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Influence of Silicon on Tuberose Plants under Drought Conditions

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ABSTRACT

The present investigation was carried out to study the effect of using Diatomite (DDM contain 89% of SiO_2) on the morphological, floral and chemical characteristics, in addition to the anatomical studies on leaves of tuberose plants (*Polianthes tuberosa* L.) grown under different water stress levels (100, 80, 60 and 40% FC). The results revealed that, a gradual increasing in days to sprouting was found with increasing the drought stress. The maximum increment was at 40% FC. The treated bulbs with DDM at 80 % FC reached the sprouting time in a shorter period than those at the other FC levels. The values of stem characters; length, diameter, fresh and dry weights were decreased from level 80% drought stress. The bulbs treated with DDM at 60 and 40% FC, the values of stem length and diameter were insignificantly increased than their controls. Highly significant increment in the stem fresh and dry weights was also occurred. The values of number of days to flowering and to cut the spike were increased by increasing the drought stress. Meanwhile, the opposite trend was occurred with the other floral characters; spike length, number of florets/spike, floret fresh and floret dry weights. The tuberose plants treated with DDM at 80 % FC reached the blooming stage significantly earlier than any other treatments, consequently the number of days to cut the spike was the shorter. Total chlorophylls and protein contents decreased gradually with increasing the stress condition. The maximum reduction was with40% FC. The bulbs treated with DDMat80% FC, led to slightly increment in the previous two characters. Also treated the tuberose plants with DDM slightly increased the carbohydrates content. Proline content increased as the drought stress increased either in the treated or untreated plants with DDM. The treated plants with DDM decreased the proline content, in contrary the Ca% in leaves was increased. All leaf tissue measurements showed a remarkable reduction as the drought stress levels increased. DDM application had slightly increased in the leaf measurements. Moreover, treated the plant by DDM at 60% FC increased the mean number of stomata. All stomata patterns on both leaf surfaces were anomocytic. It can be recommended to use Diatomite with 80% FC to produce high-quality flowers. To delay flowering, Diatomite can be used with irrigation field capacity 60%.

Key words: Tuberose, water stress, silicon, floral and anatomical characters.

Introduction

Tuberose (*Polianthes tuberosa* L.) is a perennial bulbous plant of the Agavaceae family, and is an ornamental bulbs plant native to Mexico (Benschop, 1993). It is one of a common cultivated flowering bulb used as garden plant with a great demand for its attractive tall spikes with fragrant cut flowers and also for extraction of its highly valued natural flower essential oils (Jawaharlal *et al*., 2006). In India and France, tuberose plant is widely cultivated as a source of essential oils for the perfume industry (Majid *et al*., 2012)

The effect of water stress on growth and development of bulbous plants, has been investigated to a much lesser extent than the other flowering crops.

Silicon promoting the growth of various higher plant species. It simulate the growth either indirectly by accumulation the Si in the epidermal tissue of the plant which provide defense against insect and fungal attacks and protect the plant against the effects of heavy metals contamination, or directly by originating both morphological changes and physiological processes in plants. It seems that silicon is involved directly or indirectly in cell metabolism as well, although in most cases the mode of action is still unclear (Liang *et al*., 2003; Zhu *et al*., 2004 and Sacała, 2009). Moreover,

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Hattori *et al*. (2005) mentioned that, silicon (DDM contain 89% of SiO_2) is known to increase drought tolerance of plants by maintaining plant water balance and photosynthetic activity. In addition, silicon stimulate the plant resistance to unsuitable environmental conditions, i.e. low temperature or water deficit.

The present investigation was conducted to study the effect of using Diatomite (DDM contain 89% of SiO_2) on the morphological, floral and chemical characteristics, in addition to the anatomical studies on leaves and stomata of tuberose plants (*Polianthes tuberosa* L.) grown under different water stress levels.

Materials and Methods

The present work was carried out at the Experimental Farm, Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, Egypt during the two successive seasons; 2014 and 2015 in order to ameliorate the effect of water stress conditions on growth and flowering of *Polianthes tuberosa* plants using DDM application (Table A).

Table A: Major elements in Diatomate (DDM) according to Abdalla (2011)

Major elements	SiO_2	Al_2O_3	Fe_2O_3	CaO	K_2O	MgO	Na_2O	TiO_2	H_2O
%	89.00	5.95	0.88	0.10	0.63	0.20	0.32	0.29	3.00

Bulbs of *Polianthes tuberosa* with 32mm diameter (about 30-40 g, weight) were obtained from a private farm at Menofia governorate. The bulbs split into two groups; one group was soaked in tap water for 2 h (control), the other group was soaked, as mentioned by Abdalla (2011), in 100 ppm Diatomite (DDM contain 89% of SiO_2) solution for 2 h. Bulbs were planted in the 1st season; 2014 at 12th April, and replicated in the 2nd one; 2015 at 24th April. Bulbs were planted in plastic pots; 35 cm high and 25 cm diameter, one bulb per pot, filled with a mixture of about 6 kg clay and clean sand (1:2 v/v). The untreated and treated bulbs were planted under different levels of field capacity (FC %); 100, 80, 60 and 40%. The pots were placed under field conditions. After 4 weeks of planting, the plants of Si treated bulbs were sprayed with DDM at 30 ppm, then the second spray was applied after a month from the first one. The control plants were sprayed with tap water. Tween-20 was added as a spreading agent for tested treatments. Each pot was received NPK (19:19:19) Crystalone at the recommended rates. The total watering time ranged from 135 to 150 days according to the harvest time with different FC levels. The plants irrigated twice every week. So, the total water for each treatment was differ based on the FC level as the total water was 96.90 L for 100% FC, 77.52 L for 80% FC, 58.40 L for 60% FC and 38.76 L for 40% FC

