Si can indirectly alleviate the induced damaging effects through increase in vegetative growth. © 2017 Friends Science Publishers

Keywords: *Cicer arietinum*; Vegetative growth; Proline; Carbohydrate; Abiotic stress; Factor analysis

Introduction

Legumes are the most utilized plant family with 20,000 species and in the midst of the most important crops worldwide having major impacts on agriculture, the environment, human/animal health and nutrition. Chickpea, a self-pollinated diploid plant, is the third most important food legume after common bean and field pea, globally (FAOSTAT, 2011). Chickpea is grown typically in arid and semi-arid regions. The soils in these areas are prone to salinization, while chickpea is relatively salt sensitive (Flowers *et al.*, 2010).

Soil salinity is a main environmental limitation to plant production, affecting an estimated 45 million hectares of irrigated field and expected to increase due to global climate changes as well as the consequence of irrigation activities. The damaging effects of salt stress on crop production are significant, because of slower growth rates, reduced tillering and perturbed reproductive development (Amzallag, 2005; Munns and Tester, 2008; Maqbool *et al.*, 2016). Salinity in soil or water is one of the main stresses in arid and semi-arid regions (Pitman and Lauchli, 2002), which can strongly limit crop production. Evaporation of irrigation water containing concentrated dissolved salts leads to accumulation of salinity in soil over time. Presence of salt in the soil profile reduces the plant available water capacity (Dang *et al.*, 2008) and it can intensify osmotic potential, which causes osmotic stress in plant (Munns, 2002). Shaheenuzzamn (2015) reported that the salinity stress decreased the germination rate of chickpea at high salinity level (15 dS m^{-1}). Growth decrease can be severe in chickpea when exposed to salt levels that might be regarded as moderate for most crops (Flowers *et al.*, 2010). Salt stress commonly increases the proline content in plant. Arefian *et al*. (2014) reported that proline content of chickpea leaves showed a significant increase with the increase of NaCl concentrations in all the genotypes but decreased over time (Arefian *et al.*, 2014). They also stated that salt stress significantly reduced the seeds number per pod and 1000-seed weight but did not affect the number of pods per plant (Arefian *et al.*, 2014). The salt treatment in chickpea reduced pod number, filled pod number, seed number and seed yield per plant but empty pod number was less affected in the salt treatment compared to the control plants (Pushpavalli *et al.*, 2016). Salinity may decrease biomass production due to a decrease in water potential, specific ion toxicity, or ion imbalance (Greenway and Munns, 1980).

The ultimate goal of salinity tolerance researches is to

increase the ability of plants to preserve their growth and productivity in saline soils relative to growth in non-saline soils (Roy *et al.*, 2014). Salinity can be mitigated with reclamation, water and drainage. However, the cost of engineering and management is very high. Increasing the water and energy costs emphasizes the need for an alternative strategy (Shannon, 1984). Supplementing the saline irrigation water with silicon (Si) is an alternative strategy to overcome the negative effects of salinity on the plant growth and yield (Tuna *et al.*, 2008). The absorption form of Si by plants is uncharged silicic acid, Si $(OH)_4$ and ultimately precipitated irreversibly throughout the plant as amorphous silica (Gunes *et al.*, 2007). Si as a vital element with an excellent tolerance enhancing potential against abiotic stresses, such as salinity, cold, drought, heavy metals and diseases (Mateos-Naranjo *et al.*, 2013). Silicon has been shown to ameliorate the adverse effects of salinity for plants. The salt tolerance of wheat could be markedly enhanced by adding a small amount of soluble Si (Ahmad *et al.*, 1992). Silicon as an ideal growth-promoting agent can be used to increase plant growth and productivity in various crop plants (Shahid *et al.*, 2015). Liang *et al.* (1996) demonstrated that supplementary Si, increased salinity tolerance in barley grown hydroponically. Silicon is the second most prevalent element in the soil and never found in a free form and always combined with other components, usually forming oxides or silicates.

Shahid *et al.* (2015) evaluated salinity-induced detrimental effects in pea and indicated that exogenous application of Si alone or in combination with phyto-extracts markedly alleviated the salinity-induced severe effects on growth, gas exchange attributes, and productivity. It was also concluded that exogenous use of silicon in combination with phyto-extracts was an effective ameliorative approach to alleviate salinity induced effects in plants, especially in pea, grown under a saline regime. Parande *et al.* (2013) and Saleh *et al.* (2017) have investigated the effects of Si application on the yield and its components in the common bean under salinity stress. They concluded that salinity stress significantly decreased dry matter and the interaction of Si application and salinity stress was significant on the bean yield.

In view of the importance of Si in salt tolerance, it is hypothesized that Si can protect the plant against limiting effects of salinity stress. Therefore, the main objective of this study was to investigate the effect of Si on some phenological, morphological and biochemical properties of chickpea under the salinity stress conditions.

Materials and Methods

Plant Material and Experimental Conditions

In this study, the seeds of a chickpea landrace, from Qaen, South Khorasan province, Iran were used. The experiment was performed in the research greenhouse of Agricultural Faculty, University of Birjand, Iran (20–30°C temperature, 40–60% relative humidity, and at least 16 h photoperiod) from November 2011 to May 2012. A factorial experiment was done in a completely randomized design with three replications. Salinity levels (0, 3, 5 and 7 dS m^{-1}) and silicon application (0, 0.5 and 1 mM) were the two factors. The soil was characterized for its physico-chemical properties before sowing. The pH was 7.98, EC was 0.46 dS m^{-1} and total lime, organic carbon and organic materials were 15, 17 and 29%, respectively. Field capacity was 13.5 and texture of the soil was loamy, which included clay, silt, and sand with 10, 42 and 48%, respectively. NaCl solution was added to the soil into 5 kg plastic pots before planting in order to the soil salinity. The Si powder addition, as Na_2SiO_3, was simultaneously conducted in a same procedure. Ten seeds were initially planted in each pot but five plants were eventually maintained per pot after the plant establishment. The pots irrigation was done with distilled water according to the field capacity. To do this, pots were regularly weighted throughout the duration of growth period using an electronic balance reading with an accuracy of 0.01 g.

