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Impact of silicon foliar application in enhancing antioxidants, growth, flowering and yield of squash plants under deficit irrigation condition

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ABSTRACT

Plant production under climate changes requires unique treatments to overcome the detrimental effects of abiotic stress, such as water deficiency stress. Silicon (Si) has many beneficial effects, especially in plants subjected to different types of stress. Hence, Si foliar application was used to study its potential effects on boosting osmolytes content, activity of antioxidant enzymes, growth of vegetative and flowering organs, and yield of squash plants (*Cucurbita pepo* L.) under deficit irrigation. Two field experiments were conducted during the winter seasons of 2019 and 2020 at the Experimental Farm, Faculty of Agriculture, Ain Shams University, Egypt. Three concentrations of Si at 0, 2000 and 4000 ppm in the form of monosilicic acid (H_4SiO_4) under two levels of water irrigation at 80% of water holding capacity (WHC) as control and 50% of WHC as drought treatment. Drought treatment significantly reduced the growth parameters: shoot height, shoot fresh and dry weights, leaves number/plant, average leaf area, fruit setting and yield traits, and the physiological attributes: leaf relative water content (LRWC), total chlorophylls (SPAD), total soluble proteins (TSP) and catalase (CAT) activity. Meanwhile, significant increases in the concentrations of stress indicators (total free amino acids and proline) were observed compared to the control plants (80% WHC). Both rates of Si achieved marked increases in the vegetative growth, flowering, chlorophylls, LRWC, TSP, superoxide dismutase (SOD), CAT, peroxidase (POD) and polyphenol oxidase (PPO) activity, which in turn reflects on improving fruit setting %, total fruits, and yield of squash plants under well or deficit irrigation. Spraying the high level of Si (4000 ppm) was the most effective treatment that emended the adverse effects of drought.

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1. Introduction

Squash (*Cucurbita pepo* L.) is an important vegetable crop, widely grown and consumed as food and herbal remedies in many countries around the world. It considers a natural source of carotenoids, carbohydrates, polysaccharides, flavonoids, alkaloids, tocopherols, phenols, terpenoids, saponins, sterols, minerals and fatty acids (Ratnam et al., 2017; Salehi et al., 2019).

Drought is considered as one of the most significant environmental stresses that cause adversely dangerous effects on plant growth, development, biochemical constituents, metabolic processes, plant physiology and final crop yield (Farooq et al., 2009; Kabay, 2020; Osman et al., 2021a; Rao et al., 2016; Zhu and Gong, 2013). Insufficient extension and expansion of the cell wall due to subjecting plants to osmotic stress such as drought results in slowing or stopping the plant growth entirely (Hafez et al., 2020; Osman and Salim, 2016). Under drought conditions, the content of photosynthetic pigments, stomatal conductance, and CO_2 fixation decreased, resulting in a reduction in carbohydrate accumulation (Hafez et al., 2021b; Widuri et al., 2020). In the opinion of Osman (2015), plants are more sensitive at the flowering stage when subjected to a long-term drought than short-term drought. Nevertheless, squash considers a hypersensitive vegetable crop to either deficit irrigation or excess irrigation conditions (Amer, 2011).

Many reports presented silicon (Si) as a prosperous beneficial plant nutrient for stimulating the growth and yield of different crops (Bakhat et al., 2018; Rizwan et al., 2015; Shehata and Abdelgawad, 2019). Therefore, Si has important vital roles in alleviating the adverse negative impacts of drought stress (Thorne et al., 2020; Tubana et al., 2016; Yavaş and Ünay, 2017). Silicon is widely accepted that it has two key processes in contribution to stress resistance: 1) physical and mechanical

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protection from Si deposits in the cell wall, which reduces leaves stomatal conductance and transpiration, and 2) a biochemical reaction causing metabolic changes (Luyckx et al., 2017). Under stress conditions, Si affects endogenous phytohormones, including jasmonic acid and salicylic acid, causing gene expression alterations followed by physiological changes such as antioxidant enzymes activity, chlorophyll content, polyamine content, soluble protein content, water potential, and Rubisco content (Coskun et al., 2019).

It could be summarized from previous studies (Hafez et al., 2021a; Kaushik and Saini, 2019; Mostofa et al., 2021; Sahebi et al., 2015; Savvas and Ntatsi, 2015) that the application of Si is a good strategy for maximizing crops productivity and helping vegetable crops overcome different types of environmental stresses. The objective of the current research is to study the impact of silicon at rates 2000 and 4000 ppm as foliar applications in ameliorating the harmful effects of water deficit stress and influences the vegetative growth, flowering, plant antioxidants defense system and yield of squash plant under well water and drought conditions.

