

# **$\alpha$ -TOCOPHEROL FOLIAR APPLICATION CAN ALLEVIATE THE ADVERSE EFFECT OF SALINITY STRESS ON WHEAT PLANT, *TRITICUM AESTIVUM* L.**

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(Received 12 February 2019, Revised 23 March 2019, Accepted 8 May 2019)

**ABSTRACT :** Pots experiment was conducted at the greenhouse of botanical garden belong to Department of Biology, College of Education for Pure Science, Ibn-AL-Haithum, University of Baghdad, for growth season 2018-2019. The aim of the experiment was to study the effects of foliar application of  $\alpha$ -tocopherol concentrations (0, 50, 100, 150, 200 mg.L<sup>-1</sup>) on growth parameters and the activity of some antioxidant enzymes of wheat plant irrigated with sodium chloride concentrations (0, 75, 150, 225) mM.L<sup>-1</sup>. Salinity reduced plant growth parameter, plant height, flag leaf area, flag leaf chlorophyll content and increased the activity of antioxidant enzymes, superoxide dismutase and peroxidase. Plant growth parameters were enhanced by foliar application of  $\alpha$ -tocopherol for the interaction between the two factors, the best values for plant height and flag leaf area under the effect of concentration 150mg.L<sup>-1</sup>  $\alpha$ -tocopherol and concentration 225 mM.L<sup>-1</sup> sodium chloride. There was different founds for flag leaf chlorophyll content, which  $\alpha$ -tocopherol at concentration 200 mg.L<sup>-1</sup> and the concentration 225mM.L<sup>-1</sup> sodium chloride gave the best values for the parameter, there was no significant difference between the values of flag leaf chlorophyll content at the concentrations 150, 200 mg.L<sup>-1</sup>  $\alpha$ -tocopherol under the same concentration of sodium chloride. Antioxidant enzymes were increased by  $\alpha$ -tocopherol foliar spraying, the best value for superoxide dismutase activity at height concentration 225 mM.L<sup>-1</sup> sodium chloride and 150 mg.L<sup>-1</sup>  $\alpha$ -tocopherol, the best value for peroxidase activity at concentration 225 mM.L<sup>-1</sup> sodium chloride and 100 mg.L<sup>-1</sup>  $\alpha$ -tocopherol.

**Key words :** Wheat plant, salinity stress, oxidative stress, antioxidant enzymes,  $\alpha$ -tocopherol.

## **INTRODUCTION**

Wheat is major food crop all over the world and different hard stress reduces its yield, wheat provides 55% of carbohydrates and 22% of food calories consumed globally (Hasanuzzaman *et al*, 2017). Salinity stress is the most limiting crop yield, plants under salt stress are subjected to three kind of stress including water deficit caused by osmotic unbalance, ion toxicity due to accumulation of adverse ions and nutritional alternations caused by unbalance in ion transport system (Miller *et al*, 2010). Excessive soil salinity cause accumulation of sodium in wheat leaves and reduce the growth due to the harmful effect and low water availability (Pervize *et al*, 2002). Salinity affect photosynthesis and ion distribution so it cause reduction in plant height, dry weight and leaf area (Al-Saidi and Al-Kazzaz, 2013). Salinity stress trigger oxidative stress in plant tissue, causing to accumulation of reactive oxygen species (ROS) which are harmful and very reactive in nature because they can interact with a number of other molecules and metabolites such as DNA, pigments, proteins, lipids, convert membrane properties such as fluidity and lead