Phenological and Morphological Traits

The phenological traits including time of emergence (ET), flowering (FT), pudding (PT) and maturity (MT) were recorded based on number of days after sowing (DAS). The morphological traits including plant height (PH), leaf number (LN), number of seeds (NS), percent of healthy pods (PHP), number of branches (BN) and percent of fertile branches (PFB), total dry matter (TDM) and grain yield (GY), 1000 seed weight (SW) and harvest index (HI) per plant were measured.

Leaf Proline Content

The leaf proline (LP) content was measured 60 days after sowing and shortly before flowering of control treatment. For this reason, 0.5 g of fresh leaves from the middle of the plant was collected from each pot and immediately transferred to the laboratory for extraction with 95% ethanol. Leaf extract was collected by centrifugation at 3500 rpm for 10 min. The supernatant was then used to determine the proline according to Paquin and Lechasseur (1979) procedure using a UV-visible spectrophotometer (X-ma 2000, Human crop, Seoul, South Korea) at 520 nm. The different standards of proline (0, 0.02, 0.04, 0.06, 0.08, and 1 mM) were used to draw the standard curve.

Leaf Carbohydrate

The amount of leaf carbohydrate (LC) of the obtained extracts was estimated spectrophotometrically according to Irigoyen *et al.* (1992) method. The standard curve was traced based on the reads of different concentrations of glucose (0, 0.11, 0.22, 0.33, 0.44, 0.55 and 0.66 mM) at 665 nm using a UV-visible spectrophotometer (X-ma 2000, Human crop, Seoul, South Korea).

Seed Protein Content

After maturity, the plants were harvested from the soil surface and placed in an oven at 45°C for 24 h. The total of seeds from each pot were milled and their total seed protein (SP) contents wasmeasured according to the Kjeldahl (1883) method using Auto Kjeldahl, Foss 8100, Hoganas, Sweden. Total protein of the seed was estimated using Eq. (1):

$$\text{Protein (\%)} = \frac{v \times 0.14 \times F}{m} \quad (1)$$

Where; *v*, *m* and *F* were the volume of acid used (mL), sample weight (g) and protein coefficient (6.25), respectively.

Statistical Analysis

The analysis of variance for data of the experiment was performed using the Statistical Software Package (SAS, version 9.3, SAS Institute Inc. Cary, NC, USA). The *F*-test was used to determine significant effects of each treatment and means comparison at 5% significant level was done with Fisher least significant difference test. Factor analysis by using Principal component analysis method was done for determining the most important traits in the study. Correlation analysis was also performed to investigate the type and intensity of relationships among the traits.

Results

Phenological Traits

Days to emergence, flowering, pudding and maturity significantly increased under salinity stress. Symptoms of salt stress were appeared one week after sowing and were more pronounced at the highest level of salinity at 15–18 days. Duration of all phenological stages was increased by increasing the level of salinity stress. The maximum and minimum number of days to emergence, flowering, pudding and maturity were observed in 7 and 0 dS m^{-1} of salinity, respectively. Seedling emergence was delayed 11 days at the level of 7 dS m^{-1} of salinity stress compared to the control treatment, 55 days delay in flowering time at the highest level of NaCl treatment and with delay of 52 and 61 days for pudding and maturity times, respectively (Table 1). The Si application had no significant effect on the phenological traits.

Plant Height and Harvest Index

Interactions between salinity and Si application were found significant for the plant height. Si had a significant effect on the plant height at 7 dS m^{-1} of salinity stress (Fig. 1). This morphological character was more reduced with increasing intensity of salinity stress, with highest (38.3 ± 1.1 cm) and lowest (20.3 ± 5.3 cm) plant height were observed in the control treatment and the interaction of 7 dS m^{-1} NaCl and 0 mM Si, respectively (Fig. 1). Salinity stress had no significant effect on the HI, however the largest (30.3 ± 2.5%) and smallest (19.4 ± 7.0%) HI values were obtained at the interaction of 5 dS m^{-1} of salt stress and 0.5 mM Si and 0 dS m^{-1} and 0.5 mM Si, respectively (Table 1).

Leaf and Branches Number and Total Dry Matter

Salinity stress decreased the number of leaves and branches, significantly. The lowest branch numbers (3.88 ± 0.5 per plant) was found at 7 dS m^{-1} of salt without Si application and the highest branches for the control treatment (9.6 ± 1.6 per plant) (Fig. 2). The least (47.2 ± 4.7 per plant) and the most (114.0 ± 7.4 per plant) number of leaves were obtained at 5 dS m^{-1} and 1 mM Si, and for no salt stress and 1 mM Si, respectively (Table 1).

The total dry matter was significantly decreased under salinity stress compared to the non-stress conditions. Maximum (3.4 ± 0.4 g per plant) and minimum (1.3 ± 0.0 g per plant) amounts of this character were with no salinity and 1 mM Si and 5 dS m^{-1} with 1 mM Si, respectively. The highest level of total dry matter (2.0 ± 0.5 g per plant) at the highest level of salinity was found also in Si of 1 mM (Table 1).