2. Material and methods

2.1. Cultivation

This research was conducted during two successive winter seasons in February 2019 and 2020 at the Experimental Farm, Faculty of Agriculture, Ain Shams University, Qalyubia governorate, Egypt, (located at 30° 6′ 54′′ N and 31° 14′ 51′′ E) to improve squash drought tolerance by silicon foliar applications. Squash Hybrid SAKATA- Aziad ® transplants were sown in an open field (soil pH 7.82, Clay-loamy, E.C. 0.91 ds/m, HCO_3. 0.60 meq/L, Na^+ 2.71 meq/L, Ca^{+2} 3.30 meq/L, Mg^{+2} 2.90 meq/L, K^+ 0.19 meq/L, Cl^- 5.00 meq/L, SO_4^{-2} 3.40 meq/L) in the experimental unit area, which consisted of four rows, each row was 3.50 m length and 80 cm width and the distance was 70 cm apart between transplants on one side were sown on 14 February and harvested on 25 March in the tow growing seasons of 2019 and 2020. During soil preparation, 150 kg of calcium super phosphate (15.5% P_2O_5) and 50 kg sulphur, the recommended amounts of ammonium sulphate (20.5% N) at 200 kg and potassium sulphate (48% K_2O) at 100 kg were added according to the recommendations of the Egyptian Ministry of Agriculture during the two growing seasons. The meteorological data of the study region throughout 2019 and 2020 seasons are exhibited in Table 1.

2.2. Experimental design and treatments

The experiment was arranged in a split-plot design with six treatments. Deficit irrigation treatment in the main plot as 80% and 50% Water Holding Capacity (WHC), which estimated by the following equation of the water (moisture) content of soil mass WHC% = $[mass_{saturated} - mass_{oven\ dry}] / mass_{oven\ dry} * 100$ (Brischke and Wegener, 2019). Silicon treatments as monosilicic acid (H_4SiO_4) were arranged in the subplots, which were sprayed at three times with 10 days intervals starting at the 10th day from transplanting. The complete set of treatments are as follow:

1) 80% WHC + distill water
2) 80% WHC + Si at 2000 ppm
3) 80% WHC + Si at 4000 ppm
4) 50% WHC + distill water
5) 50% WHC + Si at 2000 ppm
6) 50% WHC + Si at 4000 ppm

2.3. Vegetative growth characteristics

After 40 days from transplanting (DAT), four plants were randomly chosen from each plot to measure the shoot height, shoot fresh and dry weights/plant, number of leaves/plant, leaf area and stem diameter. The average leaf area was measured by the planimeter. Leaf chlorophylls SPAD was measured by digital chlorophyll meter, Minolta SPAD-502, (Minolta Company, Japan) for the fully expanded mature leaves of 5 plants. A percentage of the relative water content (RWC) was determined by the equation RWC = (Fresh weight − Dry weight) / (Turgid weight − Dry weight) ∗ 100 (Schonfeld et al., 1988).

2.4. Flowering and yield characteristics

Four plants from each plot labelled from sowing for recording the number of female, male and total flowers/plant and the sex ratio % was calculated according to the following formula, Sex ratio = number of male flowers/female flowers. Number of total fruits/plant, total fruits yield/plant, average fruit weight, average fruit length, average fruit diameter, fruits yield/plant, total yield/hectare and fruit shape index according to the following equation, Fruit Shape Index (FSI) = fruit length/fruit diameter according to Shehata and Abdelgawad (2019).

2.5. Organic osmolytes (total free amino acids, proline, total soluble proteins and total soluble sugars)

Total free amino acids (TFAA) concentrations in squash leaves were analyzed using the ninhydrin method and measured using a spectrophotometer (Mapada UV 1200) at 570 nm described by Swamy (2008). The method of the aqueous sulfosalicylic acid and acid ninhydrin reagent as described by Bates et al. (1973) was used to assay the concentrations of proline in leaves using a spectrophotometer at 520 nm. Concentration of total soluble protein (TSP) was determined in leaves of the squash using the method described by Bradford (1976), reading at 595 nm using bovine serum albumin as the standard. Total soluble sugars (TSS) of squash leaves were extracted in 80% hot ethanol and measured spectrophotometrically by anthrone reagent at 620 nm using glucose standard according to Sadasivam and Manickam (2010).

2.6. Catalase (CAT), superoxide dismutase (SOD), peroxidase (POD) and polyphenol oxidase (PPO) assessment

At 30 days after transplanting, samples of leaves were harvested and extracted in 5 mL of cold 50 mM K-phosphate buffer (pH 7.0), containing 1 mM EDTA, 1 mM phenylmethylsulfonyl fluoride, and 1% PVPP, to prepare the enzyme extract. The activity of CAT enzyme was determined according to the method described by Aebi (1984), which depends on monitoring the ability of CAT to catalyze a specific amount of H_2O_2 for 60 s at 240 nm. The specific activity of SOD enzyme

Table 1
Meteorological data from the experimental site collected during the 2019 and 2020 growing seasons.

Year	2019				2020			
Month	Temperature (°C)		Precipitation (mm)	Relative humidity (%)	Temperature (°C)		Precipitation (mm)	Relative humidity (%)
	Max	Min			Max	Min		
Jan	21.2	5.5	1	76	19.7	4.6	5	71
Feb	25.5	6.4	3	69	22.5	7.5	2	66
Mar	33.2	9.2	6	61	28.7	11.2	8	56
Apr	37.8	12.5	4	59	32.9	14.8	5	50

was determined according to Beauchamp and Fridovich (1971) by nitro-blue tetrazolium photochemical assay. Peroxidase activity was measured at 417 nm in the presence of H_2O_2 and enzyme extract using *o*-phenylenediamine as a chromogenic indicator (Vetter et al., 1958). The method of Oktay et al. (1995) was used to determine the activity PPO, using catechol as substrate in phosphate buffer.