Data recorded:

The following characters were recorded:

Morphological characters:

Number of days to sprouting (days), stem length (cm), stem diameter (mm), stem fresh weight(g/plant), stem dry weight(g/plant), number of leaves/plant, leaf length (cm), leaf width (cm), leaves fresh weight(g/plant) and leaves dry weight(g/plant).

Floral characters:

Number of days to flowering (days), number of days to cut spike (days, when 2-3 basal florets were opened), spike length (cm), number of florets/spike, florets fresh weight (g/plant) and florets dry weight (g/plant).

Chemical characters:

1- Total Chlorophylls: fresh leaf samples were taken for measuring the total chlorophylls (SPAD) according to Netto *et al*. (2005).
2- Total carbohydrates (%/D.W) were determined according to Herbert *et al.* (2005).

3- Free proline (mg/g F.W) in the fresh leaves was determined according to the method described by Bates *et al.* (1973). The absorbance was measured at 520 nm using Bush and Lomb spectrophotometer (model spectranic 2000).
4- Nitrogen in dry leaves was determined using the modified micro-Kjeldhahl method described by Walinga *et al* (1995). Nitrogen was multiplied by 6.25 to calculate the crude protein % (A.O.A.C., 2000).
5- 5- Calcium (%) in the fresh leaves was determined using flame photometer apparatus according to Walinga *et al.* (1995).

Anatomical studies:

For the macroscopically studies, samples of 1 cm^2 were taken from the middle portion of the leaf blade including the midrib. Specimens were killed and fixed for at least 48 hs in F.A.A. (10 ml formalin, 5 ml glacial acetic acid and 85 ml ethyl alcohol 70%), washed in 50% ethyl alcohol, dehydrated in a normal butyl alcohol series, embedded in paraffin wax (melting point 56ºC).Sections of 20 microns, were cut by using rotary microtome and stained with crystal violet/erythrosine combination safranine-light green, cleared in xylem and mounted in Canada balsam (Nassar and El-Sahhar, 1998). Slides were microscopically examined and the measurements of different tissues were taken and averages of 10 readings were calculated. Photomicrographs were also taken.

Statistical analysis:

The experiment layout was designed as a completely randomized block with 2 factors including 8 treatments and 3 replicates, each replicate contain 4 plants. The first factor had 4 treatments including the control; the second factor had 2treatments. The collected data were subjected to a convenient statistical analysis by using MSTAT software mean separations were done according to Mead *et al* (1993). The least significant difference (L.S.D.) at 0.05 levels was calculated.

Results and Discussion

Growth parameters:

This study is devoted to investigate the effect of applying Diatomite (DDM contains 89% of SiO2) on the morphological, floral, chemical and anatomical characteristics of tuberose plants grown under different drought stress levels (100, 80, 60 and 40 FC%).Regardless the FC% effects on tuberose plant, data presented in Table (1) clearly showed that applying the DDM treatment increased the mean values of all the growth parameters studied compared to their controls, in both seasons. The only exception was the mean value of number of days to sprouting where it was decreased. On the other hand, under the different levels of FC, the mean value of number of days to sprouting was increased by increasing the drought stress levels, in the same time decreasing the mean values of stem characters; length, diameter, fresh and dry weights, in addition to the mean values of number of leaves/plant and leaf length. While, the values of the leaf width, fresh and dry weights were fluctuated through the different FC levels.

Concerning the interaction between the effect of DDM and different FC levels, on the growth parameters under different drought stress levels, it was obvious that there was a gradual increase in days to sprouting with increasing the drought stress in both seasons. The increments occurred with the highest drought level (40% FC) compared with control (100% FC) were 14.59% and 26.73% in both seasons, respectively. While, with60% FC these values were 10.38% and 17.44%, in the same order. By treating the bulbs grown under 60 and 80 % FC with DDM, the bulbs sprouted time in a shorter period than those under irrigation regime of 40 and 100% FC.

In both seasons, the values of stem characters of DDM non-treated plants (length, diameter, fresh and dry weight) were decreased with 60 and 40 % drought stress. It is also noticed that the values of these characters with80% FC were significantly increased compared to those with 100% FC. The values of stem characters of bulbs treated with DDM and grown under 60 and 80% FC, were significantly increased than their control. With80% FC, the highly significant increments occurred in the values of the stem fresh weight compared to their control were 19.80% and 17.27% in both seasons, respectively. While, these values with 60% FC were 34.82% and 30.69%, in the same order.

Quite similar results, to some extent, were obtained with the fresh and dry weights with slightly significant difference.

Table 1: Effect of Diatomite (DDM) on some morphological characters of tuberose plant under different field capacity (FC%) during the two successive seasons ; 2014 and 2015.