Grain Yield and its Components

Salinity had a significant decreasing effect on the grain yield and 1000 grain weight with the intensity of salt stress, while number of seeds was not affected by the different levels of salinity. However, the highest (411.9 ± 26.3 g) and lowest 1000 grain weight (271.8 ± 25.2 g) were obtained in treatment combinations of no salinity with 0.5 mM Si and 5 dS m^{-1} with 1 mM Si, respectively. The highest grain yield (0.63 ± 0.1 g per plant) was obtained without salinity and application of 1 mM Si and the lowest grain yield (0.33 ± 0.0 g per plant) was observed in 7 dS m^{-1} with 0.5 mM Si (Table 1). Although salinity did not show significant effect on the percent of healthy pods, the results revealed that the percent of fertile branches was affected significantly by salinity stress ($P \leq 0.05$). The interaction between salinity and Si on the percent of fertile branches was significant ($P \leq 0.05$) and the highest percent of healthy pods (92.8 ± 4.1%) were observed at 5 dS m^{-1} with application of 1 mM Si and the lowest percent (65.2 ± 10.6%) was observed at highest salinity of 7 dS m^{-1} and 1 mM Si. The highest percent of fertile branches (18.2 ± 4.3%) was obtained at 3 dS m^{-1} with application of 0.5 mM Si (Table 1).

Proline, Protein and Carbohydrate Content

The salinity effect on leaf proline and carbohydrate content was significant; however, seed protein was affected not by salinity levels and different Si concentrations (Fig. 3). The highest amount of leaf proline (0.725 ± 0.0 mg g^{-1}) was

Table 1: Effect of salinity on some of phenological, anatomical, and physiological parameters with or without supplementary silicon of chickpea plants grown

Traits	Treatments													
	LSD	Si (Mm)	0				0.5				1			
	(0.05)	NaCl (dS m^{-1})	0	3	5	7	0	3	5	7	0	3	5	7
ET (Day)	2.06		7.0±0.0	10±0.0	16.6±1.3	15.6 ± 0.6	7.0 ± 0.0	10 ± 0.0	14.3 ± 1.2	18.3 ± 1.2	7.0 ± 0.00	9.3 ± 0.3	16.0 ± 2.3	17.6 ± 0.3
FT (Day)	11.31		69.7±1.7	95.9±3.8	123.8±4.3	121.5±5.7	76.3 ± 0.9	105.4±6.0	119.9±1.4	126.7±0.2	71.8 ± 3.9	107.1±6.1	121.4±1.7	124.3 ± 3.7
PT (Day)	11.85		87.5±2.7	107.5±6.9	130.0±4.0	129.0±4.8	90.1 ±1.8	115.7±4.4	126.9±1.8	135.4±1.7	84.8 ± 5.7	119.5±4.7	128.1±1.8	132.7 ± 3.6
MT (Day)	10.56		102.6±2.8	133.1±4.9	163.1±8.5	157.6±0.6	106.6±4.4	137.4±1.2	155.6±1.8	162.1±2.0	109.0±2.8	141.0±2.2	156.8±0.2	162.9 ± 2.8
T.D.M (g. plant^{-1})	0.71		2.6±0.0	2.0±0.0	1.5 ± 0.1	1.9 ± 0.2	2.8 ± 0.2	2.0 ± 0.0	1.5 ± 0.1	1.5 ± 0.0	3.4 ± 0.4	2.1 ± 0.0	1.3 ± 0.0	2.0 ± 0.5
P.H.P (%)	22.58		75.2±5.5	83.00±4.5	90.4 ± 9.5	79.4 ± 2.4	69.8±10.3	72.3 ± 7.4	72.2 ± 5.5	67.4±10.3	69.6 10.8	76.1 ± 4.7	92.8 ± 4.1	65.2 ± 10.6
P.F.B (%)	10.59		12.9±1.7	6.3±3.2	5.0 ± 3.2	13.0 ± 4.0	10.4 ± 1.7	18.2 ± 4.3	13.6 ± 6.2	0.0 ± 0.0	14.0 ± 7.1	14.7 ± 2.4	0.0 ± 0.0	0.0 ± 0.0
S.W (g)	117.29		410.5±54.1	333.7±12.7	297.0±15.1	318.8 14.0	411.9±26.3	344.4±15.1	346.4±62.1	276.1±62.0	380.4±18.5	391.7±4.9	271.8±25.2	284.9±18.6
N.S (plant^{-1})	0.93		1.5±0.3	1.7±0.3	1.3 ± 0.2	1.5 ± 0.2	1.3 ± 0.4	1.7 ± 0.4	1.3 ± 0.0	1.3 ± 0.3	1.7 ± 0.4	1.5 ± 0.2	1.4 ± 0.4	1.2 ± 0.1
GY (g.plant^{-1})	0.31		0.59±0.0	0.54±0.1	0.37 ± 0.0	0.48 ± 0.0	0.54 ± 0.1	0.57 ± 0.0	0.46 ± 0.0	0.33 ± 0.0	0.63 ± 0.1	0.58 ± 0.1	0.38 ± 0.0	0.35 ± 0.0
H.I (%)	14.43		22.5±2.3	26.3±7.1	23.6 ± 4.3	24.9 ± 3.4	19.4 ± 7.0	28.0 ± 4.7	30.3 ± 2.5	21.9 ± 2.8	19.8 ± 5.6	27.6 ± 4.7	28.4 ± 1.0	21.3 ± 8.0
LN (plant^{-1})	22.22		102.0±10.5	75.7±11.1	60.3 ± 2.9	54.1 ± 2.7	103.3±6.8	84.5±2.9	59.0±12.0	56.9 ± 5.7	114.0±7.4	79.1 ± 5.4	47.2 ± 4.7	58.6 ± 10.1

