

Effect of Salinity and ascorbic acid treatments on growth, yield and antioxidant enzymes activity of barley plant

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ABSTRACT

A pot experiment was conducted at the Faculty of Science (Girls, Al-Azhar University, Cairo, Egypt) during the winter season (November 2020) to study the efficiency of ascorbic acid (100 and 200 ppm) in improving some growth parameters, yield and its components as well as some enzymatic activities of barley plants grown under salinity stress (2000, 4000 and 8000 ppm NaCl). In general, salinity stress resulted in reduced growth parameters (plant height, number of tillers/plant, fresh and dry weight of shoot/plant) associated with a decrease in yield components (weight of spike, length of spike, number of grains/spike and weight of 100 grains) of barley plants, these reductions were gradually increased with increasing the concentration of salinity. Meanwhile, the antioxidant enzymes (peroxidase, polyphenol oxidase, superoxide dismutase, and catalase) significantly increased under salinity conditions. Foliar application of ascorbic acid, 100 and 200 ppm, on plants grown under normal conditions or grown under salinity levels showed a significant increase of all growth parameters, yield and its components as well as the antioxidant enzymes activity. It can be concluded that foliar application of ascorbic, especially at 200 ppm was the most effective treatment in amelioration the depressive effect of salinity on some growth parameters, yield characters, and some antioxidant enzymatic activities of barley plants.

Keywords: Salinity; Ascorbic acid; Barley plant.

INTRODUCTION:

More than 10000 years ago, people began growing barley for use as animal feed, drinks, soups, and barley bread (Aksakal, 2013), In addition, in comparison to other cereal crops, barley is very tolerant to salt. Nonetheless, barley has been the subject of numerous studies regarding abiotic stress environments (Ullah *et al.*, 2016). Additionally, barley is regarded as a salt excluder that restricts the movement of Na⁺ from the shoot to the leaves (EL-Sharkawyet *et al.*, 2017).

It is well-recognized that saline soils inhibit plant growth by causing osmotic stress, which is accompanied by ion toxicity. Furthermore, the detrimental effect of crucial minerals like Na and Cl on the growth and development of plants is known as salt stress (Dawood and EL-Awadi, 2015). Salinity stress affects several metabolic and physiological processes and thus eventually lowers crop output, depending on the length of time under stress and the severity of the stress (Kazemi *et al.*, 2019). Moreover, Safdar *et al.* (2019) cited that ion toxicity and osmotic stress are the two main contributors to salt stress in plants. Large levels of soluble salts depositing in the soil and reducing the amount of water available to plants cause osmotic stress; ion toxicity results from the accumulation of high salt content

within plants, this leads to the disruption of several metabolic process which includes the inactivation of specific enzymes.

Ascorbic acid is a water-soluble vitamin that effectively reduces the yield of reactive oxygen species (ROS) created in response to different abiotic stresses as well as combats oxidative stress (Foyer and Noctor, 2011). Additionally, Fatima *et al.* (2019) reported that ascorbic acid considered as vitamin C, a potent non-enzymatic antioxidant that is essential to regulating plant development and response to stress. Ascorbic acid is a cofactor for several cellular enzymes, including violaxanthin de-epoxidase, which is essential to photoprotection by the xanthophyll cycle and other enzymes and is directly involved in the removal of ROS. Addition of exogenous Ascorbic acid prevents lipid peroxidation and lowers malondialdehyde (MDA) content in plant tissue which will improve the antioxidant capacity of plant tissues (Zhou *et al.*, 2016).

The main objective of the current study was to investigate the efficiency of ascorbic acid to alleviate the adverse effect of salinity on some growth parameters, yield, and its components as well as some antioxidant enzymatic activities of barley plants.

MATERIALS AND METHODS:

Experimental design:

The present investigation was carried out at the Faculty of Science (AL- Azhar University, Cairo, Egypt- Girls) during the winter season (November 2020). A pot experiment was conducted to evaluate the effect of different salinity levels at 0, 2000, 4000, and 8000 ppm NaCl as well as foliar spray of ascorbic acid at 100 and 200 ppm beside the control and their interaction effect on growth parameters, yield components and some antioxidant enzymes activity of barley plants.

We recommended the salinity levels according to El-Sebai *et al.* (2016) , they used 4000 and 8000 mg/l NaCl on quinoa plant. Also, Hassan *et al.* (2021) applied 50, 100 and 150 mM on barley plants.

Noreen *et al.* (2020) impact 200 ppm of ascorbic acid on barley plant under salinity stress and Seleem *et al.* (2021) applied 100 and 300 ppm ascorbic acid on barley plant under salinity stress.

The plants were fertilized with NPK at the rate of 80 kg N, 30 kg P₂O₅, and 15 kg K₂O per feddan, which were added in the form of ammonium nitrate (33.5%), superphosphate, and potassium sulfate were added before planting, whereas nitrogen was added at three equal times, one before planting and the other two at three- week intervals, as recommended by the Agriculture Ministry.

