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IMPACT OF SALICYLIC ACID FOLIAR APPLICATION ON TWO WHEAT CULTIVARS GROWN UNDER SALINE CONDITIONS

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Abstract

Salinity is one of the limiting factors in agricultural production of all crops in the world specifically in cereals crops. There are different ways to decrease the effect of salinity on growth and production of all plants. One of these ways is using salicylic acid which is plant growth regulator; it is able to increase the salt tolerance in crops. The aim of this work was to study salicylic acid effects on alleviation salinity stress and enhance plant growth parameters, physical characteristics, biochemical constituents, element contents, and production of two wheat varieties grown under salinity stress conditions in the North West area of Egypt. The experiment was designed in split plot with using four replicates. The two wheat varieties; Giza 168 a salt sensitive variety and Sakha 93 a salt tolerant variety were used with salicylic acid (SA) foliar application treatments: control, SA100 (100 mg salicylic acid/ L), SA200 (200 mg salicylic acid/ L) and SA400 (400 mg salicylic acid/ L) were applied during tillering and booting initiation stages. Results cleared that salicylic acid foliar application decreased the damage effects of salinity stress in both wheat cultivars especially, in Giza168 which was more sensitive to salinity by increasing growth and production of antioxidant such as ascorbic acid and protect both protein and chlorophyll from breakdown by free radicals. Foliar application of salicylic acid increased grain and straw yield of studied wheat cultivars and also increased potassium content and decreased Na: K ratio in straw. Overall, it can be suggested that foliar application of Salicylic acid is effective strategy to improve wheat productivity under salinity stress especially, for salt sensitive cultivars.

Key words: Wheat cultivare, Salinity stress, Salicylic acid, Nutrient concentration, Growth, Yield and its components.

Introduction

Wheat (*Triticum aestivum* L.) is considered as one of the important cereal grain crops in Egypt. The nutritional value of wheat (protein) is higher than maize and rice. Moreover, wheat is considered as a main component in Egyptians food. Wheat suffers from grain yield decrements due to soil salinity by affecting nutrient uptake (Zafar *et al*., 2015). Salinity stress is type of environmental stress significantly affecting the growth, and production of all crops (Akhtar *et al*., 2013; Kausar& Gull, 2014; Mahboob *et al.*, 2016).Also, Salinity stress brings many changes in biochemical and physiological processes especially seed germination, respiration rate, mineral nutrition, organic solutes/osmolyte synthesis, enzyme activities and photosynthesis (Ebrahimi*et al*., 2012; Ashraf *et al*., 2013; Hasanuzzaman *et al*., 2013; Zafar *et al*., 2015; Mahboob *et al*., 2017). Mechanisms of salt tolerance change from plant to another and also from variety to another in the same plant because there are a lot of interactions in biochemical and physiological processes (Ashraf and McNeilly, 2004; Manaa *et al*., 2011; Mahboob *et al*., 2017). Salt tolerance may be increased in wheat by producing new varieties which are highly production under salinity conditions and also there are another ways such as leaching, drainage, using the appropriate cultural practices like natural product fertilization, and irrigation and using some growth regulators such as salicylic acid (SA). The relationship among the different characteristics of seedling growth andyield componentsunder saline conditions is very important when developing a salinity tolerant cultivar in saline conditions (Mujeeb-ur-Rahman *et al.*, 2008). Salicylic acid (SA) is considered as an endogenous hormone withPhenolic composition.It plays an important role to decrease the effect of biotic and abiotic (environmental) stress on biochemical and physiological processes (Karlidag *et al.*, 2009). These effects are resulted from salicylic acid which increase the antioxidative enzyme activity, transpiration rate, stomatal regulation , photosynthesis, germination, ion uptake and plant growth (Khan *et al*., 2003; Metwally *et al*., 2003; Khodary 2004; He *et al*., 2010; Erdal *et al*., 2011). Foliar application of salicylic acid decrease the negative effects of salinity stress in wheat varieties and produced increase in growth and production, in addition to improve some physical, biochemical constituents and nutritional value of two wheat cultivars grown under saline conditions.