Field capacity (FC)% (A)*	1st season			2nd season		
	DDM (B)*		Mean (A)	DDM (B)*		Mean(A)
	Control	DDM		Control	DDM	
No. of days to sprouting						
100	29.67	29.00	29.34	20.58	19.83	20.21
80	31.00	28.00	29.50	21.50	18.92	20.21
60	32.75	28.83	30.79	24.17	19.42	21.80
40	34.00	31.58	32.79	26.08	22.25	24.17
Mean (B)	31.86	29.35		23.08	20.11	
LSD5%	A= 1.24	B= 0.88	AB=1.76	A= 0.85	B= 0.60	AB= 1.21
Stem length cm						
100	66.76	69.16	67.96	63.48	63.41	63.45
80	67.54	72.28	69.91	63.30	64.93	64.12
60	61.13	62.71	61.92	62.61	64.87	63.74
40	54.08	57.64	55.86	53.21	56.28	54.75
Mean (B)	62.38	65.45		60.65	62.37	
LSD5%	A= 2.85	B= 2.05	AB= 4.03	A= 2.52	B= 1.78	AB= 3.56
Stem diameter mm						
100	6.50	6.88	6.69	6.76	6.89	6.83
80	7.21	7.72	7.47	7.02	7.33	7.18
60	6.39	7.24	6.82	6.45	7.07	6.76
40	5.93	6.08	6.01	6.05	6.16	6.11
Mean (B)	6.51	6.98		6.57	6.86	
LSD 5%	A= 0.33	B= 0.23	AB=0.46	A= 0.28	B= 0.20	AB= 0.39
F.W of stem g/plant						
100	24.21	28.76	26.49	18.63	21.08	19.86
80	25.35	30.37	27.86	19.17	22.48	20.83
60	18.41	24.82	21.62	16.75	21.89	19.32
40	15.19	16.14	15.67	12.96	15.29	14.13
Mean (B)	20.79	25.02		16.88	20.19	
LSD 5%	A= 1.26	B= 0.89	AB= 1.78	A= 0.67	B= 0.48	AB= 0.95
D.W of stem g/plant						
100	2.21	2.78	2.50	2.20	2.35	2.28
80	2.80	3.59	3.20	2.26	2.81	2.54
60	1.97	3.16	2.57	2.24	2.66	2.45
40	1.59	2.03	1.81	1.81	1.82	1.82
Mean (B)	2.14	2.89		2.13	2.41	
LSD 5%	A= 0.14	B= 0.10	AB= 0.20	A= 0.08	B= 0.05	AB= 0.12

Concerning the effect of drought stress, the values of all leaf characters were decreased with 60 and 40% FC. While, these values with 80% FC were greater than those values with 100% FC. The values of all the characters with 80% FC exceeded those at the highest drought stress level (40% FC). These values were 19.81- 24.10% for leaf number, 15.42-14.65% for leaf length, 7.45-9.27% for leaf width, 20.01-29.33% for leaf fresh weight and 25.20-23.14% for leaf dry weight, in both seasons, respectively. By applying DDM, significant effects on the values of leaf characters compared to the control were occurred. The highly significant effects were noticed with the plants treated with DDM grown under 60% FC compared to its control, where these values were exceeded the control by 15.68-23.95% for the leaf number, 9.44-16.75% for the leaf length, 19.76-12.35% for the leaf width, 15.42-20.82% for the leaf fresh weight and 48.69-52.65% for the leaf dry weight in the 1st and 2nd seasons, respectively. The abovementioned data showed that the values of all growth parameters of tuberose plants treated with DDM were increased compared to the untreated ones. Thus, it could be stated that DDM had beneficial effects and overcome the harmful injury occurred by stress of FC (Table 2).

Table 2: Effect of Diatomite (DDM) on some morphological characters of tuberose plant under different field capacity (FC %) during the two seasons; 2014 and 2015.

Field capacity (FC)% (A)*	1st season			2nd season		
	DDM (B)*		Mean (A)	DDM (B)*		Mean (A)
	Control	DDM		Control	DDM	
No. of Leaves/plant						
100	18.09	20.20	19.15	20.21	20.83	20.52
80	19.11	19.55	19.33	20.39	22.33	21.36
60	18.24	21.10	19.67	19.79	24.53	22.16
40	15.95	18.52	17.24	16.43	19.52	17.98
Mean (B)	17.85	19.84		19.20	21.80	
LSD5%	A= 0.95	B=0.67	AB=1.34	A= 1.42	B=1.01	AB=2.01
Leaf length cm						
100	37.31	38.44	37.88	31.54	34.10	32.82
80	37.58	39.95	38.77	31.64	36.17	33.91
60	35.29	38.62	36.96	28.71	33.52	31.12
40	32.56	37.42	34.99	27.24	30.23	28.74
Mean (B)	35.69	38.61		29.78	33.50	
LSD5%	A= 3.71	B= 2.62	AB=5.24	A= 1.70	B= 1.20	AB=2.40
Leaf width cm						
100	1.68	1.78	1.73	1.53	1.84	1.69
80	1.73	1.79	1.76	1.65	2.67	2.16
60	1.67	2.00	1.84	1.62	1.82	1.72
40	1.61	1.64	1.63	1.51	1.89	1.70
Mean (B)	1.67	1.80		1.58	2.06	
LSD5%	A= 0.06	B= 0.04	AB=0.09	A= 0.08	B= 0.06	AB=0.12
F.W. of leaves g/plant						
100	21.14	27.22	24.18	19.18	23.80	21.49
80	35.45	37.71	36.58	26.94	32.88	29.91
60	32.89	37.96	35.42	21.18	25.59	23.39
40	29.54	29.07	29.31	20.83	25.34	23.09
Mean (B)	29.76	32.99		22.03	26.90	
LSD5%	A= 1.54	B=1.09	AB=2.18	A= 1.12	B=0.79	AB=1.59
D.W. of leaves g/plant						
100	2.96	3.96	3.46	2.57	2.99	2.78
80	3.13	4.15	3.64	3.01	3.93	3.47
60	3.06	4.55	3.81	2.83	4.32	3.58
40	2.50	3.75	3.13	2.42	3.15	2.79
Mean (B)	2.91	4.10		2.71	3.60	
LSD 5%	A= 0.18	B= 0.13	AB=0.26	A= 0.17	B= 0.12	AB=0.25