Barley grains (*Hordeum vulgare* L.) Giza 134 was obtained from the Agriculture Research Center, Giza, Egypt. For grain sterilization, the grains were immersed in 95% ethanol for 5 minutes, washed three times in sterile deionized water, followed by soaking in 1.5 percent sterile sodium hypochlorite for 10 minutes, and then thoroughly rinsed with distilled water. Then, grains were planted on 15 November 2020 in porcelain pots (these pots contained loamy soil as recorded in table (1), after 10 days from emergence seedlings were thinned to 10 seedlings/pot.

The experiment was arranged in a split-plot design with three replicates, each containing 5 pots.

The pots were irrigated with various levels of salinity, and the control plants were watered with tap water to keep the soil at 70% of the field capacity. Foliar application of ascorbic acid was carried out three times (30, 50, and 75 days after sowing).

Physical and chemical properties of the soil before cultivation are analysis according to

Jackson (1973) and Page *et al.* (1982) presented in Table (1).

Growth parameters:

All samples were collected for different morphological characters, about 45 days (first sample), 65 days (second sample), and 90 days (third sample) from planting. Growth parameters for three samples were recorded, plant height (cm), number of tillers/plant, fresh weight (g/plant), and dry weight/ plant. To measure morphological parameters, ten plants were randomly selected from each treatment with the assistance of a scale, and the length of each replicate's shoot was determined.

Dry weight (g/plant) was measured as follows: each treatment's plant shoots were rinsed with distilled water before being dried in an oven at 70 C until they reached a consistent dry weight; the dry weights were expressed as g/plant.

Yield characters:

Yield characters were recorded after spike maturation (130-day-old plant) as follows: weight of spike (gm) - length of spike (cm) - number of grains/spike - weight of 100 grains (gm).

Estimation of antioxidant enzymes:

Terminal buds, in addition to the first and second young leaves, were the plant materials utilized in the estimation of peroxidase (POX), polyphenol oxidase (PPO), superoxide dismutase (SOD), and catalase enzyme. To do this, 2 g of the plant materials were mixed with 10 ml of phosphate buffer pH 6.8 (0.1 M), and they were then centrifuged in a chilled centrifuge at 2^o C for 20 min at 20,000 rpm. The enzyme supply was determined to be the clear supernatant, which included the enzymes (Mukherjee and Choudhuri, 1983).

Assay of peroxidase (POX) activity: POX was estimated according to Bergmeyer (1974).

Assay of Polyphenol oxidase (PPO) (s) activities: PPO activities were estimated according to Kar and Mishra (1976).

Assessment of Superoxide Dismutase (SOD) Productivity: Using a technique outlined by Marklund and Marklund (1974), sod activity was measured by assessing the suppression of the auto-oxidation of pyrogallol.

Assay of catalase (CAT) activity: catalase activity was assayed according to the method of Chen *et al.* (2000).

Statistical Analysis:

Computer applications Microsoft Excel version 365, SPSS version 25, and Minitab version 19 were used to do statistical calculations. At the probability range of 0.05 (Snedecor & Cochran, 1982). One-way ANOVA and Post hoc Tukey's test were used for the analysis of variance while analyzing quantitative data with parametric distribution. There was a margin of error allowed of five percent and a confidence interval of ninety-five percent.

RESULTS AND DISCUSSION:

Growth characters:

Table (2) represents data about the effect of different saline levels (2000, 4000, and 8000 ppm NaCl) on some plant growth parameters (plant height, number of tillers/plant, fresh weight, and dry weight of shoot) in barley plants (*Hordeum vulgare* L.). Results showed that all studied growth parameters at three studied stages were significantly decreased gradually by increasing salinity levels in comparison to control plants. For instance, irrigation of barley plants with 4000 ppm NaCl decreased shoot height by 9.35%, number of tillers/plant by 22.08%, fresh weight by 49.59%, and dry weight by 54.84%, at first sample compared to control plants. In addition, the same trend was also detected for the second and third sa

According to Pakar *et al.* (2016), plant height and dry weight of barley were severely lowered by salinity concentrations at 15 dsm-1. El Sharkawy *et al.* (2017) discovered that salt treatments at 10 and 15 dsm-1 decreased the shoot dry weight of barley for two distinct genotypes (tolerant and sensitive). Additionally, Ibrahim *et al.* (2019) reported that geranium plants gradually lost height, branch count, and fresh weight when saline levels were raised to 500 ppm. This reduction was observed in comparison to control plants.

The fact that salinity causes the accumulation of some ions and the lack of others, as well as lowering the external water potential in the cell, is consistent with the reduction of plant development under salt stress. Furthermore, a disruption in metabolic processes brought on by a decrease in water absorption and an imbalance in the equilibrium of water may be the cause of the decline in plant development. (Fahad *et al.*, 2015). In addition, Abd El-Monem (2010) observed that the decrease in growth could perhaps be attributed to the disruption of

phytohormone levels caused by the effects of salinity on plant hormone biosynthesis or destruction. They claimed that a significant drop in the levels of cytokinin and indole acetic acid caused a quick build-up of abscisic acid. Osmotic stress from excessive salinity accelerates water loss from leaves during the early stages of salinity stress, reducing the root system's ability to absorb water (Farooq *et al.*, 2019).