Materials and Methods

The present study was conducted during two seasons at the experimental site located at El-Hamam farm, Faculty of Agriculture, Alexandria University, at the North West area of Egypt.Physical and chemical characteristics of soil of experimental site were determined according to Chapman & Pratt (1978), The soil texture was sandy clay with EC 12.0 dS/m, pH 7.6, Calcium carbonate ($CaCO_3$) 32.1%; Organic Matter (OM) 1.50 %, P 0.22 mg/100 g soil, K 4.4 mg/100 g soil and Fe, Mn, Zn and Cu were 1.60, 4.40, 0.68 and 0.62 ppm, respectively. The experiment design was spilt plot in a randomized complete blocks arrangement with four replicates in all trials (two wheat cultivars, four Salicylic acid treatments and four replicates). The total numbers of experimental units used were 32 plots (plot area was 9 m^2). Main Treatments included two wheat cultivars; Giza 168 a salt sensitive and Sakha 93, considered as a salt tolerant cultivar with foliar application of salicylic acid (SA) treatments (control, 100 mg salicylic acid L^{-1}; SA 100, 200 mg salicylic acid L^{-1};

SA 200 and 400 mg salicylic acid L^{-1}; SA 400) randomly distributed in the subplot. These foliar application treatments were applied twice per season, during tillering and booting initiation stages**.** The experiment was started at the same time in two seasons by sowing of wheat grains, keeping all agronomic practices normal and uniform for all the treatments. After 60 days from planting plant samples were collected (above ground) to determine growth parameters (plant height and shoot dry weight (Basra *et al*., 2002), physical characteristics (membrane stability index (MSI %) as mentioned by Sairam *et al*., (2005) and relative water content (RWC%), and biochemical characteristics (chlorophyll index according to Wood *et al*., (1992), ascorbic acid content as described by Mukherjee &Choudhuri (1983), and protein content by using Bradford (1976) method. At harvesting time (maturity) sample of plants was taken from one meter/plot that were devoted to determine the yield and its components: weight of spike (gm)**,** numbers of kernels per spike, weight of 100 grain (gm), weight of straw (kg/plot) and weight of grains (kg/plot).Also at harvest samples from straw and grains were taken for the determination of nutrients by methods described by Cottenee *et al*., (1982). Statistical analysis was done according to Snedecor & Cochran (1990). The combined analysis was conducted for all data of the two seasons according to Steel *et al*., (1997), for data of two seasons the least differences (L.S.D) was used to compare the means.

Results and Discussion

Growth parameters: Results in table (1) cleared there were no significant differences in plant height between the two studied wheat cultivars. While, shoot dry weight showed marked differences. Sakah-93 showed 13% increase in shoot dry weight as compared with Giza-168. This increment might be due to the negative effect of soil salinity where, Sakha-93 is salinity tolerant cultivar and Giza-168 is a salinity sensitive one. The other explanations, the different response here related to the genetic background of the studied cultivars. These results are consistent with the results obtained by Bakry *et al.*, (2013); Ahmed, (2013) and El-Nasharty *et al.*, (2017).

Effect of salicylic acid foliar application on wheat growth parameters i.e. plant height and shoot dry weight, showed significant positive effect on wheat growth (Table 1). Only the highest doses of salicylic acid foliar spray (400 ppm) increased plant height about 11% as compared with control treatment (water spray) significantly. All salicylic acid doses showed marked increments in wheat shoot dry weights as compared with control treatment. The highest dose of salicylic acid gave the highest shoot dry weight increase (35% over control treatment). The results showed that foliar spray of salicylic acid could reduce the negative effect of salinity stress on wheat growth, recovery of growth processes and counteract the negative effect of salinity (Idrees *et al.*, 2011).

The interaction effect of wheat cultivar and foliar spray with salicylic acid showed only significance in case of shoot dry weight. Giza-168 foliar application with 400 ppm salicylic acid showed the highest shoot dry weight while, the same cultivar sprayed with water showed the lowest shoot dry weight value.