2.7. Statistical analysis

Data of the two seasons 2019 and 2020 were arranged and statistically analyzed using CoStat software (version 6.4, CoHort Software, USA). Analysis of variance (ANOVA) was used to test for significant differences among water irrigation levels and foliar applications with Si at $P < 0.05$ followed by Tukey's HSD test according to the method described by Gomez and Gomez (1984).

3. Results

3.1. Vegetative growth

The critical vegetative growth parameters affected by drought stress are shoot height, number of leaves/plant, shoot FW and shoot DW, which are presented in Table 2 reveal that drought stress expressed as irrigation at 50% of WHC significantly negative altered vegetative growth characteristics include shoot height, shoot FW and DW, leaves number per plant, average leaf area and stem diameter compared with the control (well water plants) grown under the irrigation at 80% of WHC. Among all the vegetative growth parameters, the most drought-affected parameters were shoot FW and shoot height which recorded a decrease reached 2.4 and 1.45 lower than its values in well water plants, respectively.

Silicon foliar applications at rates 2000 and 4000 ppm significantly increased the values of shoot height, shoot FW, shoot DW and stem diameter compared with the control plants sprayed with distilled water (0 ppm Si) either under the well water or drought treatments. Leaves number per plant increased significantly under well water treatment and the average leaf area under drought conditions with Si applications (Table 2). The most effective treatment in this study is the foliar application with Si at 4000 ppm recorded the highest significant values for all growth parameters either under well water and drought stress treatments. The most influenced by Si treatments were shoot fresh and dry weights, which recorded an increase of 2,00, 2.44 and 1.98 and 1.57 under well water and drought conditions, respectively. Opposite, the lowest significant values for all growth traits were produced under drought without Si treatment (Table 2).

3.2. Flowering

Flowering stage is a very sensitive stage to drought stress. It is known that the number of fruits per plant will never be over the number of flowers per plant, so the most important traits that express the flowering under drought or normal irrigation conditions are both number of flowers and fruits per plant. Water deficit stress caused a significant reduction in the number of fame flowers per plant, number of total flowers per plant, which consequently badly affected fruits number per plant and fruit setting % (Table 3). Fruits number per plant and fruit setting % reduced significantly under drought stress compared to its control. Meanwhile, the opposite trend was recorded for sex ratio (♂/♀ flowers). The number of male flowers/plant was insignificantly decreased in drought-stressed plant 13.70 flowers compared with 14.80 flowers in well water plants (Table 3). The number of female flowers, fruits number per plant and fruit set was one of the most affected traits by drought, which decreased to 66.70, 57.00 and 85.14% lower than well water plants, respectively. In this regard, the lowest values for all flowering characteristics as total, male and female flowers number per plant, fruits number per plant and fruit set were shown in plants grown under drought without Si.

The foliar applications with Si at 2000 and 4000 ppm significantly increased the number of fame flowers, total flowers/plant and fruits number/plant compared with the control level (0 ppm Si) either under 80 or 50% of WHC. In this connection, foliar spray with 4000 ppm Si significantly increased the number of male flowers/plant and fruit setting either under well water or drought treatments. Under drought stress treatment, using Si at rates 2000 and 4000 ppm led to a significant increase in the fruit setting %. Although spraying the highest level of Si (4000 ppm) recorded the highest values for flowering characteristics, number of fruits per plant and fruit setting % either under the well water or drought treatments (Table 3). The number of fame flowers, fruits number per plant and fruit set were the most affected traits by Si applications which achieved an increase reached to 1.37, 1.39 and 1.45, 1.70 and 1.07 and 1.21 over it controls. However, the highest sex ratio (♂/♀) was 0.84 resulted in drought-stressed plants without Si (Table 3). Thus, energizing the flowering, number of female flowers, fruit setting and total fruits yield by reducing the number of male flowers and sex ratio (♂/♀).

3.3. Yield

Fruit length and fruit diameter presented in Table 4 show that drought led to a significant reduction in these parameters compared with well water plants. Exogenously foliar supplied Si at 2000 and

Table 2

Effect of silicon foliar applications on vegetative growth characteristics of squash plant under water deficit during 2019/2020 seasons (main of two seasons).