The excessive salt content is toxic to plants because it inhibits their ability to grow and develop because salinity hinders the uptake of several crucial nutrients from the soil. Furthermore, plants' ability to grow and develop is ultimately hindered if a large concentration of salt builds up in their transpiration stream, causing damage to transpiring leaves (Parihar *et al.*, 2015). Furthermore, Zafar *et al.* (2015) observed that salinity stress causes a noticeable reduction in plant growth due to the germination medium's reduced osmotic capacity, which in turn induces toxicity that changes the activities of enzymes involved in the metabolism of nucleic acids, modifications in protein metabolism, disruption of hormonal balance, and reduced utilization of seed reserves. The increase in Na⁺ and decrease in K⁺ uptake by plants resulted in a reduction in the vegetative biomass. This leads to the plant being in an osmotic stress state, due to the high levels of Na⁺ in the soil, thus impeding the ability of plants to uptake water from the soil (Zeeshan *et al.*, 2020). Reduced cell elongation as a result of the water shortage as well as the inhibitory influence on growth-promoting hormones may be the cause of a reduction in plant growth in response to water stress. Furthermore, the decrease in cell turgor, volume, and growth ultimately results from reduced water availability.

The effect of ascorbic acid at 100 and 200 ppm on the mentioned growth parameters of barley plants during different growth stages was illustrated in Table (3). Foliar spray of ascorbic acid (100 and 200 ppm) caused a significant increase in plant growth parameters under normal irrigation at all growth stages. The effect is more definite at 200 ppm ascorbic acid.

Application of ascorbic acid at low concentration (100 ppm) increased plant height by 4.62 %, number of tillers/plant by 17.2 %, fresh weight by 27.38 %, and dry weight by 47.37 %, at first sample compared to unsprayed barley plants. Moreover, using a high concentration of ascorbic acid (200 ppm)

caused an increment in plant height by 7.87 %, number of tillers by 26.6 %, fresh weight by 97.02 %, and dry weight by 89.47 %, in the same mentioned sample.

The same trend was also observed in the second and third samples.

The results obtained in the present investigation concerning the effect of ascorbic acid on plant growth are in agreement with previous data obtained by Loutfy *et al.* (2019) on fava bean plants; Gaafar *et al.* (2020) on common bean plants; Hassan *et al.* (2021) on barley plants.

Abdel-Hafeez *et al.* (2019) showed that the measurements of growth parameters (plant height, number of leaves/plant, shoot dry weight, and leaf area) in sunflower plants were greatly raised when the addition of 1mM of ascorbic acid in sunflower plants. Ascorbate or ascorbic acid, is the most prevalent antioxidant, it contributes to the plant's redox system by acting as an electron donor in many processes (Bilska *et al.*, 2019). Ascorbic acid is also essential for cell metabolism and the synthesis of proteins containing hydroxyproline (Al-Hakimi and Hamada, 2011). Ascorbic acid may help cells proliferate by controlling the cell cycle and encouraging quiescent cell division and elongation (Akram *et al.*, 2017). Since ascorbic acid is a key regulator of plant growth, it increases the amount of IAA, which promotes cell division and regulates the development and growth of plants (Khan *et al.*, 2011). In addition, ascorbic acid is very important for the regulation of photosynthesis, flowering and senescence (Barth *et al.*, 2006). Ascorbic acid would affect the metabolism of plant reactions which accounted for adaptabilities that increase the resistance of plants against the environmental factors (Metwally *et al.*, 2003). El-Kobisy *et al.* (2005) reported that reactive oxygen species (ROS) can be scavenged by ascorbic acid, ROS are known to impair the growth of plants. It is a byproduct of the metabolism of D-glucose and is essential to the electron transport system as well as influencing certain aspects of the nutritional cycle in higher plants.

Results recorded in Table (4) illustrated the interactive effect between NaCl concentrations in irrigation water and the application of ascorbic acid as a foliar spray on the growth parameters of barley plants at three growth stages (45,65 and 90 days after planting). Data showed that the application of ascorbic acid at both concentrations (100 and 200 ppm) had a positive and significant increment of all

growth parameters in barley plants grown under different salinity levels (2000, 4000. and 8000 ppm NaCl). For instance, plant height, the number of tillers/plant, fresh, and dry weights were increased by 9.99 %, 20.18 %, 71.77 %, and 121.43 %, respectively at the first sample in plants grown under 4000 ppm NaCl and sprayed with 200 ppm ascorbic acid, compared to salinized unsprayed plants.

Data showed that the application of ascorbic acid especially at 200 ppm seemed to be more effective than 100 ppm for increasing all growth parameters of plants grown under saline conditions in comparison to salinized unsprayed plants. It is true to mention that the application of ascorbic acid ameliorated the negative effect of salinity in all growth parameters of barley plants.