Physical characteristics: Results of wheat leaves membrane stability index showed significant differences between wheat cultivars (Table 2). Sakha-93 showed 20% increase as compared with Giza-168 which was considered as sensitive cultivar. These results were consistent with Sairam *et al*., (2005) who demonstrated that salinity sensitive wheat cultivars grown in saline media showed decline in membrane stability index than tolerant cultivars which reflected their tolerant nature. While, no significant effect between the two studied cultivars in their leaves relative water content was recorded.

Results in table (2) also indicated that foliar spray with salicylic acid had marked positive effect on both leaves membrane stability and leaves relative content. Spraying wheat plants with 200 ppm salicylic acid resulted in the highest values for leaves membrane stability and leaves relative content as compared to control and other salicylic acid treatments. Khan *et al*., (2010) showed that foliar application of salicylic acid resulted in increasing Ca^{+2} accumulations which helped in decreasing membrane damages. Yusuf *et al*., (2008) also reported that salicylic acid decreased the intensity of damages of reactive oxygen species (ROS) on membranes which produced under stress by enhanced production of enzymatic and non-enzymatic antioxidant. He (2010) and Sakhabutdinova *el al*., (2003) found that application of salicylic acid enhanced photosynthesis and also increased photosynthetic products which affected leaf sap and increased water leaf content. Similar finding have been reported by Yildirim *et al*., (2008)

Marked positive interactions effect were found. Sakha-93 foliar application with 400 ppm salicylic acid for leaf membrane stability while, Giza-168 foliar application with 200 ppm salicylic acid for leaf relative water content showed the highest significant increments.

Biochemical characteristics: The significant differences in ascorbic acid content and chlorophyll index were found between the two studied wheat cultivars. However, no marked effect was recorded concerning the total protein (Table 3). Sakah-93 which was considered as a salinity tolerant cultivar recorded the highest values of most studied biochemical characteristics as compared with Giza-168. Thus Sakah-93 could successfully be sown in salinity affected soils. This cultivar also, could be considered as plant material in breeding programs for producing salt tolerant wheat cultivars.

It is also clear that foliar spraying plants with 200 ppm salicylic acid showed the highest significant increments in ascorbic acid and total protein as compared with the other treatments.

Moreover, the highest significant increment in chlorophyll index was achieved as a result of spraying plants with 400 ppm salicylic acid. Salicylic acid application affected the activity of Rubisco which plays an important role on the efficiency of photosynthesis process. Results of this study were in agreement with those of (Yildrim *et al*., 2008) who found that under salinity stress chlorophyll contents were decreased because of formation of proteolytic chemicals such as cholropyllase enzyme.

Table 1. Effect of cultivar, salicylic acid foliar spray and their interaction on wheat growth (60 days old) grown on salinity calcareous soil (Combined analysis of two seasons).

Treatment	Plant height (cm)			Shoot dry weight (g/plant)		
	Giza 168	Sakha 93	Mean	Giza 168	Sakha 93	Mean
Control (water spray)	79.83	80.17	80.0	1.64	2.54	2.09
SA 100 ppm	80.89	83.05	81.97	2.06	2.69	2.37
SA 200 ppm	82.00	80.58	81.29	2.27	2.18	2.23
SA 400 ppm	90.50	86.75	88.63	2.97	2.68	2.83
Mean	83.31	82.64		2.23	2.52	
L.S.D. 0.05						
Cultivar		N.S.			0.22	
SA		2.99			0.15	
Cultivar X SA		N.S.			0.26	

Table 2. Effect of Cultivar, Salicylic acid foliar spray and their interaction on wheat physical characteristics (60 days old) grown on salinity calcareous soil (Combined analysis of two seasons).