Each value is the means of three replicates ± standard error (SE). Fisher protected LSD at $P \leq 0.05$. ET: emergence time; FT: flowering time; PT: pudding time; MT: maturity time; TDM: total dry matter; PHP: percent of healthy pod; PFB: percent of fertile branch; SW: 1000-seed weight; NS: number of seed; GY: grain yield; HI: harvest index; LN: leaf number

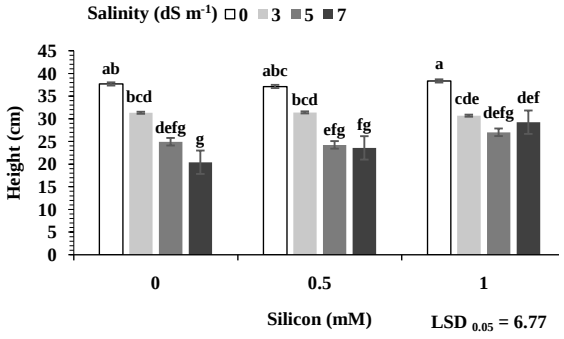

Fig. 1: Effect of salinity (0, 3, 5 and 7 dS m^{-1} NaCl) on the height of chickpea plants grown with supplementary silicon (0, 0.5 and 1 mM). Different small letters on bars represent statistically significant differences at 0.05 significant level

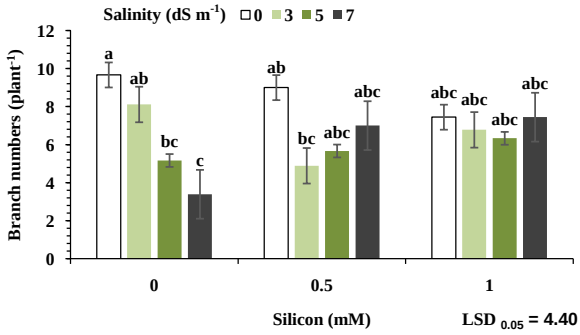

Fig. 2: Effect of salinity (0, 3, 5 and 7 dS m^{-1} NaCl) on number of branches in chickpea plants grown with supplementary silicon (0, 0.5 and 1 mM). Different small letters on bars represent statistically significant differences at 0.05 significant level

observed at 7 dS m^{-1} with 1 mM of silicon, whereas the lowest amount (0.008 ± 0.0 mg g^{-1}) in control without salinity and 0.5 mM Si (Fig. 4). The highest (50.0 ± 10.2 mg 0.5 g^{-1}) and lowest (12.6 ± 2.0 mg 0.5 g^{-1}) leaf carbohydrate contents were observed in control with application of 0.5 mM Si and at 7 dS m^{-1} with 0.5 mM Si application, respectively (Fig. 5).

Correlation Analysis

A negative correlation was found between among the grain yield and days to emergence (r = -0.941, $P \leq 0.01$), flowering (r = -0.816, $P \leq 0.01$), pudding (r = -0.811, $P \leq 0.01$), maturity (r = -0.737, $P \leq 0.01$) and leaf proline content (r = -0.838, $P \leq 0.01$). A positive significant correlation was also obtained between grain yield and plant height (r = 0.708, $P \leq 0.05$), 1000-seed weight (r = 0.871, $P \leq 0.01$), seed numbers (r = 0.775, $P \leq 0.01$), percent of fertile branches (r = 0.839, $P \leq 0.01$), branch number (r = 0.832, $P \leq 0.01$), total dry matter (r = 0.752, $P \leq 0.01$) and leaf carbohydrate (r = 0.647, $P \leq 0.05$). However, correlations between grain yield and percent of healthy pods, seed protein and harvest index were not significant (Table 2).

Factor Analysis

Since correlation coefficients may not provide sufficient information to determine relationships between the different traits, factor analysis was used due to the multiple benefits of multivariate statistical analysis. According to observation in Eigen values, three components by amount over 1 of Eigen values were found, which together explained 88.66% of total variations. The first component that explained most of the variations (63.94%) had large and negative coefficients for days to emergence, flowering, pudding, maturity and leaf proline. This component had a positive coefficient for total dry matter, fertile branches percent, 1000-seed weight, seed number, yield, leaf carbohydrate, number of leaves and branches and plant height.

Table 2: Bivariate correlation coefficients between various studied characters

	ET	FT	PT	MT	TDM	PHP	PFB	SW	NS	GY	HI	PR	LC	LP	LN	BN
FT	0.933**															
PT	0.924**	0.997**														
MT	0.945**	0.995**	0.989**													
TDM	-0.815**	-0.884**	-0.888**	-0.865**												
PHP	0.225^{ns}	0.239^{ns}	0.214^{ns}	0.257^{ns}	0.458^{ns}											
PFB	-0.682**	-0.490*	-0.490*	-0.517^{ns}	0.473^{ns}	-0.213^{ns}										
SW	-0.920**	-0.847**	-0.832**	-0.868**	0.744**	-0.310^{ns}	0.751**									
NS	-0.626**	-0.502^{ns}	-0.503^{ns}	-0.509^{ns}	0.465^{ns}	-0.054^{ns}	0.657*	0.408^{ns}								
GY	-0.941**	-0.816**	-0.811**	-0.737**	0.752**	-0.192^{ns}	0.839**	0.871**	0.775**							
HI	0.207^{ns}	0.446^{ns}	0.444^{ns}	0.448^{ns}	-0.637*	0.436^{ns}	0.215^{ns}	-0.190^{ns}	0.150^{ns}	0.021^{ns}						
SP	0.027^{ns}	-0.158^{ns}	-0.146^{ns}	-0.176^{ns}	0.312^{ns}	-0.330^{ns}	-0.295^{ns}	0.027^{ns}	-0.264^{ns}	-0.162^{ns}	-0.668*					
LC	-0.826**	-0.920**	-0.930**	-0.949**	0.830**	-0.285^{ns}	0.385^{ns}	0.805**	0.251^{ns}	0.647*	-0.521^{ns}	0.278^{ns}				
LP	0.892**	0.814**	0.825**	0.819**	-0.571*	-0.114^{ns}	-0.647*	-0.817**	-0.548^{ns}	-0.838**	-0.064^{ns}	0.289^{ns}	-0.759**			
LN	-0.928**	-0.953**	-0.950**	-0.942**	0.923**	-0.387^{ns}	0.574^{ns}	0.857**	0.515^{ns}	0.832**	-0.488^{ns}	0.069^{ns}	0.887**	-0.768**		
BN	-0.542^{ns}	-0.685*	-0.656*	-0.677*	0.517^{ns}	-0.296^{ns}	-0.154^{ns}	0.486^{ns}	-0.060^{ns}	0.279^{ns}	-0.497^{ns}	0.297^{ns}	0.673*	-0.403^{ns}	0.595*	
PH	-0.870**	-0.921**	-0.913**	-0.915**	0.842**	-0.296^{ns}	0.336^{ns}	0.742**	0.378^{ns}	0.708*	-0.455^{ns}	0.113^{ns}	0.868**	-0.727**	0.920**	0.767**