to cell death (Sharma *et al*, 2012). Plants have developed detoxification mechanisms including the synthesis of antioxidant molecules such as tocopherol and ascorbic acid and various antioxidative enzymes such as superoxide dismutase, peroxidase and catalase (Ahmad *et al*, 2010).  $\alpha$ -tocopherol known as vitamin E are lipid soluble antioxidants, its synthesized by plants and photosynthetic organisms, it can accumulate in plant leave in response to a biotic stress (Mene-saffrane and Della Penna, 2010).  $\alpha$ -Tocopherol have number of functions in plants, the most prominent one is protection of chloroplast membrane polyunsaturated fatty acid from lipid peroxidation (Colombo, 2010).  $\alpha$ -Tocopherol quench<sup>1</sup>O<sub>2</sub> in photosynthetic membrane organelles and limit lipid peroxidation by reducing lipid peroxy radicals to hydroperoxides (Munne-Bosch, 2007). In the study of (Farouk, 2011) mentioned that salinity increased the senescence of wheat leaves, decreased the concentrations of chlorophyll, potassium, K<sup>+</sup>/Na<sup>+</sup> ratio and soluble proteins and increased sodium, chloride content in plant tissue, application  $\alpha$ -tocopherol reduced sodium and chloride content, increased potassium content,

antioxidants enzymes activity and reduced lipid peroxidation.

In our study, we aimed to evaluate the extent to which wheat plant AL-Baraka cultivar can withstand salinity stress and determine  $\alpha$ -tocopherol concentration that alleviate the adverse effect of sodium chloride and gave the best values for parameters studied.

## MATERIALS AND METHODS

Pots experiment was conducted at green house of botanical garden belong to Department of Biology, College of Education for pure Science, Ibn-AL-Haithum, University of Baghdad on the growing season 2018-2019 to study the effect of  $\alpha$ -tocopherol foliar application on growth parameters of wheat plant (AL-Baraka cultivar) irrigated with sodium chloride concentrations. The experiment was arranged in completely randomized design (CRD) 4x5 with three replicates. Fifteen uniform wheat grains (bring from the General Authority for Agricultural Research) were sown on 12-11-2018 in pottery pots containing 11 Kg soil, two weeks after sowing the seedling were thinned to twelve uniform seedling per pots. Plants were irrigated at 30-12-2018 with sodium chloride concentrations (0, 75, 150, 225mM.L<sup>-1</sup>), plants were sprayed with  $\alpha$ -tocopherol concentration (0, 50, 100, 150, 200mg.L<sup>-1</sup>) at 9-1-2019 in the early morning. Four randomly selected plants were carefully harvest per pots 22-1-2019, plant height (cm) were measured, flag leaf area (cm<sup>2</sup>) were calculated according to (Mckee, 1964) flag leaf chlorophyll content were measured using (SPAD), Superoxide dismutase (SOD) was estimated according to method (Beyer and Fridovich, 1987), Peroxidase (POD) was determined according to Nezh (1985). The data were analyzed using analysis of variance ANOVA using statistically computer programe (SAS, 2012). Significance between treatment were compared at 0.05 probability level.

## RESULTS AND DISCUSSION

Data in Table 1 indicated that average of plant height was significantly reduced by increasing sodium chloride concentrations up to 225 mM.L<sup>-1</sup>, the reduction ratio was 15.85% in contrast application of  $\alpha$ -tocopherol concentration from 0 to 200 mg.L<sup>-1</sup> caused significant increase for parameter average and the best increase was at concentration 150 mg.L<sup>-1</sup>  $\alpha$ -tocopherol by about 18.85%. The combination between the two factors, data illustrated that the concentration 225 mM.L<sup>-1</sup> sodium chloride with the concentration 150 mg.L<sup>-1</sup>  $\alpha$ -tocopherol, the value of plant height was 65.25 cm compared with the value 48.51 cm at the same concentration of sodium chloride but without  $\alpha$ -tocopherol spraying. The highest

value for plant height was 73.80 cm at the concentration 150 mg.L<sup>-1</sup>  $\alpha$ -tocopherol and without sodium chloride in comparison with the value 64.20 cm at control treatment.