Irrigation (I)	80% WHC	50% WHC	Mean	80% WHC	50% WHC	Mean	80% WHC	50% WHC	Mean
Treatments (T)	Shoot height (cm)			Shoot fresh weight (g)			Shoot dry weight (g)		
Control	43.9 ± 2.4 b	30.3 ± 1.6 c	37.1 B	401.8 ± 26.8 c	196.7 ± 20.8 d	299.3C	56.1 ± 7.0 b	39.3 ± 3.5 c	47.7 B
2000 PPM Si	55.3 ± 2.4 a	45.0 ± 2.8 b	50.1 A	683.0 ± 27.0 b	399.2 ± 34.0 c	541.1 B	99.2 ± 9.4 a	55.1 ± 5.7 b	77.2 A
4000 PPM Si	62.2 ± 2.9 a	45.6 ± 3.8 b	53.9 A	803.3 ± 27.5 a	481.8 ± 29.3 c	642.6 A	111.2 ± 9.2 a	61.7 ± 6.2 b	86.4 A
Mean	53.8 A	40.3 B		629.4 A	359.2 B		88.8 A	52.0 B	
P value I			0.0200			0.0008			0.0056
P value T			0.0000			0.0000			0.0001
P value I × T			0.0083			0.0370			0.0151
	Leaves number plant^{-1}			Average leaf area (cm^2)			Stem diameter (cm)		
Control	27.0 ± 2.0 b	19.7 ± 1.5 c	23.3 B	300.2 ± 10.4 bc	248.2 ± 10.8 d	274.2C	1.51 ± 0.13 c	0.91 ± 0.23 d	1.21C
2000 PPM Si	35.3 ± 2.1 a	24.3 ± 1.5 bc	29.8 A	336.1 ± 19.0 b	286.4 ± 15.0 c	311.3 B	2.21 ± 0.23 ab	1.74 ± 0.20 bc	1.97 B
4000 PPM Si	37.7 ± 2.5 a	27.7 ± 2.5 b	32.7 A	430.2 ± 21.3 a	297.7 ± 10.9 bc	363.9 A	2.68 ± 0.32 a	2.18 ± 0.16 ab	2.43 A
Mean	33.3 A	23.9 B		355.5 A	277.4 B		2.13 A	1.61 B	
P value I			0.0091			0.0004			0.0142
P value T			0.0002			0.0000			0.0001
P value I × T			0.0342			0.0019			0.0087

Means (±SD) followed by different letters are significantly different at $P < 0.05$ level; Tukey's HSD test. The uppercase letters to compare means of overall treatment or irrigation level, whereas the lowercase letters for the interaction between treatments and irrigation levels. WHC = Water Holding Capacity.

Table 3
Effect of silicon foliar applications on flowering characteristics of squash plant under water deficit during 2019/2020 seasons (main of two seasons).

Irrigation (I)	80% WHC	50% WHC	Mean	80% WHC	50% WHC	Mean	80% WHC	50% WHC	Mean
Treatments (T)	Number of male flowers			Number of female flowers			Sex ratio (♂/♀)		
Control	14.8 ± 1.3 bc	13.7 ± 1.1 c	14.3 B	24.3 ± 1.3 c	16.2 ± 1.0 e	20.3C	0.61 ± 0.02 c	0.84 ± 0.03 a	0.73 A
2000 PPM Si	16.7 ± 1.0 ab	15.0 ± 1.0 bc	15.8 B	29.8 ± 1.8 b	20.0 ± 1.0 d	24.9 B	0.56 ± 0.01 c	0.75 ± 0.01 b	0.65 B
4000 PPM Si	18.5 ± 1.5 a	17.5 ± 1.0 a	18.0 A	33.2 ± 1.3 a	22.5 ± 1.5 cd	27.8 A	0.56 ± 0.04 c	0.78 ± 0.01 ab	0.67 B
Mean	16.7 A	15.4 A		29.1 A	19.6 B		0.57 B	0.79 A	
P value I			0.1904			0.0015			0.0026
P value T			0.0018			0.0001			0.0045
P value I × T			0.0241			0.0040			0.0413
	Number of total flowers			Fruits number Plant^{-1}			Fruit setting %		
Control	39.2 ± 2.5 c	29.8 ± 2.0 d	34.5C	18.8 ± 1.0 c	10.7 ± 1.3 e	14.8C	77.4 ± 1.6 bc	65.9 ± 4.1 d	71.6C
2000 PPM Si	46.5 ± 2.8 b	35.0 ± 2.0 c	40.7 B	24.0 ± 1.5 b	14.8 ± 0.8 d	19.4 B	80.5 ± 0.5 ab	74.2 ± 0.7 c	77.3 B
4000 PPM Si	51.7 ± 2.3 a	40.0 ± 2.5 c	45.8 A	27.3 ± 1.5 a	18.0 ± 1.0 c	22.7 A	82.4 ± 1.5 a	80.0 ± 0.9 ab	81.2 A
Mean	45.8 A	34.9 B		23.4 A	14.5 B		80.1 A	73.4 B	
P value I			0.0041			0.0025			0.0209
P value T			0.0003			0.0000			0.0001
P value I × T			0.0007			0.0074			0.0115

Means (±SD) followed by different letters are significantly different at $P < 0.05$ level; Tukey's HSD test. The uppercase letters to compare means of overall treatment or irrigation level, whereas the lowercase letters for the interaction between treatments and irrigation levels. WHC = Water Holding Capacity.

4000 ppm significantly increased the fruit length and fruit diameter under water deficit treatment. However, the foliar application with silicon levels didn't significantly affect fruit length and fruit diameter under well water conditions compared with drought-stressed plants. In addition, spray Si at 4000 ppm under 80% of WHC significantly increased the fruit diameter as compared to it controls. Therefore, the high level of Si recorded the highest values in fruit length and fruit diameter 16.2 and 14.1 cm and 3.83 and 3.53 cm either under well water and drought stress treatments, respectively. The opposite trend showed for the highest significant values in the fruit shape index (FSI) 4.74 in drought-stressed plants without Si application (Table 4) because drought significantly reduced values of the fruit diameter.