*, **: Significant at 0.05 and 0.01 levels, respectively. ns: non-significant. ET: emergence time; FT: flowering time; PT: pudding time; MT: maturity time; TDM: total dry matter; PHP: percent of healthy pod; PFB: percent of fertile branch; SW: 1000-seed weight; NS: number of seed; GY: grain yield; HI: harvest index; SP: seed protein; LC: leaf carbohydrate; LP: leaf proline; LN: leaf number; BN: branch number; PH: plant height

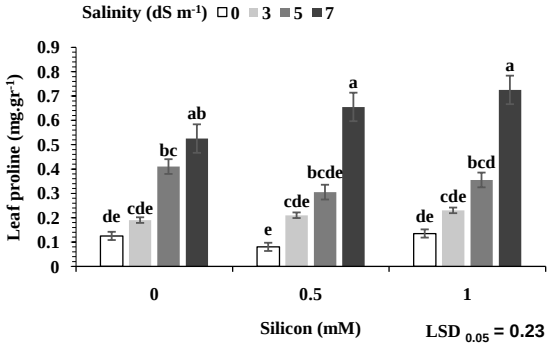

Fig. 3: Effect of salinity (0, 3, 5 and 7 dS m^{-1} NaCl) on leaf proline content of chickpea plants grown with supplementary silicon (0, 0.5 and 1 mM). Different small letters on bars represent statistically significant differences at 0.05 significant level

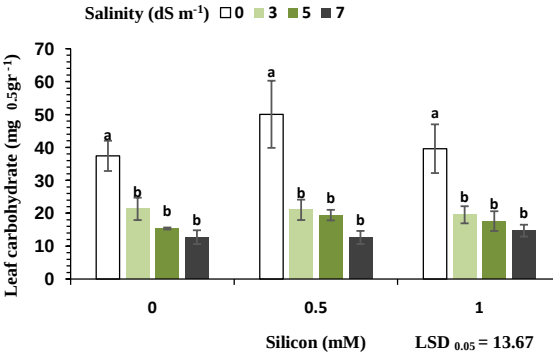

Fig. 4: Effect of salinity (0, 3, 5 and 7 dS m^{-1} NaCl) on leaf carbohydrate content of chickpea plants grown with supplementary silicon (0, 0.5 and 1 mM). Different small letters on bars represent statistically significant differences at 0.05 significant level

The most important coefficients in second component were seed number, harvest index, seed protein and number of branches, as this component explained 17.41% of the variations. Third component also explained 7.30% of variations that percent of healthy pods was considered as a trait with the highest negative coefficient (Table 3 and 4).

Discussion

Salinity is an important environmental constraints limiting chickpea production, which require breeding different varieties with improved salt tolerance. Alleviation of salinity stress in this study was assessed through Si supplementation to the soil. Results showed the positive and protective effects of Si in some of desired traits. One of the initial effects of salt stress is the growth rate reduction. Salt in soil or water can inhibit the plant growth by osmotic or water-deficit stress, which can reduce the plant's ability to take up water leading to a slower growth and further reduction of the growth via ion toxicity (Horgan and Henderson, 2015). Plants can adjust their phenology under environmental conditions through an accelerating activity between the vegetative growth and leaf senescence, which the action can rapidly promote time of reproductive phases of flowering and pudding (Allu *et al.*, 2014). Days to emergence, flowering, pudding and maturity traits were delayed by salt stress, which can be indirectly, declined the yield due to the negative correlation between the phenological traits and chickpea performance (Table 2). Slower growth is an adaptive attribute for plant survival under the different stress conditions because this action provide a possible for plants to rely on multiple resources (e.g., building blocks and energy) to cope with the stress (Zhu, 2001). Al-Mutawa (2003) determined the germination and seedling growth responses of 30 genotypes of chickpea under the various salinity levels of irrigation water. All genotypes showed a salt tolerance either at germination or seedling growth stage at low salinity level (4 dS m^{-1}) (Al-Mutawa, 2003). The

Table 3: Total variance explained

Component	Total	% of variance	Cumulative %
1	10.988	63.947	63.947
2	2.851	17.418	81.404
3	1.325	7.300	88.665

Extraction method: Principle Component Analysis (PCA)