Data in Table 2 showed that increasing sodium chloride concentration from 0 to 225mM.L<sup>-1</sup> decreased average of flag leaf area by about 23.33%. It can be indicated that a positive response to  $\alpha$ -tocopherol application on average of flag leaf area by increasing the concentrations from 0 to 200 mg.L<sup>-1</sup>  $\alpha$ -tocopherol, the increase ratio was 25.10%. The difference between the average of flag leaf area was not significant at the concentration 150, 200 mg.L<sup>-1</sup>  $\alpha$ -tocopherol. Regarding the combination between the two factors,  $\alpha$ -tocopherol at the concentration 150 mg.L<sup>-1</sup> was positive that can diminish the adverse effect of 225 mM.L<sup>-1</sup> sodium chloride on flag leaf area and gave the value 43.50 cm<sup>2</sup>, in comparison with the value 27.14 cm<sup>2</sup> for flag leaf area at the same sodium chloride concentration, but without spraying with  $\alpha$ -tocopherol. The highest value for the parameter was 52.25 cm<sup>2</sup> at the concentration 150 mg.L<sup>-1</sup>  $\alpha$ -tocopherol and without sodium chloride in comparison with the value 44.61 cm<sup>2</sup> at control treatment.

Data Presented in Table 3 indicated that salinity had pronounced effect on average of flag leaf chlorophyll content, increasing concentrations of sodium chloride up to 225 mM.L<sup>-1</sup> reduced the average of parameter, the reduction ratio 18.65%,  $\alpha$ -tocopherol foliar spraying from 0 to 200 mg.L<sup>-1</sup> enhance average of flag leaf chlorophyll content by about 27.62%, the difference between the average for flag leaf chlorophyll content was not significant at the concentrations 150, 200 mg.L<sup>-1</sup>  $\alpha$ -tocopherol. The interaction between the two factors was illustrated, the value for flag leaf chlorophyll content at the treatment of height sodium chloride concentration 225 mM.L<sup>-1</sup> and concentration 200 mg.L<sup>-1</sup>  $\alpha$ -tocopherol 41.32 SPAD comparing with the value 28.33SPAD at the same sodium chloride concentration but without  $\alpha$ -tocopherol spraying. The difference was not significant between the values for flag leaf chlorophyll content at the concentration 150, 200 mg.L<sup>-1</sup>  $\alpha$ -tocopherol and 225 mM.L<sup>-1</sup> sodium chloride. The highest value for the parameter was 46.30 SPAD at the concentration 150 mg.L<sup>-1</sup>  $\alpha$ -tocopherol and without sodium chloride in comparison with the value 40.50 SPAD at control treatment.

To determine whether antioxidant enzymes enhanced in response to salinity stress, Table 4 shows that average of superoxide dismutase activity was characterized by increasing with increasing sodium chloride concentration up to 225 mM.L<sup>-1</sup> with increase ratio 39.13%. Application of  $\alpha$ -tocopherol up to 200 mg.L<sup>-1</sup> increased average of

**Table 1 :** Role of  $\alpha$ -tocopherol in wheat plant height (cm) subjected to salinity stress.

Sodium chloride concentrations (mM.L <sup>-1</sup> )	$\alpha$ -tocopherol concentrations (mg.L <sup>-1</sup> )					Sodium chloride average
	0	50	100	150	200	
0	64.20	66.60	68.00	73.80	73.55	69.23
75	61.15	63.00	65.38	68.90	66.21	64.93
150	58.35	62.20	64.21	68.00	65.33	63.62
225	48.51	54.20	60.34	65.25	63.00	58.26
$\alpha$ -tocopherolaverage	58.05	61.50	64.48	68.99	67.02	
LSD(0.05)	Sodium chloride concentrations=1.55, $\alpha$ -tocopherol concentrations = 1.73, Interaction = 3.45					

**Table 2 :** Role of  $\alpha$ -tocopherol in flag leaf area (cm<sup>2</sup>) of wheat plant subjected to salinity stress.