Drought stress significantly reduced the average fruit fresh weight, fruits yield plant^{-1} and total fruits yield (ton ha^{-1}) compared with well water plants (Table 4). Water deficit decreased fruit FW, fruits yield plant^{-1} (g) and total fruits yield (ton ha^{-1}) 73.5%, 41.6% and 41.5% compared with well water plants, respectively. Silicon foliar applications achieved significant increases in average fruit fresh weight, fruits yield plant^{-1} and total fruit yield/ha^{-1} either under well water and drought stress treatments compared with controls, except for the fruit FW recorded a significant increase by Si at 2000 ppm treatment under well water. Additionally, the highest values of all yield traits were recorded by using the high level of Si either under well water or drought stress conditions whereas, the lowest values of average fruit fresh weight, fruits yield plant^{-1} (g) and total fruits yield (ton ha^{-1}) were produced under drought without Si (Table 4). Overall, higher yields produced by Si foliar applications at 2000 and 4000 ppm than its controls (Table 4) related to that silicon encourage higher flowering (Table 3), higher chlorophylls, relative water, total amino acids, soluble proteins and total soluble sugars as well as stimulating SOD, CAT, POD and PPO enzymes activity (Figs. 1-3).

3.4. Biochemical and physiological aspects

Results presented in Fig. 1A and B clearly show that drought gave a significant decrease for total chlorophylls values (SPAD) and leaf relative water content (LRWC) % compared to the well water treatment. In contrast, silicon foliar applications markedly increased the chlorophylls and LRWC either under well water or water deficit conditions in comparison to the respective controls. Using the high level of Si (4000 ppm) recorded the highest significant values of total chlorophylls 45.53 and 39.44 and LRWC 82.69 and 71.59% either under well water or water deficit conditions, respectively. The lowest values of total chlorophylls and LRWC were 28.25 and 49.64% showed in drought-stressed plants without Si. Data show a positive correlation between the LRWC with shoot fresh and dry weights under drought stress and Si indicating that the plant water status improves drought tolerance.

Table 4
Effect of silicon foliar applications on fruits yield characteristics of squash plant under water deficit during 2019/2020 seasons (main of two seasons).

Irrigation (I)	80% WHC	50% WHC	Mean	80% WHC	50% WHC	Mean	80% WHC	50% WHC	Mean
Treatments (T)	Fruit length (cm)			Fruit diameter (cm)			Fruit shape index		
Control	14.7 ± 0.8 ab	12.3 ± 0.6 c	13.5 B	3.38 ± 0.17 bc	2.62 ± 0.28 d	3.0 B	4.34 ± 0.06 b	4.74 ± 0.42 a	4.54
2000 PPM Si	15.6 ± 0.8 ab	14.0 ± 0.5 b	14.8 AB	3.67 ± 0.18 ab	3.21 ± 0.18 c	3.4 A	4.24 ± 0.09 b	4.36 ± 0.08 b	4.29
4000 PPM Si	16.2 ± 1.0 a	14.1 ± 0.9 b	15.1 A	3.83 ± 0.16 a	3.33 ± 0.15 bc	3.6 A	4.22 ± 0.14 b	4.23 ± 0.07 b	4.23
Mean	15.5 A	13.5 B		3.6 A	3.1 B		4.26	4.44	
P value I			0.0151			0.0339			NS
P value T			0.0165			0.0012			NS
P value I × T			0.0141			0.0017			0.0506
	Average fruit fresh weight (g)			Fruits yield Plant^{-1} (g)			Fruits yield (t ha^{-1})		
Control	52.4 ± 2.3 b	38.5 ± 2.5 d	45.5C	985 ± 13 c	410 ± 47 e	698C	19.5 ± 0.3 c	8.1 ± 0.9 e	13.8 C
2000 PPM Si	56.3 ± 2.5 ab	44.8 ± 2.3 c	50.6 B	1349 ± 40 b	666 ± 67 d	1007 B	26.7 ± 0.8 b	13.2 ± 1.3 d	20.0 B
4000 PPM Si	60.0 ± 3.0 a	51.7 ± 3.1 b	55.8 A	1638 ± 97 a	931 ± 99 c	1284 A	32.5 ± 1.9 a	18.5 ± 2.0 c	25.5 A
Mean	56.2 A	45.0 B		1324 A	669 B		26.2 A	13.3 B	
P value I			0.0024			0.0057			0.0057
P value T			0.0015			0.0000			0.0000
P value I × T			0.0016			0.0084			0.0085

Means (±SD) followed by different letters are significantly different at $P < 0.05$ level; Tukey's HSD test. The uppercase letters to compare means of overall treatment or irrigation level, whereas the lowercase letters for the interaction between treatments and irrigation levels. WHC = Water Holding Capacity; NS = Not significant.