The obtained data were in agreement with Hwang *et al.* (2005). They stated that Si promotes the growth of various higher plant species. The simulation of growth by silicon may be either indirect; owing to the protective effects of Si against pathogens, or direct originating from implications of Si to both morphological changes and physiological processes in plants. It seems that silicon is involved directly or indirectly in cell metabolism as well, although in most cases the mode of action is still unclear (Liang *et al.*, 2003 and Zhu *et al.*, 2004). It is possible that Si could increase total GA content and increases shoot height (Reezi *et al.*, 2009).

Hattor*i et al*. (2005) mentioned that, silicon treatment (DDM, contain 86-89% of SiO_2) is known to increase drought tolerance of plants by maintaining plant water balance and photosynthetic activity. Moreover, Silicon is known for its stimulation of plant resistance to unfavorable environmental conditions, *i.e.* low temperature or water deficit. One of the mechanisms for supporting plants under water stress conditions is decreased transpiration resulting in reduction in water loss, but silicon also partakes in osmo-regulation, maintaining water status and adequate supply of nutrients (Sacala, 2009). Faraz *et al.*, (2007) reported that increased levels of Si (50-150g/kg soil) significantly increased plant biomass, plant weights and dry matter production under water stress at 50% FC. Si fertilization increases crop yield and quality and thus insures high production under severe biotic and abiotic stresses (Hou *et al.*, 2006 and Ali *et al.*, 2015).

Floral characters:

Regardless the FC% effects on tuberose plant, data in Tables (3) showed that applying the DDM treatment on tuberose plants increased the mean values of spike length, number of florets/spike, floret fresh and dry weights compared to their controls, in both seasons, but it decreased the number of days to flowering and to cut the spike. The opposite situation was occurred on the mean values of these characters under the different drought levels (100, 80, 60 and 40% FC).

Table 3: Effect of Diatomite (DDM) on some floral characters of tuberose plant under different field capacity (FC%) during the two s seasons; 2014 and 2015.

Field capacity (FC)% (A)*	1st season			2nd season		
	DDM (B)*		Mean (A)	DDM (B)*		Mean (A)
	Control	DDM		Control	DDM	
No. of days to flowering						
100	110.11	106.61	108.36	102.22	101.56	101.89
80	108.50	99.72	104.11	100.25	83.14	91.70
60	112.67	110.75	111.71	109.81	97.33	103.57
40	120.39	118.67	119.53	113.67	102.00	107.84
Mean (B)	112.92	108.94		106.49	96.01	
LSD5%	A= 5.07	B= 3.58	AB=7.17	A= 4.47	B= 3.16	AB=6.32
No. of days to cut spike						
100	137.58	132.09	134.84	134.44	132.72	133.58
80	136.61	118.54	127.58	12976	123.22	126.49
60	143.87	140.86	142.37	136.01	128.94	132.48
40	156.11	153.34	154.73	155.58	142.40	148.99
Mean (B)	143.54	136.21		138.95	131.82	
LSD 5%	A= 7.73	B=5.47	AB=10.94	A= 6.08	B=4.30	AB=8.60
Length of spike cm						
100	19.00	21.07	20.04	17.18	17.36	17.27
80	19.41	22.68	21.05	17.28	18.90	18.09
60	17.75	18.60	18.18	15.96	16.71	16.34
40	12.32	12.48	12.40	11.56	12.44	12.00
Mean (B)	17.12	18.71		15.50	16.35	
LSD5%	A= 0.89	B= 0.63	AB=1.26	A= 0.74	B= 0.53	AB=1.05
No. of florets/spike						
100	11.45	12.11	11.78	9.13	10.11	9.62
80	11.82	12.25	12.04	9.41	10.31	9.86
60	9.89	10.73	10.31	8.98	9.60	9.29
40	6.60	7.72	7.16	6.15	7.38	6.77
Mean (B)	9.94	10.70		8.42	9.35	
LSD5%	A= 0.62	B= 0.43	AB=0.87	A= 0.29	B= 0.21	AB=0.41
F.W. of florets g/plant						
100	21.44	30.57	26.01	20.52	24.34	22.43
80	29.02	35.94	32.48	22.90	24.94	23.81
60	21.10	29.66	25.38	17.35	19.13	18.24
40	13.64	14.97	14.31	10.80	11.65	11.23
Mean (B)	21.30	27.79		17.84	20.02	
LSD5%	A= 1.78	B= 1.26	AB= 2.52	A= 0.94	B= 0.64	AB= 1.33
D.W. of florets g/plant						
100	2.24	2.89	2.57	1.77	2.52	2.15
80	2.86	4.22	3.54	2.10	3.40	2.75
60	2.11	2.86	2.49	1.99	2.46	2.23
40	2.08	1.36	1.72	1.52	2.39	1.96
Mean (B)	2.32	2.83		1.85	2.69	
LSD5%	A= 0.14	B= 0.10	AB= 0.20	A= 0.14	B= 0.11	AB= 0.21