Treatment	Membrane stability index (%)			Relative water content (%)		
	Giza 168	Sakha 93	Mean	Giza 168	Sakha 93	Mean
Control (water spray)	48.02	57.04	52.53	71.11	64.81	67.96
SA 100 ppm	51.29	42.45	46.87	66.84	58.69	62.77
SA 200 ppm	51.47	60.28	55.87	72.24	68.79	70.52
SA 400 ppm	34.07	61.76	47.92	54.45	67.32	60.89
Mean	46.21	55.38		66.16	64.90	
L.S.D. 0.05						
Cultivar		4.61			N.S.	
SA		6.52			7.27	
Cultivar X SA		9.22			10.29	

Table 3. Effect of cultivar, salicylic acid foliar spray and their interaction on wheat biochemical characteristics (60 days old) grown on salinity calcareous soil (Combined analysis of two seasons).

Treatment	Ascorbic acid (ppm)			Chlorophyll index			Total protein (%)		
	Giza168	Sakha 93	Mean	Giza168	Sakha 93	Mean	Giza168	Sakha 93	Mean
Control (water spray)	13.04	12.94	12.98	40.93	42.56	41.75	6.37	8.63	7.50
SA 100 ppm	13.03	12.92	12.97	41.58	41.72	41.65	9.06	9.88	9.47
SA 200 ppm	13.12	13.04	13.08	42.79	44.47	43.63	10.30	11.72	11.01
SA 400 ppm	12.97	13.01	12.99	42.00	47.68	45.14	8.86	10.60	9.73
Mean	13.04	12.98		41.98	44.11		8.65	10.21	
L.S.D. 0.05									
Cultivar		0.05			2.00			N.S.	
SA		0.09			1.25			1.02	
Cultivar X SA		N.S.			1.76			N.S.	

Salicylic acid foliar application enhanced maize growth under saline conditions which might have been because of proline accumulation in plants (Kiran *et al.*, 2015). Some studies showed that foliar application of tomatoes (Javaheri *et al*., 2012) and strawberry (Aghaeifard *et al*., 2015) with SA produced high concentration of ascorbic acid compared to control, this effect of SA was due to activation of ascorbate peroxidase enzyme, which played an important role to protect ascorbic acid (vitamin C) and caused ascorbic acid accumulation in cell.

The interaction effect between the two studied factors, showed only significant effect on chlorophyll index (Table 3). The highest chlorophyll index was achieved by spraying Sakah-93 with 400 ppm salicylic acid.

Yield and related traits: Sakha-93 showed 5 % increase in its grain yield as compared with Giza-168 (Table 4). This increment was statistically significant. While no significant differences between the two wheat cultivars were observed in their straw yield.

Spraying plants with salicylic acid with either the lowest does or the highest does resulted in the increase of the wheat grain yield by about 11% as compared with control treatment. The increments ranged between 11-16 % over control treatment. The highest grain yield increment was achieved by spraying the plants with 200 ppm salicylic acid. However, most does of salicylic acid caused significant increases in straw yield.

Regarding to the interaction between the two studied factors, significant effects were found.The highest grain and straw yields were obtained as a result of the interaction of spraying Sakha-93 with the lowest salicylic does (100 ppm). However, the lowest grain and straw yields were achieved by spraying Giza-168 with water (control).

Table 4. Effect of cultivar, salicylic acid foliar spray and their interaction on weight of spike, numbers of kernels per spike, weight of 100 grain, yield of straw and grains of two wheat cultivars grown on salinity calcareous soil (Combined analysis of two seasons).