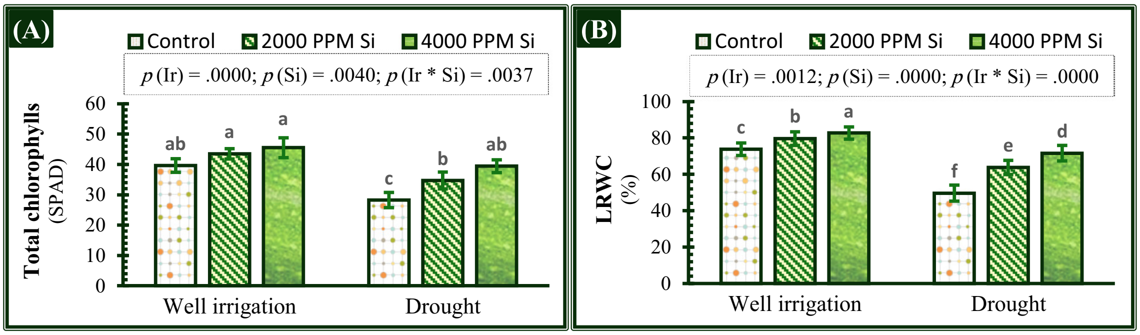

Fig. 1. Effect of silicon foliar applications on the content of total chlorophylls (SPAD) and leaf relative water content in squash plants under deficit irrigation condition during 2019 and 2020 seasons (main of two seasons).

Concentrations of total free amino acids (TFAA), proline, total soluble proteins (TSP) and total soluble sugars (TSS) in leaves of squash plants were presented in Fig. 2A-D observed that drought significantly increased total free amino acids, proline and total soluble sugars concentrations whereas, the total soluble proteins significantly reduced when compared with well water plants. Regarding the foliar applications with Si treatments improved the concentrations of organic osmolytes and total soluble proteins under the well water treatment. The same trend for leaf TSP and TSS concentrations under drought (Fig. 2C and D). Silicon foliar treatments led to a significant decrease in total free amino acids and proline concentrations under drought stress (Fig. 2A and B).

The highest significant concentration of total free amino acids and proline in leaves 15.1 and 6.47 mg g^{-1} FW was recorded under drought treatment, while the lowest concentrations of both 6.01 and 2.21 mg g^{-1} FW resulted in well water plants without Si applications, respectively (Fig. 2A and B). In this regard, using Si at 4000 ppm as foliar spray produced the highest concentrations of total soluble proteins (19.75, 12.22 mg g^{-1} FW) and total soluble sugars (15.54 and 16.71 mg g^{-1} FW) in leaves of the well water and drought-stressed plants, respectively. Meanwhile, the lowest concentration for total soluble protein 6.68 mg g^{-1} FW was recorded in drought-stressed plants without Si. The lowest values of total soluble sugars 7.15 mg g^{-1} FW resulted in well water treatment without Si.

Regarding, activities of enzymatic antioxidants such as superoxide dismutase (SOD), catalase (CAT), peroxidase (POD) and polyphenol oxidase (PPO) presented in Fig. 3 reveal that water deficit treatment significantly increased SOD, POD and PPO activities compared with well water plants (Fig. 3A, C and D) which improve squash drought tolerance via maximizing activities of these enzymes. In contrast, drought stress gave the lowest significant activity 2.35 units mg^{-1} protein of CAT enzyme, compared with rest treatments (Fig. 3B). Foliar applications with Si at rates 2000 or 4000 ppm increased the activity of SOD, CAT, POD and PPO enzymes either under the well water and drought treatments especially using the high rate of Si recorded the highest levels in these enzymes. Higher SOD activity is the first one in the enzymatic antioxidant defense system of squash plants grown under drought stress 73.09 units mg^{-1} protein.

4. Discussion

In the present study, the hamper effects of water deficit (50% of WHC) on the vegetative growth, flowering, physiological, biochemical constituents and yield productivity of squash in compared to the