Ascorbic acid is one of the antioxidant enzymes that has synergistic effects on the growth of many plant species. According to Foyer *et al.* (1991), these substances help absorb radicals or active oxygen that is created during photosynthesis and respiration. According to the same pattern, Barakat (2003) found that applying ascorbic acid directly to plants improved their physiological traits, increasing their resistance to stress and allowing them to absorb more water and nutrients. The potential use of exogenous antioxidant application to mitigate the negative impacts of stress on plants and enhance their growth and development has garnered significant interest (El-Bassiouny *et al.*, 2005). Ascorbic acid regulates cell proliferation and mitosis in plants. It also has an impact on signaling mechanisms mediated by phytohormones during the development and senescence stages, as well as the transition from the vegetative to the reproductive phase (Barth *et al.*, 2006) Furthermore, as demonstrated by Wang and Kao (2007), ascorbic acid is thought to mitigate the harmful effects of salt on plants during biotic and a biotic stress. The favorable effect of ascorbic acid on plant height may be related to the fact that ascorbic acid plays a role in regulating the processes of shoot and root enlargement, cell vacuole, leaf area, and cell growth (Farahat *et al.*, 2013). Moreover, Hassan *et al.* (2022) found that high soil salinity (150 mM) depressed plant growth and biomass in barley plants (plant height, leaf area, number of leaves, fresh and dry biomass), but the foliar addition of ascorbic acid (30 and 60 Mm) of salinized plants enhanced plant growth and development.

Yield components:

The pattern changes in several yield components of barley plants including spike length (cm), the weight of 100 grains, number of grains per spike, and weight of grains per spike were significantly reduced by increasing the concentration of NaCl in growth media (Table 5). The most decreased percentage in yield components of barley plants was in plants grown under high salinity levels (8000 ppm NaCl), which caused a reduction in the weight of spike, length of the spike, number of grains /spike, and weight of 100 grains by 45.41 %, 14.12 %, 22.12 %, and 24.41 %, respectively compared to control plants.

These results are in harmony with those obtained by Hafez and Gharib (2016) on wheat plants, El-Beltagi *et al.* (2020) on chickpea plants, and Seleem *et al.* (2021) on barley plants.

The detrimental effects of salt on growth and the interruption of mineral uptake could be responsible for of the decrease in wheat yield/plant induced by salinity stress (Abd El-Haleem *et al.*, 1995). According to Bybordi (2010), barley could suffer reproductive difficulties as a result of salinity, which would therefore influence yield output. Changes in osmotic potential brought on by decreased water content together with specific harmful effects from the formation of salt and chloride ions, which are seen in many plants, could be the reason of the reduction in yield components (Abu-Muriefah, 2015). According to Pakar *et al.* (2016), this disorder may be caused by an interruption in the photosynthetic machinery, which in turn influences assimilate synthesis and glucose metabolism. Additionally, El-Sebai (2016) found that quinoa plants progressively decreased yield components such as shoot length, number of fruiting branches per plant, and seed weight per plant when salt levels increased to 0.0, 4000, and 8000 mg/l. According to Seleem *et al.* (2021), barley plants treated with salt at 9.3 and 14 dsm⁻¹ NaCl showed a substantial decrease in all yield parameters when compared to control plants

Data presented in Table (6) declare the effect of ascorbic acid (100 and 200 ppm) on yield components of barley plants. Both concentrations of ascorbic acid had a positive and significant increment at all yield components of barley plants. The highest concentration of ascorbic acid (200 ppm) increased yield components as follows: weight of spike (23.81 %), length of spike (6.70 %), number of grains /spike (8.15 %), and weight

of 100 grains (32.04 %) compared to unsprayed plants.

The favorable effects of ascorbic acid on yield parameters were reported in grain weight by Dolatabadian *et al.* (2010), which is consistent with the results of our study. Furthermore, when ascorbic acid at 300 ppm was applied to wheat cultivars, Bakry *et al.* (2013) showed a substantial increase in all yield features, however, Desoky and Merwad (2015) observed that ascorbic acid at 0.2% caused a significant increase in just grain yield/plant and seed index. Ascorbic acid affects sunflower plant species' yield and yield quality synergistically. Ascorbic acid is one of the most significant water-soluble antioxidants in plants, functioning as a coenzyme in reactions that metabolize proteins, fats, and carbohydrates as well as a modulator of plant development through hormone signaling. These compounds have positive effects on capturing free radicals or the active oxygen that is produced during photosynthesis and respiration processes (Maiorana *et al.*, 2005).

According to Hafez and Gharib (2016), ascorbic acid's contribution to the transfer of metabolites from leaves into reproductive organs may explain its beneficial effects on grain and straw yields. Furthermore, wheat plants yield more grain and straw when protein and nucleic acid synthesis proceeds.

Results recorded in Table (7) showed the interactive effect between salinity levels in irrigation water and the application of ascorbic acid on measured yield parameters. It can be noticed that, ascorbic acid applications considerably increased yield components of barley plants grown under different salinity concentrations at the harvest time (130-day-old plants). The best increment values were recorded in plants treated with 200 ppm ascorbic acid and grown under 8000 ppm NaCl by 38.32% in weight of spike, 9.40% in length of spike, 15.26% in number of grains/spike, and 41.63% in weight of 100 grains.

The depressive effect of salinity on grain yield that resulted from the harmful outcome of salt stress on barley plants and positive increases of yield and its components in response to ascorbic acid treatments are in agreement with this obtained by Gaafar *et al.* (2020) on common bean plants and Seleem *et al.* (2021) on barley plants.