Table 4: Rotated component matrix

Trait	Factors		
	1	2	3
ET	-0.971	-0.182	0.029
FT	-0.982	0.081	0.111
PT	-0.978	0.069	0.121
MT	-0.985	0.063	0.096
TDM	0.902	-0.227	0.231
PHP	-0.318	0.399	-0.692
PFB	0.603	0.621	0.410
SW	0.903	0.145	0.064
NS	0.546	0.560	0.272
GY	0.880	0.415	0.153
HI	-0.392	0.814	-0.156
SP	0.102	-0.786	0.235
LC	0.910	-0.239	-0.141
LP	-0.835	-0.406	0.323
LN	0.978	-0.054	0.060
BN	0.617	-0.523	-0.403
PH	0.918	-0.176	-0.179

Extraction method: Principle Component Analysis (PCA). ET: emergence time; FT: flowering time; PT: pudding time; MT: maturity time; TDM: total dry matter; PHP: percent of healthy pod; PFB: percent of fertile branch; SW: 1000-seed weight; NS: number of seed; GY: grain yield; HI: harvest index; SP: seed protein; LC: leaf carbohydrate; LP: leaf proline; LN: leaf number; BN: branch number; PH: plant height

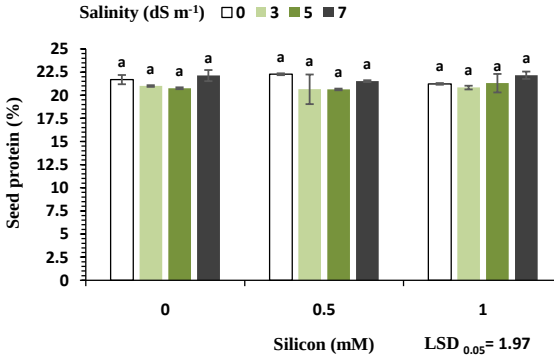

Fig. 5: Effect of salinity (0, 3, 5, and 7 dS m^{-1} NaCl) on seed protein percent of chickpea plants grown with supplementary silicon (0, 0.5, and 1 mM). Different small letters on bars represent statistically significant differences at 0.05 significant level

results of present study also revealed that the chickpea well tolerated low salt levels (up to 3 dS m^{-1}). According to the high negative correlation between the examined phenological traits and chickpea yield (Table 2), the effect of Si addition delayed the emergence of phenological traits (Table 1). The results of this study showed that the production rate of branches and leaves and the increase of the plant height were limited due to the salt existence in root growth environment compared to the condition of without salt stress. However, addition of the Si could considerably compensate these defects (Table 1; Figs. 1 and 2). The increase of plant height and numbers of leaf and branch considerably increased in presence of Si under high salinity conditions at reproductive phase. The free proline accumulation increased under salt stress in chickpea as the maximum proline amount was observed at 7 dS m^{-1} salinity (Fig. 3). Proline, a compatible osmolyte, plays a vital role in several multiple functions particularly in stress adaptation, recovery and signaling, proteins stabilization and complexes in the chloroplast, cytosol and protection of the photosynthetic organs in plants (Szabados and Savouré, 2010). Ashraf and Foolad (2007) suggested that the proline could successfully improve stress tolerance in plants. Our findings demonstrated that Si could provide a greater yield by protecting chickpea at high salt concentrations. Delauney and Verma (1993) presented the yield enhancement under saline stress can be due to the osmotic adjustment of proline and carbohydrates accumulation in plant tissues. However, the correlation between concentration of salt and these functional compounds is generally poor (Cordovilla *et al.*, 1996). The results of current research showed a significant relationship between increase of NaCl concentration and proline accumulation in leaves, while a decrease in concentration of leaf carbohydrates was found due to the salinity stress (Figs. 3 and 4). Kaur *et al.* (2014) reported that salinity decreased the number of filled pods per plant, however the reduction in number of filled pods was associated with an increase in pod abortion of pods in salt-sensitive chickpea genotypes (Kaur *et al.*, 2014). It was observed that percent of healthy pods and fertile branches under high saline levels with Si supplementation were larger than the controlled condition without Si (Table 1). Si accumulation among the plant species have a widespread variation, as Poaceae family showing high Si build-up, while Fabaceae are considered as low Si accumulators (Hodson *et al.*, 2005; Ma and Yamaji, 2008; Meena *et al.*, 2014). Flowers *et al.* (2010) also detailed that chickpea has low ability to uptake Si from soil (Flowers *et al.*, 2010). Langdale *et al.* (1973) stated that grain filling in plants have a close relationship with remobilization of photo-assimilation (Langdale *et al.*, 1973). Results of this study showed no significant difference among of seed protein at the different salt-stress conditions (Fig. 5).

Singla and Garg (2005) reported a more drastic reduction of pod and seed numbers, which resulted in a significant decline in weight of seeds, 100 seed weight and harvest index of all the chickpea cultivars under salinity stress (Singla and Garg, 2005). Results of present study demonstrated that the number of seeds, 1000 seed weight and harvest index showed no significant differences during the multiple salinity levels. Therefore, the grain yield decrease was more affected through other factors or it may be due to the supporting role of Si in mitigating high level of

salinity stress (Tables 1 and 2). Salinity stress up to 5 dS m^{-1} had no significant effect on the total dry matter of chickpea; however, both the total dry matter and grain yield in over 7 dS m^{-1} of salt stress were particularly decreased. Observations of this study showed that the total dry matter was decreased under stress condition without Si application. However, the Si application did not show a regular process in protecting of plant for both traits of yield and total dry matter under salinity-stressed conditions. Parande *et al.* (2013) also reported that total dry matter of common bean decreased with an increase in salinity level. They also pointed out that added Si did not significantly affected the dry matter compared to the control treatment (Parande *et al.*, 2013). Supplementary Si resulted in a significant increase in dry matter and chlorophyll contents of plants grown at high NaCl level but the obtained values in the highest Si treatment were lower than the control treatment (Tuna *et al.*, 2008). Miyake (1992), Bonilla and Tsuchiya (1998) and Liang (1999) observed such response for cucumber and tomato, rice and barley, respectively.