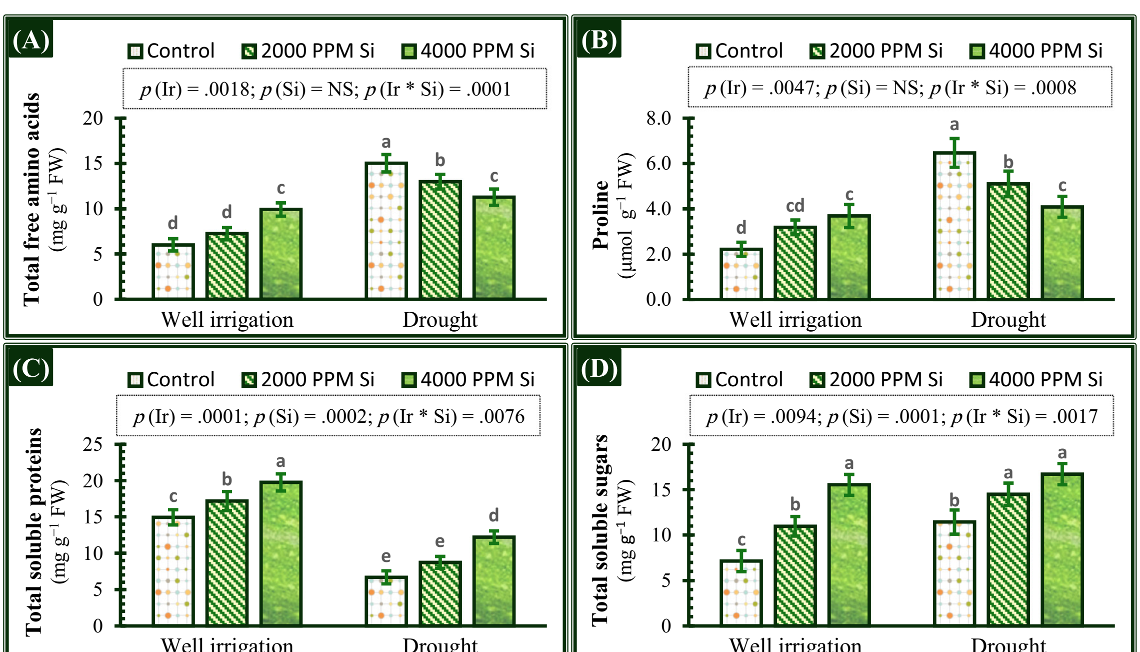

Fig. 2. Effect of silicon foliar applications on concentrations of free amino acids, proline, total soluble proteins and total soluble sugars in the leaves of squash plants under deficit irrigation condition during 2019 and 2020 seasons (main of two seasons).

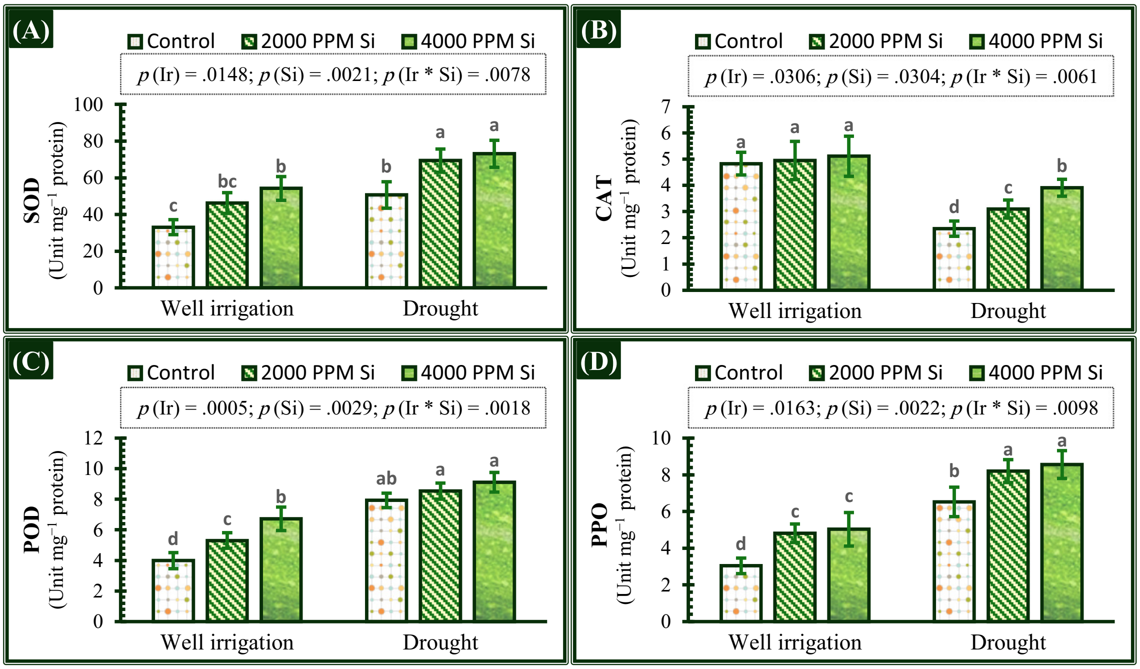

Fig. 3. Effect of silicon foliar applications on SOD, CAT, POD and PPO activity in the leaves of squash plants under deficit irrigation condition during 2019 and 2020 seasons (main of two seasons).

respective control grown under well water (80% of WHC) in both tested seasons 2019 and 2020. These results were explained by many authors who suggested that drought inhibited morphological, physiological and metabolic pathways (Farooq et al., 2009; Kapoor et al., 2020; Rao et al., 2016; Yavaş and Ünay, 2017).

Our results indicate that water deficit led to a significant reduction for plant shoot height, shoot FW and DW, leaves number per plant, average leaf area and the stem diameter (Table 2) are in agreement with the literature studies of (Abd El-Mageed et al., 2016; Abd El-Mageed et al., 2017) indicating that drought stress reduced growth characteristics of squash plants. Similar results were also observed on other vegetable crops like snap bean by Salim and Abou El-Yazied (2015), pea by Osman (2015), bean by Kabay (2020) and tomato plants by Ors et al. (2021). Inhibited vegetative growth of squash under drought stress (Table 2) may be due to the reduction in photosynthetic rate by drought (Ma, 2004; Ors et al., 2021). Therefore, Abd El-Mageed and Semida (2015) obtained the highest values in plant height, number of leaves per plant, total dry weights/plant and leaf area index under adequate water irrigation. Enhancing the vegetative growth of well water plants may be due to improvements in chlorophylls and LRWC (Fig. 1).