The foliar treatment of ascorbic acid could reduce the negative impacts of salt stress on the quinoa plants' yield and yield-related characteristics when compared to the

equivalent salinity levels that were being tested. Furthermore, the alterations observed could be related to the rise in the absorption and use of nutrients. Furthermore, the impact of antioxidants on promoting protein synthesis and postponing senescence might be the cause of the rise in yield and its constituent parts (Hammam et al., 2001). In addition, according to Mittler (2002), ascorbic acid is a non-enzymatic antioxidant that functions as a reaction substrate in the enzymatic cycle and as an electron donor to decrease the buildup of reactive oxygen species (ROS). According to Athar et al. (2009), ascorbic acid plays a critical role in enhancing flag leaf area and leaf chlorophyll content, which in turn promotes vegetative and reproductive growth, the accumulation of carbohydrates, and the seed set. This helps wheat plants yield grown under water stress. Ascorbic acid is essential for the maintenance of multiple metabolic pathways in stressed plants (Sadak et al., 2010). Ascorbic acid has been proven to be essential for both cell growth and division as well as for enhancing antioxidant enzymes, which counteracts the inhibitory effects of oxidative stress (Agami, 2014). According to Farooq et al. (2015), small grain size and weight during water stress may be caused by decreased photosynthesis, rapid leaf senescence, and sink restriction, which all contribute to yield reduction. Finally, Noreen et al. (2020) found that foliar spray of ascorbic acid on barley plants grown under saline conditions improved yield components.

Antioxidant enzymes:

Data in Table (8) displays the effect of different salinity levels (2000, 4000, 8000 ppm) on the activities of a few antioxidant enzymes (peroxidase, superoxide dismutase, polyphenol oxidase, and catalase) in leaves of barley plants at first sample (45 days after sowing) and second sample (65 days after sowing) during the tested period. The activities of these enzymes were significantly increased as a result of salinity treatments compared to un-salinized control plants.

The higher increment percentage in enzymatic activities was recorded in barley plants grown under salinity level (8000 ppm NaCl) at both growth stages compared to un-salinized control plants. For instance, the increment percentage of POX, PPO, SOD, and CAT was 82.35, 91.30, 84.62 and 85.71%, respectively in the first sample compared to control as a result of salinity stress, and the same trend was detected in the second sample.

These results are in parallel with Seleem et al. (2021) who recorded a significant increment for antioxidant enzymes (CAT and POD) activities in leaves of barley plants under both salinity levels (9.3 and 14 dsm-1 NaCl).

According to Moussa and Abdel-Aziz (2008), protection against oxidative damage, lignifications, and cross-linking of the cell wall are associated with increased peroxidase (POD) activity under different stress conditions. Furthermore, in a soil salinity environment, an imbalance between the generation and elimination of ROS poses the greatest hazard to plant cells; yet, extra ROS creation is also believed to function as a signaling molecule to the active plant defense system (Saleem et al., (2019).

Increases in reactive oxygen species (ROS) may be the cause of the increases in antioxidant enzyme activity, which may lead to the peroxidation of numerous vital cellular components, including lipids, chlorophyll, and pigments involved in photosynthetic processes (Ali et al., 2020). In keeping with this, Hassan et al. (2021) discovered that in contrast to plants grown in non-saline soil, barley plant grown in salinity (0, 50, 100, and 150 mM NaCl) exhibit significantly higher activity of antioxidant enzymes (SOD, POD, CAT, and APX).

Results presented in Table (9) demonstrated the effect of ascorbic acid treatments (100 and 200 ppm) on antioxidant enzyme activities (POD, PPO, SOD, and CAT) in leaves of barley plants. Antioxidant enzyme activities were significantly increased under both treatments of ascorbic acid, while 200 ppm ascorbic acid was more effective and the high increment percentage was 76.8% for SOD and 50% for POD at the heading stage.

Madany and Khalil (2017) discovered that ascorbic acid treatment enhanced the activity of antioxidant enzymes (PAL, POX, and PPO) in sunflower plants. According to Athar et al. (2009), using ascorbic acid may help protect plants from oxidative damage brought on by water stress. As the primary substrate of APX, an important enzyme in the ascorbate-glutathione pathway, ascorbic acid serves as plants' first line of defense against oxidative stress by eliminating several free radicals, including O₂, OH⁻, and H₂O₂. (Sharma et al., (2019).

Data presented in Table (10) showed the interactive effect between different concentrations of salinity and ascorbic acid at 100 and 200 ppm on some activities of

antioxidant enzymes (POD, PPO, SOD, and CAT) at the heading and tillering stages in leaves of barley plants.