The effect of DDM on the floral characters of tuberose plant grown under different drought stress levels, was obvious on the values of number of days to flowering and to cut the spike, where these values were increased by increasing the drought stress. These values under 40% FC were exceeded those under 80% FC by 10.96-14.38% for number of days to flowering and 14.27-19.90% for number of days to cut the spike, in both seasons, respectively. Meanwhile, the opposite trend had been occurred with the other floral characters; spike length, number of florets, floret fresh and dry weights.

It is worthy to mention that tuberose plants treated with DDM either grown under 80 and 100% FC levels in the 1st season or those under 60 and 80% in the 2ndseason, reached the blooming stage significantly earlier than any other treatments. Number of days to cut the spike had the same behavior with the same drought levels. The plants treated with DDM and grown under 80% FC reached the flowering stage earlier than its control by 8.10 and 23.95%, in the 1st and 2nd seasons, respectively. These values were 13.22 and 5.04% for the number of days to cut the spike, in the same order. The other studied floral characters; spike length, number of florets/spike, fresh weight of florets/plant and dry weight of florets/plant, behave the same compared with their controls, where these values were 16.85-9.38%, 3.64-10.67%, 23.85-8.91% and 47.55-61.91%, in the 1st and 2nd seasons, respectively (Table 3).

The effect of Si on blooming is becoming more marked if the plants grown under stress conditions (Sacała 2009 and Denisow *et al.*, 2015). Mirabbasi *et al.* (2013) reported that, drench of 25 mg/l potassium silicate produced the maximum number of florets per Asiatic lily plant. Rubinowska *et al.* (2014) mentioned that the application of Si as a foliar fertilizer positively affect the floral traits (flower size, nectar and pollen production).While, Kamenidou *et al.* (2009 and 2010) stated that, the beneficial effect of Si supplementation on the flower or spike characters has been earlier documented for several ornamental plant species, *e.g.* sunflower, zinnia and gerbera. Moreover, Wróblewska and Dębicz (2011) mentioned that, the foliar treatment of Si increased the number of buds and flowers for many ornamental plants; *i.e. Argyranthemum frutescens* 'Blazer Rose', *Xerochrysum bracteatum* 'Gold', *Osteospermum ecklonis* 'Grande Pink Bush', and *Gauralind heimeri* Corinas.

Chemical characters:

Regardless the FC% effects on tuberose plant, applying the DDM treatment on tuberose plant increased the mean values of total chlorophylls, total carbohydrates, protein and calcium percentages compared to their controls, in both seasons. While, it decreased the proline content. Meanwhile, under the different levels of FC, the mean values of total chlorophylls and protein content were decreased, in time of increasing the proline content. Moreover, at these levels, the mean values of total carbohydrates and calcium content were fluctuated (Table 4).

Concerning the interaction between the effect of DDM on the chemical characters under different drought stress levels, it is clear that total chlorophylls and protein contents decreased gradually with increasing the stress condition. The maximum reductions observed in total chlorophylls were37.20 and 33.37% at 40% FC, in both seasons, respectively. The same trend was obtained with protein content, where the maximum reductions were2.88 and 3.38%, in the two seasons, respectively. At the different levels of field capacity (FC%), DDM treatment led to slightly increment in the previous two characters than in the untreated plants. The chlorophyll and protein contents in the treated plants with DDM under 60 and 80% FC increased significantly than their controls.

The reduction of photosynthesis under drought stress could be attributed to the formation of proteolytic enzymes such as chlorophyllase, destruction of chlorophyll molecules by ROS, decline in membrane permeability, reducing water availability and nutrients particularly magnesium (Anjum *et al*. 2011). The result corroborate with the findings of Zhu and Gong (2014)and Rady and Mohamed (2015).The maximum chlorophyll index at harvesting time of Asiatic lily was related to spraying 25 mg/l potassium silicate (Mirabbasi *et al.*, 2013). Mauad *et al.* (2016) stated that, silicon can affect biochemical, physiological, and photosynthetic processes and, consequently, reduce drought stress. Silicon is one the important elements for the plants and plays an important role in tolerance of plants to environmental, stress heavy metal and biotic stress (Gong *et al*., 2005 and Tahir *et al.*, 2006).