Sodium chloride concentrations (mM.L <sup>-1</sup> )	$\alpha$ -tocopherol concentrations (mg.L <sup>-1</sup> )					Sodium chloride average
	0	50	100	150	200	
0	44.61	46.54	48.62	52.25	50.62	48.53
75	40.42	42.97	44.59	45.90	46.77	44.13
150	36.83	40.52	43.21	44.66	45.90	42.22
225	27.14	33.82	38.50	43.50	43.10	37.21
$\alpha$ -tocopherolaverage	37.25	40.96	43.73	46.58	46.60	
LSD(0.05)	Sodium chloride concentrations=0.98, $\alpha$ -tocopherol concentrations= 1.09, Interaction= 2.19					

superoxide dismutase activity with the superiority to concentration 150 mg.L<sup>-1</sup>  $\alpha$ -tocopherol for giving the highest average for enzyme activity with increase ratio 41.79%. The interaction between the dual factors was significant,  $\alpha$ -tocopherol at the concentration 150 mg.L<sup>-1</sup> can minimize the adverse effect of 225 mM.L<sup>-1</sup> sodium chloride and raised superoxide dismutase activity and gave the best value 200.81 Unit.ml<sup>-1</sup> in comparison with the value for enzyme activity 168.42 Unit.ml<sup>-1</sup> at the same sodium chloride concentration but without  $\alpha$ -tocopherol spraying and with the value of enzyme activity 80.76 Unit.ml<sup>-1</sup> at control treatment.

Data in Table 5 shows increase in average of peroxidase activity with increasing concentrations of sodium chloride from 0 to 225 mM.L<sup>-1</sup> with increase ratio 121.72%,  $\alpha$ -tocopherol foliar application increase average of enzyme activity by increasing concentrations of  $\alpha$ -tocopherol from 0 to 200 mg.L<sup>-1</sup>, the superiority was to concentration 100 mg.L<sup>-1</sup>  $\alpha$ -tocopherol that gave the highest average for peroxidase activity with increase ratio 51.77%. The combination between the two factors was significant, the value for enzyme activity at the concentration 225 mM.L<sup>-1</sup> sodium chloride and 100mg.L<sup>-1</sup>  $\alpha$ -tocopherol was 363.00 Unit.ml<sup>-1</sup> in comparison with the value 270.00 Unit.ml<sup>-1</sup> at the same sodium chloride concentration but without spraying with  $\alpha$ -tocopherol and with the value 100 Unit.ml<sup>-1</sup> at control treatment.

Salt stress can lead to stomata closure ,reduces gasses

changes, that limit CO<sub>2</sub> supply to the leaf, caused over reduction of photosynthetic electron transport chain leading to height production of ROS and induced oxidative stress (Perez-lopez, 2009). The toxic ROS oxidize proteins, DNA, lipids, reduce chlorophyll content and leaf area lead to leaf senescence which disturbed photosynthesis, associated with nutrient relocation to grain and cause reduction in plant growth (Hameed *et al*, 2008). The reduction in chlorophyll content can be due to the reduction in its synthesis or increase chlorophyll degradation in the leaves by enhancing the activity of chlorophyllase, or due to the plant stopping chlorophyll biosynthesis and activate the biosynthesis of  $\alpha$ -tocopherol non-enzymatic antioxidant (Zhao *et al*, 2007). To prevent cellular components damaging, plant have develop many detoxification mechanisms such as antioxidant enzymes including superoxide dismutase (SOD), peroxidase (POD), both enzymes can play vitalrole in alleviating the adverse effects of salinity stress (Flak and Munn-Bosch, 2010). SOD is an importantenzyme, it is the first line of defense that convert superoxide radical in to hydrogen peroxide and water and prevent hydroxyl radicals formation, which are responsible for lipid peroxidation (Abogadalla, 2010). Hydrogen peroxide is converted to H<sub>2</sub>O byperoxidase (Chen and Jiang, 2010).

Foliar application of  $\alpha$ -tocopherol affect photosynthesis, regulate jasmonicacide in leave, control cellular redox state in chloroplast and enhance antioxidant

**Table 3 :** Role of  $\alpha$ -tocopherol in flag leaf chlorophyll content (SPAD) of wheat plant subject to salinity stress.

Sodium chloride concentrations (mM.L <sup>-1</sup> )	$\alpha$ -tocopherol concentrations (mg.L <sup>-1</sup> )					Sodium chloride average
	0	50	100	150	200	
0	40.50	43.75	44.71	46.30	45.92	44.24
75	36.50	39.82	41.87	43.28	44.72	41.24
150	32.24	37.77	39.12	41.00	43.60	38.75
225	28.33	33.27	36.60	40.41	41.32	35.99
$\alpha$ -tocopherolaverage	34.39	38.65	40.57	42.75	43.89	
LSD(0.05)	Sodium chloride concentrations=1.52, $\alpha$ -tocopherol concentrations =1.69, Interaction=3.39					

**Table 4 :** Role of  $\alpha$ -tocopherol in superoxide dismutase activity (Unit.ml<sup>-1</sup>) of wheat plant subjected to salinity stress.

Sodium chloride concentrations (mM.L <sup>-1</sup> )	$\alpha$ -tocopherol concentrations (mg.L <sup>-1</sup> )					Sodium chloride average
	0	50	100	150	200	
0	80.76	125.29	134.21	160.65	150.63	130.31
75	128.20	160.17	190.77	194.79	193.37	173.46
150	153.00	163.15	197.04	195.77	177.52	177.30
225	168.42	173.29	183.27	200.81	180.73	181.30
$\alpha$ -tocopherolaverage	132.60	155.48	176.32	188.01	175.56	
LSD(0.05)	Sodium chloride concentrations =1.93, $\alpha$ -tocopherol concentrations=2.16, Interaction= 4.32					

**Table 5 :** Role of  $\alpha$ -tocopherol in peroxidase activity (Unit.ml<sup>-1</sup>) of wheat plant subjected to salinity stress .

Sodium chloride concentrations (mM.L <sup>-1</sup> )	$\alpha$ -tocopherol concentrations (mg.L <sup>-1</sup> )					Sodium chloride average
	0	50	100	150	200	
0	100.00	128.30	140.67	172.33	146.34	137.53
75	180.34	199.33	281.34	236.67	205.33	220.60
150	210.67	247.33	370.00	277.00	250.00	271.00
225	270.00	288.67	363.00	311.00	292.00	304.93
$\alpha$ -tocopherolaverage	190.25	215.91	288.75	249.25	223.42	
LSD(0.05)	Sodium chloride concentrations=1.92, $\alpha$ -tocopherol concentrations=2.15, Interaction=4.30					

system (Hunter and Cahoon, 2007).  $\alpha$ -Tocopherol increased total chlorophyll through delayit's degradation and stimulate its biosynthesis, this increase might be related to antioxidant enzymes that scavenge ROS, because chloroplast is amajor source of ROS production (Sakr and El-Matwally, 2009).  $\alpha$ -tocopherol protect metabolic processes, cell membrane against the damage caused by ROS, it increase the activity of many enzymes, minimizes the adverse damage caused by oxidative processes through synergic function with several antioxidants that is lead to healthy growth and good yield under effect of salt stress (Shao *et al*, 2008). Our results showed that sodium chloride with height concentration caused reduction in plant height, flag leaf area, flag leaf chlorophyll these finding agree with Al-Saedy *et al* (2012).  $\alpha$ -Tocopherol foliar application can alleviate the adverse effect of salinity stress and reflected significantly on growth parameters mentioned and increased antioxidant

enzymes, these finding agree with Farouk (2011). The highest values of growth parameters was with concentration 150 mg.L<sup>-1</sup>  $\alpha$ -tocopherol that minimized the adverse effects of the concentration 225 mM.L<sup>-1</sup> sodium chloride.

## CONCLUSION

Sodium chloride with height concentration caused reduction in plant height, flag leaf area and flag leaf chlorophyll.  $\alpha$ -Tocopherol foliar application can alleviate the adverse effect of salinity stress and reflected significantly on growth parameters mentioned and increased antioxidant enzymes.

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