The results illustrate a pronounced and significant increase of antioxidant enzyme activity which was detected in leaves of barley plants sprayed with 200 ppm ascorbic acid and grown under 8000 ppm NaCl, compared to the control one. These results are in harmony with, El-Sayed *et al.* (2015) who found that Catalase, peroxidase, and superoxide dismutase activities were all markedly elevated by salinity levels on sweet pepper plants in the presence or absence of ascorbic acid; however, the activity was increased more significantly in the presence of ascorbic acid than in control plants. In the same sense, according to Hafez and Gharib (2016), ascorbic acid treatment and water stress both enhanced the activity of antioxidant enzymes (CAT and POX) in wheat plants. According to Younis *et al.* (2010), exogenous administration of ascorbic acid improved plant survival under overall environmental stress by dramatically increasing the activity of most antioxidant enzymes, including SOD, POD, CAT, and APX, under NaCl stress conditions. This could be because ascorbic acid, one of the most effective non-enzymatic antioxidants available, can both scavenge reactive oxygen species (ROS) and modify several essential plant processes in both stressed and unstressed environments (Akram *et al.*, 2017). Treatment of quinoa seeds with ascorbic acid improves stress tolerance by the increase in enzymatic activities as compared with corresponding salinity levels (El-Sebai *et al.*, 2016).

CONCLUSION:

The findings of this study demonstrate that soil salinity at 2000, 4000, and 8000 ppm NaCl irrigation has a major negative impact on plant growth parameters (plant height, number of tillers/plant, fresh and dry weights of shoot) and plant yield traits (weight of spike, length of spike, number of grains/spike and weight of 100 grains). Furthermore, plants have powerful defensive mechanisms, one of which is antioxidant enzymes that scavenge ROS generation while simultaneously boosting antioxidant activity. Although barley plants are thought to be a cereal crop that tolerates salt, we employed ascorbic acid application to improve plant growth and yield components. Applying ascorbic acid to plants enhances their biomass, defensive mechanisms, and ability to absorb essential nutrients from the soil. By limiting the synthesis of reactive

oxygen species (ROS), ascorbic acid, a non-enzymatic antioxidant molecule, lowers oxidative stress in plants. Furthermore, ascorbic acid treatment enhanced the antioxidant enzymes and yield components of barley plants grown in salinity.

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Table 1: Physical and chemical properties of the soil used.

Particle size distribution (%)				pH	EC	Chemical properties							
Clay	Silt	Sand	Texture			Cations, meq/L				Anions, meq/L			
						K ⁺	Na ⁺	Mg ⁺⁺	Ca ⁺⁺	SO ₄ ⁻	Cl ⁻	HCO ₃ ⁻	CO ₃ ⁻
2.50	18.0	79.5	Loamy clay	8.32	1.64	0.21	8.15	3.5	4.5	5.86	9.5	1.0	-

Table 2: Effect of different concentrations of salinity in irrigation water on some growth parameters of barley plants at different growth stages :

Parameters	Plant height (cm)			Number of tillers/plant			Fresh weight g / plant			Dry weight g / plant		
	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample
Control	34.33	52.60	59.13	5.66	6.60	8.22	2.46	8.85	10.65	0.31	1.81	2.93
Salinity 1	32.67	50.59	55.82	5.00	6.34	8.00	1.75	7.01	8.70	0.24	1.12	2.40
Salinity 2	31.12	48.27	52.70	4.41	6.01	7.30	1.24	4.29	8.31	0.14	0.84	2.15
Salinity 3	28.04	37.00	44.00	4.12	5.00	7.18	0.92	2.88	7.84	0.07	0.57	1.79
LSD	1.38	1.01	1.21	0.43	0.22	0.19	0.31	1.23	1.32	0.04	0.60	0.31

Salinity 1 = 2000 ppm NaCl, Salinity 2= 4000 ppm NaCl, Salinity 3= 8000 ppm NaCl

Table 3: Effect of Ascorbic acid application on some growth parameters of barley plants at different growth stages :

Parameters	Plant height (cm)			Number of tillers/plant			Fresh weight g / plant			Dry weight g / plant		
	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample
Control	35.95	52.11	58.80	5.40	6.41	8.40	2.68	8.71	10.81	0.29	2.25	3.06
AS 1	37.61	53.41	61.70	5.86	6.63	8.90	3.24	9.61	11.52	0.36	2.94	3.62
AS 2	38.78	55.36	63.51	6.33	7.30	9.31	4.31	10.88	12.04	0.43	3.44	4.31
LSD	1.15	1.30	2.76	0.41	0.31	0.42	0.42	0.24	0.60	0.05	0.41	0.25

AS 1 = Ascorbic acid (100 ppm), AS2 = Ascorbic acid (200 ppm)

Table 4: Effect of interaction between salinity concentrations in irrigated water and application of ascorbic acid on some growth parameters of barley plants at different growth stages.