Silicon is a beneficial element to many metabolic processes, *i.e.* suppresses the chlorophyll degradation(Shen *et al.,* 2010) or encourages the photosynthetic apparatus by promoting the chlorophyll contents, as well as positively impact on the water balance (Rubinowska *et al.*, 2014), which altogether results in the increase of the photosynthetic efficiency. Photosynthetic activity of leaves which supplies flower structures with organic compounds (primarily carbohydrates) most probably implicated the nectar and pollen results, as primarily carbohydrates are being utilized to flower growth and/or production of floral reward. Si impact on both the photosynthetic efficiency and water balance (Ma and Takahashi, 2002). The beneficial impact of silicon on osmo-regulation and effects of *water impoundment* was confirmed by different authors (Kazemi *et al.*, 2012 and Rubinowska *et al.*, 2014).

Table 4: Effect of Diatomite (DDM) on some chemical characters of tuberose plant under different field capacity FC% during the two seasons; 2014and 2015.

Field capacity (FC)% (A)*	1st season			2nd season		
	DDM (B)*		Mean (A)	DDM (B)*		Mean(A)
	Control	DDM		Control	DDM	
Total chrolophylls (SPDA units)						
100	38.87	39.60	39.24	39.77	40.07	39.92
80	38.57	41.33	39.95	38.50	41.90	40.20
60	38.40	40.37	39.39	35.73	39.23	37.48
40	37.20	39.57	38.39	33.37	35.67	34.52
Mean (B)	38.26	40.22		36.84	39.22	
LSD 5%	A= 1.82	B= 1.28	AB=2.57	A= 1.65	B= 1.17	AB=2.34
Total carbohydrates %						
100	17.67	19.10	18.39	16.77	17.52	17.15
80	20.23	21.37	20.80	18.28	18.99	18.64
60	21.77	21.98	21.88	19.09	19.21	19.15
40	19.39	19.79	19.59	18.05	18.70	18.38
Mean(B)	19.77	20.56		18.05	18.61	
LSD 5%	A= 0.78	B= 0.55	AB= 1.10	A= 0.69	B= 0.49	AB=0.98
Protein content %						
100	5.44	6.00	5.72	4.94	5.44	5.19
80	4.38	6.69	5.54	4.19	5.88	5.04
60	3.56	6.44	5.00	3.47	5.75	4.61
40	2.88	5.69	4.29	3.38	4.25	3.82
Mean (B)	4.07	6.21		3.99	5.33	
LSD 5%	A= 0.28	B= 0.20	AB= 0.40	A= 0.24	B= 0.17	AB= 0.35
Proline content mg/g F.W						
100	0.36	0.21	0.29	0.52	0.46	0.49
80	0.51	0.30	0.41	0.68	0.50	0.59
60	0.67	0.46	0.57	0.80	0.66	0.73
40	0.92	0.48	0.70	1.41	0.87	1.14
Mean (B)	0.62	0.36		0.85	0.62	
LSD 5%	A= 0.03	B= 0.02	AB= 0.04	A= 0.03	B= 0.02	AB= 0.04
Calcium content %						
100	1.28	1.50	1.39	1.38	1.41	1.40
80	1.54	1.67	1.61	1.56	1.68	1.62
60	1.52	1.54	1.53	1.51	1.60	1.55
40	0.80	1.14	0.97	1.22	1.55	1.39
Mean (B)	1.29	1.46		1.42	1.56	
LSD 5%	A= 0.11	B= 0.08	AB= 0.15	A= 0.07	B= 0.05	AB= 0.10

Concerning the carbohydrates content, data revealed that, the content was slightly increased with 60 and 80% FC, then insignificantly reduced with 40% FC. Treated the tuberose plants with DDM slightly increased the carbohydrate content. The maximum increments; 21.98 and 19.21% were in the plants treated with DDM grown under 60% FC in both seasons, respectively (Table 4). Zhu *et al.* (2004) and Gong *et al.* (2005)stated that DDM treatment increased the protein and total carbohydrates contents in the drought stressed plants by increasing protein biosynthesis or decreasing its oxidation which is accomplished by an increase in the activities of certain antioxidant and hydrolytic enzymes as peroxidases (POD), catalases (CAT), esterases (EST) and acid phosphatases (ACP).

Proline content increased as the drought stress increased either in the treated or untreated plants with DDM. The highest content of proline in the untreated plants (0.92-1.41%) were recorded with 40% FC, in both seasons. Although treated the plants with DDM and grown under 40 % FC, decreased the proline content (0.48-0.87%), these contents still higher than those with the other FC levels (Table 4).Numerous studies have shown that the proline content in higher plants increases under different environmental stresses (Laszlo and Arnould, 2009).Silicon fertilization may have reduced water loss as the Silicon considered as an osmotic regulator (Gunes, 2008). Moreover, Si fertilization reduces the proline content in the vegetative and reproductive plant tissues and increases the activity of peroxidase in the reproductive phase, which could berevealing of stress tolerance (Mauad *et al.,* 2016).

Drought stress had a significant effect on the Ca % in tuberose plants. The Ca content in leaves decreased in both seasons with increasing drought stress. Application of DDM increased the Ca content than the control plants with different FC levels. The maximum increments in Ca% (1.67 and 1.68%) were recorded in the plants treated with DDM at 80% FC, in both seasons compared with the other treatments (Table 4).

These results could be due to the important role of DDM as known to increase drought tolerance in plants by maintaining plant water balance and photosynthetic activity (Hattori *et al*., 2005). Faraz *et al.* (2007) reported that increased levels of Si significantly increased plant biomass, plant weights and dry matter production under different levels of water stress. Si fertilization increases crop yield and quality and thus insures high production under severe biotic and abiotic stresses (Hou *et al.*, 2006). Therefore, it has the ability to reduce the adverse effect of drought and to protect the plant against the build-up of toxic ions by maintenance of the ionic homeostasis under stress and this makes N, P, K, Ca more available to the plant. These results are in parallel with the finding of Abd El–Hamied and El-Amary (2015).

Anatomy:

Microscopical counts and measurements of certain histological characters in transverse sections through the blade of tuberose leaf grown under drought stress and treated by DDM are presented in Table (5) and Fig. (1).

Table 5: Means of counts and measurements (μ) of different tissues of tuberose leaf as affected by DDM treatments (Si) under different field capacity (FC %) (av. of 10 readings).

Field capacity (FC%)	DDM treatment (Si)	Leaf thick. (μ)	Upper epidermis thick. (μ)	Lower epidermis thick. (μ)	Mesophyll thick. (μ)	Mean No. of stomata on lower surface
100	0 DDM	397.53	13.84	10.15	375.52	73.20
	DDM	345.17	9.80	9.18	323.49	74.00
80	0 DDM	345.04	12.63	9.54	312.17	71.00
	DDM	359.65	12.42	10.10	342.37	73.83
60	0 DDM	307.49	11.55	9.27	276.80	67.40
	DDM	335.17	11.32	8.45	295.98	74.00
40	0 DDM	297.62	10.53	8.32	284.34	55.50
	DDM	302.35	10.30	8.42	272.33	59.00

concerning the drought levels, it is obvious that, all measurements of the characters studied showed a remarkable reduction as the drought levels increased. The highest decrements were at 40% FC comparing to 100% FC were 25.13%for leaf thickness, 23.92% for upper epidermis thickness, 18.02% for lower epidermis thickness and 24.28% for mesophyll thickness. DDM application had slightly increase in the leaf measurements. For example, the increments occurred at 60% and 80% FC compared with their controls were 9% for leaf thickness and 9.67% for mesophyll thickness, respectively.DDM treatments had no obvious effect on the upper and lower epidermis thickness

(Fig.2). Moreover, at 60% FC, treated the plant by DDM increased the mean number of stomata by 9.79% comparing to its control. All stomata patterns on both leaf surfaces were anomocytic (Fig.3).

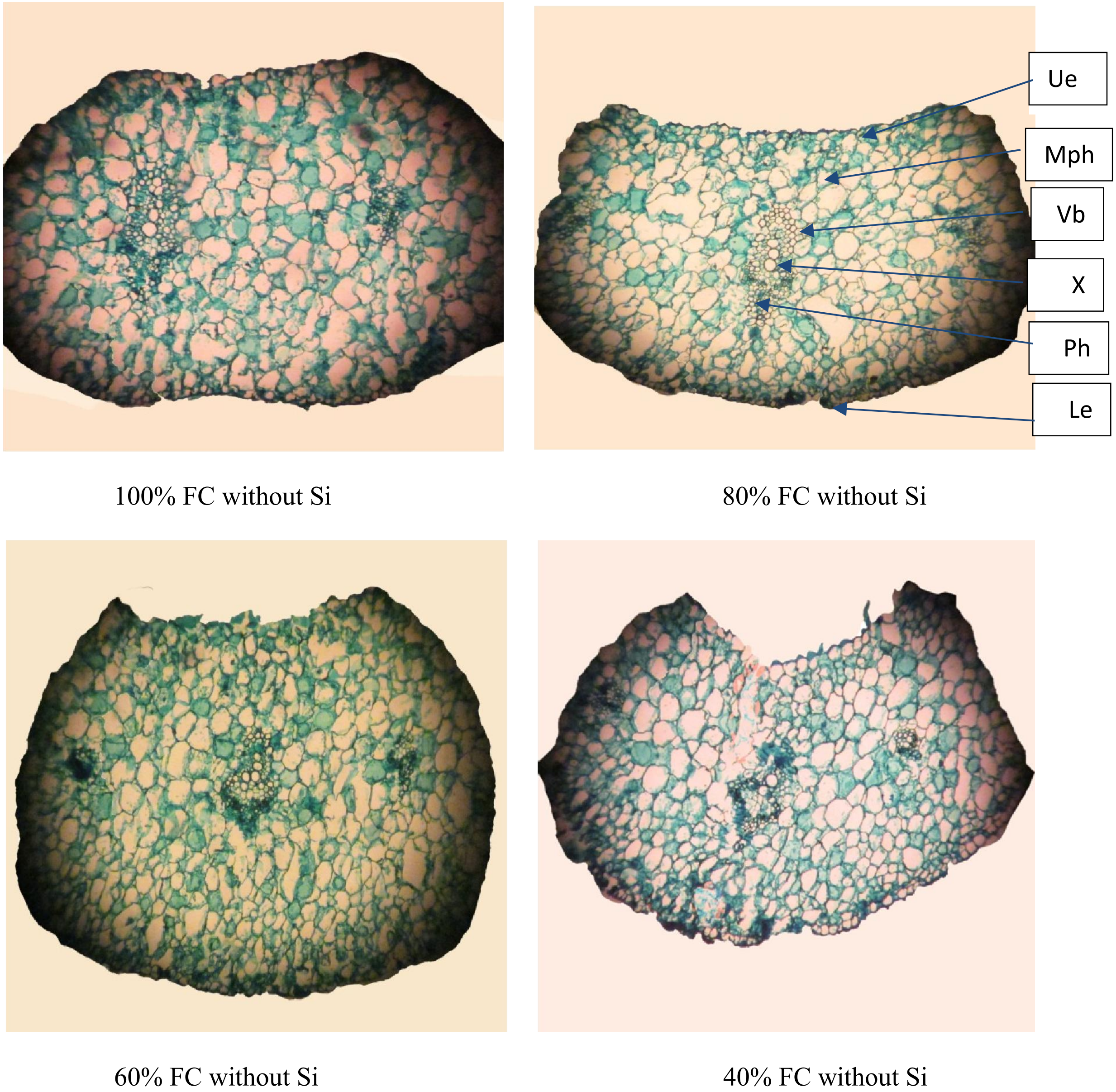
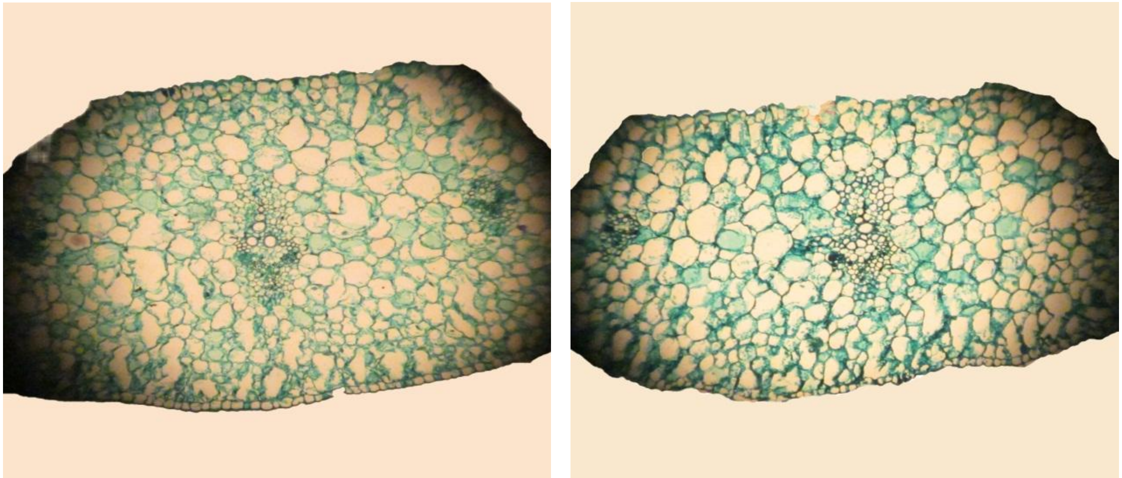


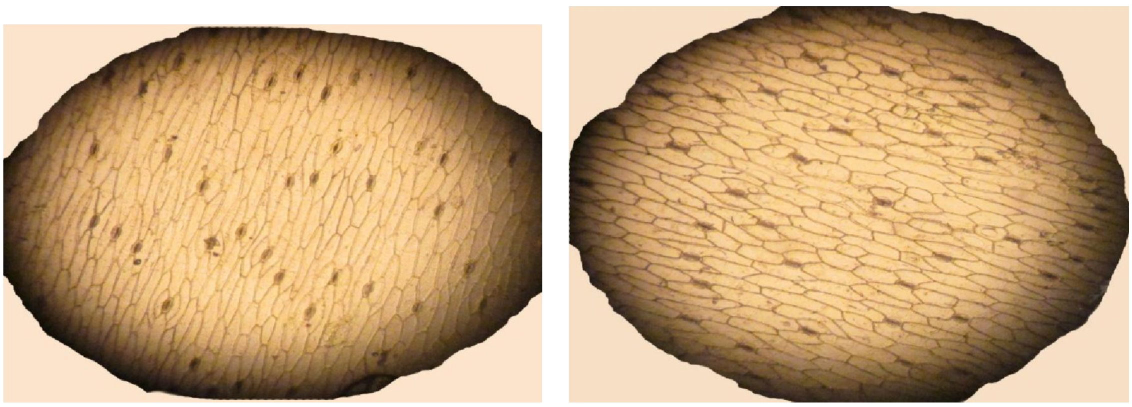
Fig. 1: Effect of different levels of drought stress (FC%) on the leaf internal structure of tuberose plant without using Silicon (Si). (100 x)
Key : Ue: Upper epidermis, Le: Lower epidermis, Mph: Mesophyll, Vb: vascular bundle, X: Xylem and Ph: Phloem.



80% FC with Si 40% FC with Si

Fig. 2: Effect of applying Silicon on the leaf internal structure of tuberose plant under different drought levels (FC%). (100 x)

Stomata type (anomocytic) (400x)



60% FC with Si 60% FC without Si

Fig. 3: Stomatal type and effect of treated the tuberose plant by Silicon on the number of stomata. (100 x)

It can be recommended to use Diatomite with 80% FC to produce tuberose flowers with high marketing quality. To delay flowering, Diatomite can be used with 60% FC irrigation.

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