Taking into account that, the ability of silicon to influence the anatomical-morphological, physiological and biochemical reactions in plants during multiple abiotic stresses such as salinity, drought, metal toxicity and UV-radiation, which can be expected that silicon will have a protective function in plants in environmental stress conditions (Balakhnina and Borkowska, 2013). Si can decrease stability of permeability of the plasma membrane of leaf cells and improve the ultrastructure of chloroplast cells accumulation of polysilicic acids inside cells (Biel *et al.*, 2008), improved the water uptake in salt-stressed cucumber by up-regulating the aquaporin gene expression,decreased Na^+ uptake by decreasing transpiration and adjusting the levels of solutes and phytohormones (Rizwan *et al.*, 2015).

Conclusion

Si supplementation is a strategy to alleviate salinity stress effects. Results showed that the amount of Si in the soil must be appropriate by salinity level because of adverse effects observed in some of traits such as percent of healthy pod. However, it has a positive role in some of traits including total dry matter, leaf number, plant height, branch number, leaf carbohydrate, leaf proline and percent of fertile branch at high salinity stress levels. It is concluded that application of Si against salinity could alleviate salinity effects on crop production through increasing of plant growth and development.

References

Ahmad, R., S.H. Zaheer and S. Ismail, 1992. Role of silicon in salt tolerance of wheat (*Triticum aestivum* L.). *Plant Sci.*, 85: 43–50

Al-Mutawa, M., 2003. Effect of salinity on germination and seedling growth of chickpea (*Cicer arietinum* L.) genotypes. *Int. J. Agric. Biol.*, 5: 226–229

Allu, A.D., A.M. Soja, A. Wu, J. Szymanski and S. Balazadeh, 2014. Salt stress and senescence: identification of cross-talk regulatory components. *J. Exp. Bot.*, 65: 3993–4008

Amzallag, G.N., 2005. Perturbed reproductive development in salt-treated *Sorghum bicolor*: a consequence of modifications in regulation networks? *J. Exp. Bot.*, 56: 2821–2829

Arefian, M., S. Vessal and A. Bagheri, 2014. Biochemical changes and SDS-PAGE analyses of chickpea (*Cicer arietinum* L.) genotypes in response to salinity during the early stages of seedling growth. *J. Biol. Environ. Sci.*, 8: 99–109

Ashraf, M. and M. Foolad, 2007. Roles of glycine betaine and proline in improving plant abiotic stress resistance. *Environ. Exp. Bot.*, 59: 206–216

Balakhnina, T. and A. Borkowska, 2013. Effects of silicone on plant resistance to environmental stresses. *Int. Agrophys.*, 27: 225–232. Biel, K.Y., V.V. Matichenkov and I.R. Fomina, 2008. Protective role of silicon in living systems. In: Functional Foods For Chronic Diseases. Martirosyan, D.M. (ed.). D and A inc., Richardson Press, Dallas, Texas, USA.

Biel, K.Y., V.V. Matichenkov and I.R. Fomina, 2008. Protective role of silicon in living systems. *In: Functional Foods For Chronic Diseases.* Martirosyan, D.M. (ed.). D and A Inc., Richardson Press, Dallas, Texas, USA

Bonilla, P. and M. Tsuchiya, 1998. Induction of salt tolerance in rice by silica treatment. *Philip. J. Crop. Sci.*, 23: 35–44

Cordovilla, M., F. Ligero and C. Lluch, 1996. Growth and nitrogen assimilation in nodules in response to nitrate levels in *Vicia faba* under salt stress. *J. Exp. Bot.*, 47: 203–210

Dang, Y., R. Dalal, D. Mayer, M. McDonald, R. Routley, G. Schwenke, S. Buck, I. Daniells, D. Singh and W. Manning, 2008. High subsoil chloride concentrations reduce soil water extraction and crop yield on vertosols in north-eastern Australia. *Crop. Pasture Sci.*, 59: 321–330

Delauney, A.J. and D.P.S. Verma, 1993. Proline biosynthesis and osmoregulation in plants. *Plant J.*, 4: 215–223

FAOSTAT, 2011. http://faostat.fao.org/. Accessed 11 July 2011

Flowers, T.J., P.M. Gaur, C.L. Gowda, L. Krishnamurthy, S. Samineni, K.H. Siddique, N.C. Turner, V. Vadez, R.K. Varshney and T.D. Colmer, 2010. Salt sensitivity in chickpea. *Plant. Cell. Environ.*, 33: 490–509

Greenway, H. and R. Munns, 1980. Mechanisms of salt tolerance in non-halophytes. *Annu. Rev. Plant Biol.*, 31: 149–190

Gunes, A., D.J. Pilbeam, A. Inal, E.G. Bagci and S. Coban, 2007. Influence of silicon on antioxidant mechanisms and lipid peroxidation in chickpea (*Cicer arietinum* L.) cultivars under drought stress. *J. Plant Interact.*, 2: 105–113

Hodson, M.J., P.J. White, A. Mead, and M.R. Broadley, 2005. Phylogenetic variation in the silicon composition of plants. *Ann. Bot.*, 96: 1027–1046.

Horgan, K.J. and J.O. Henderson, 2015. miRNA mediated post-transcriptional gene regulation in response to abiotic stress in plants. *J. Stud. Res.*, 4: 21–28

Irigoyen, J., D. Einerich and M. Sánchez-Díaz, 1992. Water stress induced changes in concentrations of proline and total soluble sugars in nodulated alfalfa (*Medicago sativa* L.) plants. *Physiol. Plant.*, 84: 55–60

Kaur, P., J. Kaur, S. Kaur, S. Singh and I. Singh, 2014. Salinity induced physiological and biochemical changes in chickpea (*Cicer arietinum* L.) genotypes. *J. Appl. Nat. Sci.*, 6: 578–588

Kjeldahl, J., 1883. Neue methode zur bestimmung des stickstoffs in organischen körpern. *Fresen. J. Anal. Chem.*, 22: 366–382

Langdale, G.W., J. Thomas and T. Littleton, 1973. Nitrogen metabolism of stargrass as affected by nitrogen and soil salinity. *Agron. J.*, 65: 468–470

Liang, Y., 1999. Effects of silicon on enzyme activity and sodium, potassium and calcium concentration in barley under salt stress. *Plant Soil*, 209: 217–224

Liang, Y., Q. Shen, Z. Shen and T. Ma, 1996. Effects of silicon on salinity tolerance of two barley cultivars. *J. Plant. Nutr.*, 19: 173–183

Ma, J.F. and N. Yamaji, 2008. Functions and transport of silicon in plants. *Cell. Mol. Life. Sci.*, 65: 3049–3057

Maqbool, N., A. Wahid and S.M.A. Basra, 2016. Varied patterns of sprouting and nutrient status of sugarcane sprouts in simulated and natural saline/sodic soils across two growing seasons. *Int. J. Agric. Biol*., 18: 873–880

Mateos-Naranjo, E., L. Andrades-Moreno and A.J. Davy, 2013. Silicon alleviates deleterious effects of high salinity on the halophytic grass Spartina densiflora. *Plant. Physiol. Biochem*., 63: 115–121

Meena, V.D., M.L. Dotaniya, V. Coumar, S. Rajendiran, S. Kundu, and A.S. Rao, 2014. A case for silicon fertilization to improve crop yields in tropical soils. *Proc. Natl. Acad. Sci. USA, India Section B: Biol. Sci*., 84: 505–518

Miyake, Y., 1992. The effect of silicon on the salt tolerance of cucumber and tomato plants. *Sci. Rep. Fac. Agric*., 80: 93–99

Munns, R., 2002. Comparative physiology of salt and water stress. *Plant. Cell. Environ*., 25: 239–250

Munns, R. and M. Tester, 2008. Mechanisms of salinity tolerance. *Annu. Rev. Plant Biol*., 59: 651–681

Paquin, R. and P. Lechasseur, 1979. Observations sur une méthode de dosage de la proline libre dans les extraits de plantes. *Can. J. Bot*., 57: 1851–1854

Parande, S., G.R. Zamani, M. Syyari Zahan and M. Ghaderi, 2013. Effects of silicon application on the yield and component of yield in the common bean (*Phaseolus vulgaris* L.) under salinity stress. *Int. J. Agron. Plant Prod*., 4: 1574–1579

Pitman, M.G. and A. Läuchli, 2002. Global impact of salinity and agricultural ecosystems. *In: Salinity: Environment-Plants-Molecules*, pp: 3-20. Kluwer Academic Publishers, Springer, The Netherlands

Pushpavalli, R., J. Quealy, T. Colmer, N. Turner, K.H.M. Siddique, M. Rao and V. Vadez, 2016. Salt stress delayed flowering and reduced reproductive success of chickpea (*Cicer arietinum* L.), a response associated with Na^+ accumulation in leaves. *J. Agron. Crop Sci*., 202: 125–138

Rizwan, M., S. Ali, M. Ibrahim, M. Farid, M. Adrees, S.A. Bharwana, M. Zia-ur-Rehman, M.F. Qayyum and F. Abbas, 2015. Mechanisms of silicon-mediated alleviation of drought and salt stress in plants: a review. *Environ. Sci. Pollut. Res. Int.*, 22: 15416–15431

Roy, S.J., S. Negrão and M. Tester, 2014. Salt resistant crop plants. *Curr. Opin. Biotechnol*., 26: 115-124

Saleh, M.H.G., H.S.M. Al-Zahrani and E.M.R. Metwali, 2017. Improving the salinity tolerance in potato (*Solanum tuberosum*) by exogenous application of silicon dioxide nanoparticles. *Int. J. Agric. Biol.*, 19: 183–194

Shaheenuzzamn, M., 2015. Screening of chickpea genotypes against salinity stress. *Bangl. J. Agric. Res.*, 39: 605–619

Shahid, M.A., R.M. Balal, M.A. Pervez, T. Abbas, M.A. Aqeel, M.M. Javid and F. Garcia-sanchez, 2015. Foliar spray of phyto-extracts supplemented with silicon: an efficacious strategy to alleviate the salinity-induced deleterious effects in pea (*Pisum sativum* L.). *Turk. J. Bot.*, 39: 408–419

Shannon, M., 1984. Breeding, selection and the genetics of salt tolerance. In: *Salinity Tolerance In Plants-Strategies For Crop Improvement*. pp: 313–331. Staples, R.C. and G.H. Toenniessen (eds.). Wiley International, New York, USA

Singla, R. and N. Garg, 2005. Influence of salinity on growth and yield attributes in chickpea cultivars. *Turk. J. Agric. For*., 29: 231–235

Szabados, L. and A. Savouré, 2010. Proline: a multifunctional amino acid. *Trends. Plant. Sci*., 15: 89–97

Tuna, A.L., C. Kaya, D. Higgs, B. Murillo-Amador, S. Aydemir and A.R. Girgin, 2008. Silicon improves salinity tolerance in wheat plants. *Environ. Exp. Bot*., 62: 10–16

Zhu, J.K., 2001. Plant salt tolerance. *Trends Plant. Sci*., 6: 66–71

(Received 06 May 2016; Accepted 27 February 2017)