Parameters	Plant height (cm)			Number of tillers/plant			Fresh weight g / plant			Dry weight g / plant		
	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample	1 st sample	2 nd sample	3 rd sample
S1	32.67	50.59	55.82	5.00	6.34	8.00	1.75	7.01	8.70	0.24	1.12	2.40
S1 + AS 1	34.00	53.23	57.71	5.35	6.50	8.36	1.89	7.37	8.84	0.26	1.32	2.75
S1 + AS 2	35.34	55.19	59.18	5.67	7.00	8.58	2.11	7.81	10.12	0.34	2.04	3.21
S2	31.12	48.27	52.70	4.41	6.01	7.30	1.24	4.29	8.31	0.14	0.84	2.15
S2 + AS 1	32.89	50.22	53.24	5.00	6.57	7.66	1.85	5.56	9.09	0.23	1.24	2.54
S2 + AS2	34.23	52.88	55.87	5.30	6.92	8.33	2.13	6.12	11.04	0.31	1.31	3.04
S3	28.04	37.00	44.00	4.12	5.00	7.18	0.92	2.88	7.84	0.07	0.57	1.79
S3 + AS 1	30.21	40.56	48.56	4.66	5.68	7.65	1.12	5.39	10.06	0.15	0.70	2.28
S3 + AS2	32.57	48.21	51.21	5.00	6.01	7.70	1.93	6.04	10.86	0.27	1.15	3.18
LSD	1.30	1.86	1.03	0.20	0.13	0.21	0.09	0.10	0.13	0.01	0.03	0.27

S1 = salinity at 2000 ppm NaCl, S2 = salinity at 4000 ppm NaCl, S3 = salinity at 8000ppm NaCl

S1+AS1= 2000ppm NaCl + 100 ppm ascorbic acid, S1+AS2 = 2000 ppm NaCl + 200 ppm ascorbic acid.

S2+AS1= 4000 ppm NaCl + 100 ppm ascorbic acid, S2+ AS2 = 4000ppm NaCl + 200 ppm ascorbic acid.

S3+AS1 = 8000ppm NaCl + 100 ppm ascorbic acid, S3+AS2 = 8000 ppm NaCl+ 200 ppm ascorbic acid.

Table 5: Effect of salinity concentrations in irrigation water on some yield components of barley plants.

	Weight of spike/ Plant (g)	Length of spike/plant (cm)	Number of grains / spike	Weight of 100 grains (g)
Control	3.92	17.00	34.00	3.40
S 1	3.77	17.63	33.29	3.17
S2	2.38	16.30	31.00	2.84
S3	2.14	14.90	26.48	2.57
LSD	0.15	0.61	0.71	0.18

S1 = salinity at 2000 ppm NaCl, S2 =Salinity at 4000 ppm NaCl, S3 = Salinity at 8000 ppm NaCl.

Table 6: Effect of ascorbic acid application on some yield components of barley plants.

	Weight of spike (g)	Length spike/plant (cm)	Number of grains / spike	Weight of 100 grains
Control	3.78	16.87	35.70	3.87
AS 1	4.05	17.46	36.88	4.46
AS2	4.68	18.00	38.61	5.11
LSD	0.10	0.33	0.48	0.31

AS1 = Ascorbic acid at (100 ppm), AS2 = Ascorbic acid at (200 ppm)

Table 7: Effect of interaction between salinity concentrations in irrigation water and ascorbic acid application on some yield components of barley plants .

	Weight of spike (g)	Length of spike/plant(cm)	Number of grains / spike	Weight of 100 grains (g)
S1	3.77	17.63	33.29	3.17
S1 + AS1	3.80	17.96	35.70	3.46
S1 + AS2	4.13	18.30	38.51	4.14
S2	2.38	16.30	31.00	2.84
S2 + AS1	2.57	16.89	33.40	3.22
S3 + AS2	2.89	17.30	36.00	3.86
S3	2.14	14.90	26.48	2.57
S3 + AS1	2.48	15.52	28.71	2.88
S3 + AS2	2.96	16.30	30.52	3.64
LSD	0.01	0.21	1.40	0.20

S1+AS1=Salinity 2000 ppm NaCl + Ascorbic acid 100 ppm, S1+AS2=Salinity 2000 ppm NaCl +Ascorbic acid 200 ppm.

S2+AS1=Salinity 4000 ppm NaCl+ Ascorbic acid100 ppm, S2+AS2=Salinity 4000 ppm NaCl +Ascorbic acid 200 ppm.

S3+AS1=Salinity 8000 ppm NaCl+ Ascorbic acid 100 ppm, S3+AS2=Salinity 8000 ppm NaCl +Ascorbic acid 200 ppm.

Table 8: Effect of different concentrations of salinity in irrigation water on some antioxidant enzymes activity in leaves of barley plants at two growth stages (ug/g fresh weight)

	Peroxidase		Polyphenol oxidase		Superoxide dismutase		catalase	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	0.17	2.04	0.23	1.53	1.43	1.86	0.14	0.78
S1	0.21	2.40	0.30	1.99	1.81	2.82	0.18	0.86
S2	0.23	2.59	0.32	2.41	1.86	3.29	0.19	0.89
S3	0.31	3.29	0.44	2.91	2.64	4.60	0.26	0.95
LSD	0.01	0.19	0.01	0.37	0.10	0.27	0.01	0.02

S1= Salinity at 2000 ppm NaCl, S2 Salinity at 4000 ppm NaCl, S3=Salinity at 8000 ppm NaCl.

Table 9: Effect of ascorbic acid application on some antioxidant enzyme activities in leaves of barley plants at two growth stages (ug/g fresh weight).