Cultivar (C)	Salicylic acid (SA), ppm	Weight of spike (gm)	Numbers of kernels per spike	Weight of 100 grain (gm)	Yield of straw (kg/plot)	Yield of grains (kg/plot)
Giza 168 (C1)	Cont.	1.82	13.33	3.77	5.06	5.40
	SA 100	1.96	16.00	3.87	5.12	5.74
	SA 200	1.79	15.67	3.58	5.07	6.74
	SA 400	1.73	14.33	3.51	6.26	6.30
Sakha 93 (C2)	Cont.	1.46	12.33	3.71	5.31	5.93
	SA 100	1.83	13.00	3.81	6.75	6.93
	SA 200	1.89	15.33	3.99	5.18	6.46
	SA 400	1.54	14.33	3.93	5.61	5.25
Mean values of Cultivars (C)	Giza 168 (C1)	1.83	14.83	3.68	5.38	6.05
	Sakha93 (C2)	1.68	13.75	3.86	5.71	6.37
Mean values of Salicylic acid (SA), ppm	Cont.	1.64	12.83	3.74	5.19	5.67
	SA 100	1.90	14.50	3.84	5.94	6.29
	SA 200	1.84	15.50	3.79	5.13	6.60
	SA 400	1.64	14.33	3.72	5.94	6.28
LSD at 5 %	(C)	N.S.	0.95	N.S.	N.S.	0.30
	(SA)	0.13	1.17	N.S.	0.55	0.36
	(C) X (SA)	0.19	N.S.	N.S.	0.77	0.51

Table 4 showed the effect of spraying different rates of salicylic acid at two wheat cultivars and their interaction on yield components. Results revealed that, the two studied cultivars only differed significantly in number of kernels/spike but Giza-168 showed 8% increase as compared with Sakha-93. In contrast, Giza 168 gave the lowest weight of grains and this decrease in weight of grains was associated with decreased 100-kernel weight compared to Sakha 93.This might be because Sakha 93 headed earlier than Giza 168, therefore, it had enough time to fill its kernels which was reflected in heavier weight of grains than Giza 168, Ali *et al*., (2004).

Spraying wheat plants with salicylic acid significantly increased weight of spike and number of kernel/spike. These effects of SA foliar application may be due to its effect on total fertile florets which resulted in increment of numbers of kernels per spike followed by increased spike weight. Generally spraying wheat plants with 200 ppm salicylic acid showed the highest increments in the aforementioned traits as compared with the other treatments. In spite of salicylic acid foliar spray increased 100 grain weight but the increment did not reach to the significantly level. Jafar *et al.*, 2012 found that Salicylic acid application on two wheat varieties under salinity stress improved plant growth and yield component (tillers number, spikelet number per spike, numbers of kernels per spike, biological yield and grain yield).The improvement effect of SA foliar application on grain yield might be due to its effects on physiological and biochemical processes such as photosynthesis and activation of rubisco enzyme (Ashraf *et al*., 2013). Kaydan *et al*., (2007) and Afshari *et al*., (2013) found that the lowest concentrations of salicylic acid were the most effective on wheat growth compared to the highest concentrations. This effect might be due to decreasing the activity of oxygenase, nitrate reductase, ribulose-1, 5-biphosphate carboxylase and antioxidant enzymes which were produced naturally (Hayat *et al*., 2005; Hayat *et al*., 2010; Ashraf *et al*., 2013).

Concerning the interaction effect of the two studied factors on weight of spike, straw and grain yield showed marked significant increase. Giza-168 foliar application with salicylic acid (SA100) showed the highest increase weight of spike but Sakha-93foliar application with salicylic acid (SA100) gave the highest increase of straw and grain yield.

Element contents: The mean estimates concentration (%) of Potassium (K), Sodium (Na), Calcium (Ca) and Na: K ratio in straw and grains of two wheat cultivars grown on calcareous soil as influenced by high salinity indicated that the differences among cultivars were significant except for concentration of K and Na: K ratio in grains (Table 5).

Giza168 cultivar (sensitive genotype) gave the highest values of K and Ca content in straw and Na content in grain but Na and Na: K ratio content in straw and Ca content in grain were the highest in Sakha93 cultivar (tolerant genotype). Tolerant variety showed high significant increase in K/Na ratio in straw in comparison with sensitive variety. These results are in agreement with the results obtained by Oyiga *et al*., (2016).

Foliar application of salicylic acid showed significant effects on K,Na,Ca and Na;K in both straw and grains of the two studied wheat cultivars (Table 5). The highest content value of K, Na and low Na: K ratio in straw was produced by foliar spray of 400 ppm salicylic acid. Whereas in grains the same values were produced by foliar spray of 200 ppm salicylic acid.

These results are agreed with by Baghizadeh *et al*., (2012) who found that SA increased potassium concentration in leaf and root of tomato plant under salinity condition.

Table (5). Effect of cultivar, salicylic acid foliar spray and their interaction on sodium, potassium, calcium (%) concentrations and Na: K ratio in straw and grains of two wheat cultivars grown on calcareous soil as influenced by high salinity (Combined analysis of two seasons).

Cultivar (C)	Salicylic acid (SA), ppm	Straw				Grains			
		K	Na	Ca	Na:K	K	Na	Ca	Na:K
Giza 168 (C1)	Cont.	2.21	0.47	2.02	0.21	0.38	0.14	0.31	0.37
	SA 100	2.24	0.52	2.37	0.23	0.39	0.15	0.29	0.38
	SA 200	2.92	0.25	2.03	0.09	0.43	0.16	0.31	0.37
	SA 400	3.02	0.49	2.18	0.16	0.39	0.17	0.33	0.44
Sakha 93 (C2)	Cont.	1.66	0.58	1.57	0.35	0.43	0.12	0.40	0.28
	SA 100	2.57	0.51	2.32	0.20	0.43	0.14	0.35	0.33
	SA 200	1.63	0.48	1.48	0.29	0.47	0.17	0.39	0.36
	SA 400	2.38	0.61	2.04	0.26	0.41	0.14	0.41	0.34
Mean values of Cultivars (C)	Giza 168 (C1)	2.60	0.43	2.15	0.17	0.40	0.16	0.31	0.40
	Sakha93 (C2)	2.06	0.55	1.85	0.27	0.44	0.14	0.39	0.32
Mean values of Salicylic acid (SA), ppm	Cont.	1.94	0.53	1.80	0.28	0.41	0.13	0.36	0.33
	SA 100	2.41	0.52	2.35	0.22	0.41	0.15	0.32	0.36
	SA 200	2.28	0.37	1.76	0.19	0.45	0.17	0.35	0.37
	SA 400	2.70	0.55	2.11	0.21	0.40	0.16	0.37	0.39
LSD at 5 %	(C)	0.22	0.02	0.08	0.04	NS	0.01	0.02	NS
	(SA)	0.09	0.03	0.07	0.01	0.02	0.01	0.01	0.05
	(C) X (SA)	0.13	0.04	0.11	0.01	NS	0.04	NS	NS

According to Baghizadeh *et al*., (2012) that salysilic acid alleviates the effects of salts on plasma membrane which can damage it under saline conditions for that SA plays an important role in increased nutrients uptake. Khan *et al.,* (2014) also found that salicylic acid application changed Na^+ and K^+ selectivity uptake and decreased Na^+/K^+ ratio, which protected membrane from damage.

On the others hand, the interaction between foliar spray of salicylic acid and cultivaes were significantly in all nutrient concentrations of straw and grains except K and Na: K ratio in grains of two wheat cultivars (Table 5). Foliar application of Giza168 gave the highest value of K with SA 400 and highest value of Ca with SA 100. With respect to Sakha 93, higher Na concentration was obtained with SA 400 in straw. On the other hand, in grains the highest value of Na concentration was obtained by Sakha 93 with SA 200 however, the same value of Na concentration was obtained by Giza168 with SA400.

Conclusion

Salicylic acid is considered as an endogenous hormone with phenolic composition, it plays an important role to decrease the effect of biotic and abiotic (environmental) stress on biochemical and physiological processes. Foliar application of salicylic acid alleviated the salinity-induced damage in both wheat cultivars especially in Giza168 which was more sensitive to salinity by increasing growth and production of antioxidant such as proline and ascorbic acid and protect protein and chlorophyll from breakdown by free radicals. Foliar application of salicylic acid increased yield of both wheat varieties and also increased potassium content in grains and decreased Na: K ratio in grains. These results suggest that foliar application of salicylic acid is an effective strategy to improve wheat productivity under salinity stress especially, for salt sensitive cultivars.

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