Data presented in Tables 3 and 4 summarized that drought inhibits flowering consequently decreases the fruit setting and fruit yield compared with well water plants. The same effects have been shown in squash plants by Shehata and Abdelgawad (2019) and tomato plants by Sivakumar and Srividhya (2016). In this regard, Salim et al. (2019) reported that flowering considers a hypersensitive stage to drought. Moreover, water deficit reduced days to flowers initiation and increased flowers abscission percentage of tomato plants. Additionally, drought stress inhibited nutrient uptake, translocation and minerals relations within plants which obstructed plant growth, development, flowering and yield (Sivakumar and Srividhya, 2016). Moreover, all biological processes, from germination to reproduction and maturity, are negatively affected by water deficit (Kapoor et al., 2020).

Drought significantly reduced the total chlorophylls, LRWC %, total soluble proteins and CAT enzyme activity, whereas improved considerably the content of total amino acids, total soluble sugars, proline and activity of SOD, POD and PPO enzymes (Fig. 1-3). Similar studies suggested that water deficit decreased leaf photosynthetic pigments, relative water content and increased proline and soluble sugars concentrations of squash plants (Abd El-Mageed and Semida, 2015; Abd El-Mageed et al., 2016). Also, Sivakumar and Srividhya (2016) indicated that drought decreased chlorophylls and soluble protein of tomato. Results are in agreement with Farooq et al. (2009); Rizwan et al. (2015); Abd El-Mageed et al. (2017); Kapoor et al. (2020); Osman et al. (2021b) that drought-tolerant plants induced biosynthesis higher osmolites and enzymatic antioxidant activities in plant defense system to protect plant organelles.

On the contrary, spraying Si at 2000 and 4000 ppm had synergism impacts on the vegetative growth, flowering and yield traits of squash plants either under well water or water deficit treatments (Tables 2, 3 and 4). Foliar silicic acid is only effective at very low levels during the vegetative growth stage as a biostimulant enhancing plant growth starting from root growth resulting in higher nutrients absorption (Laane, 2016). Enhancing the vegetative growth characteristics (Table 2) by Si foliar applications related to improving chlorophylls, LRWC (Fig. 1), non-enzymatic (Fig. 2) and enzymatic antioxidants (Fig. 3) either under well water or water deficit conditions in compared to the respective controls (sprayed with distilled water). Our data are similar to results obtained by Shehata and Abdelgawad (2019), who reported that the plant length, number of leaves/plant, leaf area/plant, leaves fresh and dry weights/plant, flowering, sex ratio, fruit length, fruit diameter, fruit shape index, fruit weight and total fruits yield of squash were stimulated by potassium silicate treatments. In the same context, higher significant fruits yield of tomato plants correlated with better flowering and better fruit set with Si than control plants (Jarosz, 2014). These data are supported with the opinions recommended Si for alleviating drought stress (Kaushik and Saini, 2019; Laane, 2018; Rizwan et al., 2015; Sahebi et al., 2015; Savvas and Ntatsi, 2015).

Positive significant impacts of Si treatments on the vegetative growth, flowering and yield traits of squash in the present study might be attributed to silicon reducing the transpiration and maintaining cell membranes stability (Ma, 2004). Moreover, Savvas and Ntatsi (2015) reported that Si retards senescence processes and improves antioxidant systems. Also, Si improves cell wall strength, water uptake and photosynthesis (Yavaş and Ünay, 2017). Therefore, Si promotes cells division and cells enlargement, which controls the physiological processes and metabolic activities such as membrane permeability, water and nutrients uptake, photosynthesis, transpiration and endogenous phytohormones biosynthesis (Thorne et al., 2020; Zhu and Gong, 2013).

In analyzing data of the present study, we can summarize a trend towards improved growth characteristics, flowering and yield traits (Tables 2-4) in response to the foliar Si applications are correlated with the superior capacity of leaf chlorophylls, LRWC, organic osmolytes i.e. total soluble sugars and total soluble proteins as well as, higher activities of the SOD, CAT, POD and PPO enzymes than its controls are beneficial in improving plant tolerance to drought. In this concern, Rizwan et al. (2015) reported that, Si increased chlorophylls pigments, leaf water content, compatible solutes and antioxidant enzymes. In this regard, Si has a positive effect on chlorophylls (Thorne et al., 2020). Plants can overcome injurious effects of water deficit stress by Si applications through inducing various metabolic pathways, biochemical processes and accumulation of higher organic solutes, higher leaf water content and enzymatic antioxidants activity (Bakhat et al., 2018; Osman et al., 2021a; Sahebi et al., 2015; Savvas and Ntatsi, 2015; Zhu and Gong, 2013).

5. Conclusion

Water deficit inhibits the vegetative growth, flowering, yield productivity, LRWC%, chlorophylls and total soluble proteins. Foliar applications of silicon at 2000 or 4000 ppm helping squash plants to correct the harmful impacts of drought stress. Spraying the high level of Si was the best treatment under stressed and unstressed conditions.

Declaration of competing interest

All the authors declare there is no conflict of interest.

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