	Peroxidase		Polyphenol oxidase		Superoxide dismutase		Catalase	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
Control	0.16	2.24	0.23	1.46	1.75	2.12	0.16	0.81
AS 1	0.18	2.60	0.27	1.84	2.04	2.60	1.05	1.13
AS 2	0.23	2.86	0.32	2.03	2.65	2.87	1.42	1.84
LSD	0.01	0.19	0.02	1.40	0.09	0.31	0.05	0.13

AS1= Ascorbic acid at 100 ppm. AS2 =Ascorbic acid at 200 ppm.

Table 10: Effect of interaction between salinity concentrations and ascorbic acid on some antioxidant enzymes activity in leaves of barley plants at two growth stages (ug/g fresh wt.)

	Peroxidase		polyphenol oxidase		Superoxide dismutase		catalase	
	1 st	2 nd	1 st	2 nd	1 st	2 nd	1 st	2 nd
S1	0.21	2.40	0.30	1.99	1.81	2.82	0.18	0.86
S1 + AS 1	0.28	2.63	0.36	2.36	1.98	3.09	0.20	1.01
S1 + AS2	0.31	2.84	0.39	2.65	2.34	3.31	0.22	1.35
S2	0.23	2.59	0.32	2.41	1.86	3.29	0.19	0.89
S2 + AS1	0.27	2.74	0.38	2.76	1.93	3.48	0.22	1.05
S2 + AS 2	0.29	2.93	0.41	3.02	2.38	3.73	0.27	1.39
S3	0.31	3.29	0.44	2.91	2.64	4.60	0.26	0.95
S3 + AS1	0.36	3.57	0.48	3.15	2.81	4.91	0.28	1.23
S3 + AS2	0.40	3.95	0.51	3.56	3.15	5.11	0.35	1.64
LSD	0.01	0.19	0.03	0.22	0.15	0.26	0.02	0.09

S1+As1=salinity 2000 ppm NaCl+ Ascorbic acid 100 ppm, S1+As2=salinity 2000 ppm NaCl+Ascorbicacid200 ppm.

S2+As1=salinity 4000 ppm NaCl+ Ascorbic acid 100 ppm, S2+As2=salinity 4000 ppm NaCl+ Ascorbic acid 200 ppm.

S3+As1=salinity 8000 ppm NaCl+ Ascorbic acid 100 ppm, S2+As2=salinity 8000 ppm NaCl+ Ascorbic acid 200 ppm

تأثير معاملات الملوحة وحامض الاسكوربيك في النمو والحاصل ونشاط الانزيمات المضادة للاكسدة لنباتات الشعير

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الملخص العربي :

أجريت تجربة أصص في كلية العلوم جامعة الأزهر فرع النبات في الموسم الزراعي 2020 بهدف دراسة إستجابة نباتات الشعير لتركيزات مختلفة من الملوحة (صفر، 2000، 4000، 8000 جزء من المليون) من كلوريد الصوديوم وكذلك تطبيق حمض الأسكوربيك (100، 200 جزء من المليون) رشاً على النباتات المنزرعة ثلاث مرات أثناء فترة النمو سواء النباتات النامية في الظروف الطبيعية أو النامية تحت ظروف الملوحة بغرض التغلب على التأثير الضار للملوحة وأخذت ثلاث عينات أثناء فترة النمو عند عمر 45، 65، 90 يوم من الإنبات لدراسة قياسات النمو (طول النبات - عدد الفروع - الوزن الغض و الوزن الجاف) وفي نهاية عمر النبات أخذ المحصول عند عمر 130 يوم لدراسة وزن كل سنبل - طول السنبل - عدد الحبوب في السنبل و وزن 100 حبة) كما أجريت تحاليل لبعض الأنزيمات المضادة للاكسدة (إنزيم البروكسيداز - إنزيم البوليفينول اوكسيداز - إنزيم السوبراوكسيدديسموتاز - إنزيم الكاتالاز) عند عمر 45 و 65 يوماً من الإنبات. أدت تركيزات الملوحة إلى حدوث نقص معنوي في قياسات النمو الخضري والمحصول وبعض الأنزيمات المضادة للاكسدة ويزداد هذا النقص بزيادة تركيز الملوحة في التربة. أشارت النتائج إلى وجود زيادة معنوية واضحة في قياسات النمو والمحصول ونشاط بعض الأنزيمات المضادة للاكسدة لنبات الشعير عند تطبيق حمض الأسكوربيك خاصة عند التركيز المرتفع. أظهرت نتائج التفاعل المشترك بين تركيزات الملوحة وتطبيق حمض الأسكوربيك وجود زيادة معنوية واضحة في كل القياسات مقارنة بالنباتات المعاملة بالملوحة فقط. وكان التركيز العالي من حمض الأسكوربيك أكثر فاعلية في تسجيل أعلى القيم للقياسات المختلفة. أثبتت نتائج الدراسة مدى فاعلية استخدام حمض الأسكوربيك في تقليل الأثار الضارة الناجمة من الإجهاد الملحي على قياسات النمو الخضري والمحصول ونشاط بعض الأنزيمات المضادة للاكسدة. وهذا يدل على تحسين إنتاجية نبات الشعير.

الكلمات الاسترشادية: