Morphological and biochemical responses to application of ascorbic acid on some lupine (*Lupinus termis* L.) cultivars under salinity stress

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ABSTRACT

Salinity is abiotic stress and is an effective factor in the production of all crops, especially in semi-arid and arid areas. Ascorbic acid (ASA) is an important antioxidant in plant tissues and has a great role in tolerance to various stresses. The aim of this work is to study the effects of ascorbic acid on growth traits, leaf photosynthetic pigments, biochemical attributes such as total phenolic content, isozymes electrophoresis and protein electrophoresis of three lupine cultivars under salinity stress. The effects of ascorbic acid on germination percentage and different growth parameters, as well as on chlorophyll pigments, total phenolics, electrophoresis of protein and isozymes profiles of esterase (EST), peroxidase (POX) and catalase (CAT) in three cultivars of Egyptian lupine (Giza 1, Giza 2 and Giza 3), which were grown under salinity stress using concentrations of NaCl (0 and 50 mM), were investigated. The results showed a significant decrease in germination and all growth parameters, as well as chlorophyll pigments (Chl a, Chl b) and total phenolics (TP). Also, salinity stress showed noticeable changes in isozymes profiles and an increase in the total number of protein bands in all cultivars. Application of ascorbic acid (ASA) caused an increase in shoot length (SL), root length (RL), seedling fresh weight (SFW) and seedling dry weight (SDW) in all cultivars compared to the salinity without ASA. Also, pretreatment with ASA led to increase of Chl a, Chl b and TP as well as led to the induction of new isozymes bands of EST, POX and CAT that may be related to the tolerance of lupine cultivars to salinity stress. On the other hand, ASA stimulated the appearance of new protein bands and the disappearance of others with different molecular weights under salinity in all cultivars. It could be concluded that soaking lupine seeds in ascorbic acid at a concentration of 200 mg/L before being exposed to salinity reduced the adverse effects of salinity stress by improving morphological and biochemical characteristics.

Keywords: Lupine; Salinity stress; Ascorbic acid; Phenolics; Isozymes; Protein electrophoresis.

INTRODUCTION

Lupine *(Lupinus termis* L.) is one of the oldest agricultural legume crops in Egypt. Since ancient times, lupine seeds are cultivated in Egypt and have a significant role in nutrition due to their high protein content (35-45%) and oil content (10-15%). Lupine seeds are used as a snack for human consumption and they are also of great medical importance in Egypt and all the countries of the world for thousands of years (Kattab, 1986; Rady *et al*., 2016). Due to population growth and high human consumption, there is a need to increase production through expansion reclaimed lands. Increasing crops production is affected by different environmental stresses such as salinity, chilling, drought and heat (Almansouri *et al*., 2001). The germination period is the most important and sensitive stage in the plant life cycle to environmental stresses (Cook, 1997). Salinity is abiotic stress and is an effective factor in the production of all crops, especially in semi-arid and arid areas (Khajeh *et al*., 2003). In general, leguminous crops are sensitive to salinity (Ashraf and Waheed, 1990). The morphological and biochemical processes of plants are negatively affected under salinity stress (Nazar *et al*., 2011 and Benzarti *et al*., 2014). Reduction in growth parameters, photosynthetic pigments and increasing in total protein contents of lupine (*Lupinus termis* L.) plants under salinity stress were recorded by Akladious and Hanafy (2018). The accumulation of salts in the medium of growth leads to the formation of reactive oxygen species that cause significant damage to the chloroplasts and mitochondria. Plants use enzymatic antioxidant (such as catalase and peroxidase) and non-enzymatic antioxidant (such as phenolic compound and ascorbic acid) to protect cells against the harmful effects of free radicals (Mittler, 2002). On the other hand, salinity stress led to changes in the expression

gene of the genetic information inclusive protein and isozymes profiles in lupine plants. These changes caused the appearance of new proteins and the disappearance of others (Akladious and Hanafy, 2018). Ascorbic acid (ASA) is an important antioxidant in plant tissues and has a great role in tolerance to various stresses. It is also the most important metabolite that interacts with HO, H_2O_2, O_3 and lipid hydroxyl peroxidase (Conklin and Barth, 2004). Ascorbic acid is associated with different types of biological activities in the plant. It acts as an enzyme co-factor, a donor and an acceptor in electron transport. Soaking seeds in ascorbic acid before exposure to salinity counteracted the adverse effects of salinity stress on seed germination and seedling growth as well as on some metabolic mechanisms of *Lupinus termis* L. and *Brassica rapa* L. plants (Shaddad *et al*., 1990; Mittal *et al*., 2018).

Therefore, the aim of this work is to study the effects of ascorbic acid on growth traits, leaf photosynthetic pigments, biochemical attributes such as total phenolic content, isozymes electrophoresis and protein electrophoresis of three lupine cultivars under salinity stress.

MATERIAL AND METHODS

Plant materials and growth conditions

Lupine seeds (*Lupinus termis* L.) cultivars (Giza 1, Giza 2 and Giza 3) were obtained from the Legume Crops Research Department of the Agriculture Research Center, Giza, Egypt. This study was conducted in the Laboratories of Seed Technology Research Dept., Field Crops Research Institute, Agricultural Research Center (ARC), and Biochemistry department, Faculty of Agriculture, Al-Azhar University. The seeds were sterilized by soaking in 1% sodium hypochlorite solution for 10 minutes to prevent fungal infections and then washing several times with distilled water to remove any sodium hypochlorite residue. After washing by distilled water, the seeds were soaked in ascorbic acid (ASA) concentrations (0 and 200 mg/L) for 24 h, then dried at room temperature (25 °C). Seeds were transferred into Petri dishes containing two layers of Whatman's filter paper (three replicates) and moistened with 10 mL of distilled water or saline solution of NaCl concentrations (0 and 50 mM), then these dishes were moved to a growth chamber at 20 °C for 21 days. Seed germination was observed daily to maintain moisture levels in Petri dishes. Germination percentage was calculated, according to (Krishnasamy and Seshu, 1990). Growth parameters (shoot and root lengths), (seedling fresh weight and seedling dry weight) were measured according to (Krishnasamy and Seshu, 1990; ISTA, 1999). The calculation of seedling vigor index was done using the formula (seedling length x germination percentage), according to (ISTA, 1999).

Biochemical analysis:

Chlorophyll a and chlorophyll b of seedling leaves were measured using the spectrophotometric method and were calculated according to Lichtentaler and Wellburn (1985). Total phenolics were assayed according to Noreen and Ashraf (2009).

Isozymes of esterase, peroxidase and catalase were separated via 8% native polyacrylamide gel electrophoresis (Native-PAGE) using the procedures outlined by Wendel and Weeden (1989). Isozymes fractionation was performed on vertical slab (19.8cm x26.8cm x.02cm) using the gel laconic electrophoresis apparatus according to Jonathan and Wendel (1990).

Protein extracts of seedling leaves of three lupine cultivars were separated by Sodium Dodecyl Sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) according to Laemmli (1970). The protein gel was scanned, and analysis was carried out by image analysis software.

Statistical analysis was performed using randomized complete block design (three replicates) and the differences between means were calculated by L.S.D test according to Steel and Torrie (1980).

RESULTS AND DISCUSSION

Ascorbic acid and growth parameters

The data in Table 1 showed the effect of ascorbic acid (ASA) on seed germination and growth characteristics of three cultivars of lupine under salinity during the germination stage (21 days). The results showed that seed germination and growth parameters in lupine cultivars significantly decreased at 50 mM NaCl compared to the control. Under normal conditions, germination percentage increased in Giza 1 and Giza 2 with application of ASA (200 mg/L) from 90% and 95% to 100% and 100%, respectively. Under salinity stress, maximum reduction in germination percentage was observed in Giza 2 (75%), while Giza 3 was recorded as salt-tolerant (100%) with ASA application. In addition, the application of ASA led to increasing shoot length

(SL) by 52.33%, 55.69% and 24.45, root length (RL) by 70.51%, 110.68% and 9.15, seedling fresh weight (SFW) by 16.44%, 21.53% and 8.64%, seedling dry weight (SDW) by 14.28%, 18.18%, 9.37% in Giza 1, Giza 2 and Giza 3 cultivars, respectively compared to the salinity without application of ASA. Accumulation of salts and ions in the growth environment led to osmotic and drought stress, causing the difficulty of absorbing water by plant tissue, which led to a reduction in germination percentage and all different growth parameters in lupine cultivars. ASA plays a major role in increasing cell division and enlargement in treated plants. Also, it scavenges the reactive oxygen species (ROS), which cause severe damage to the plant and help in many different biological processes (El-Kobisy *et al*., 2005). Several studies have confirmed the role of ASA in improving plant tolerance to salinity stress (Athar *et al*., 2008). These results are consistent with EL-Afry *et al*. (2018) who found that using ASA reduces the adverse effects of salinity stress on flax (*Linum usitatissimum* L.).

Table 1. Effect of ascorbic acid on germination percentage and growth parameters in seedling of three lupine cultivars under salinity stress.

Cultivars	Treatments	GP%	SL (cm)	RL (cm)	SRL (cm)	SFW (g)	SDW (g)	SVI
	0+0 ASA	90	12.50	9.01	21.51	3.05	0.29	1935.9
Giza 1	0+200 ASA	100	12.25	10.90	23.15	2.95	0.27	2315
	S+0 ASA	80	7.51	6.75	14.26	2.25	0.21	1140.8
	S+200 ASA	85	11.44	11.51	22.95	2.62	0.24	1950.75
	0+0 ASA	95	13.01	12.40	25.41	3.00	0.25	2413.95
Giza 2	0+200 ASA	100	14.10	14.44	28.54	2.92	0.25	2854
	S+0 ASA	75	6.50	5.80	12.30	2.09	0.22	922.5
	S+200 ASA	100	10.12	12.22	22.34	2.54	0.26	2234
	0+0 ASA	100	14.10	13.60	27.70	4.21	0.39	2770
Giza 3	0+200 ASA	100	13.30	13.15	26.45	3.87	0.38	2645
	S+0 ASA	85	10.14	11.80	21.94	3.24	0.32	1864.9
	S+200 ASA	100	12.62	12.88	25.50	3.52	0.35	2550
LSD 5%		1.90	0.48	0.59	1.02	0.09	0.007	110

S= 50 mM salinity by NaCl, 0 and 200 ASA= 0 and 200 mg/L ascorbic acid, GP= Germination Percentage; SL= Shoot Length; RL=Root Length; SRL=Shoot length + Root Length; SFW=Seedling Fresh Weight, SDW= Seedling Dry Weight; SVI, Seedling Vigor Index.

Ascorbic acid and total chlorophyll

Data presented in table (2) indicated a significant decrease in chlorophyll a (Chl a) and chlorophyll b (Chl b) at 50 mM NaCl salinity, thus a decrease in total chlorophyll in all cultivars of lupine. Giza 3 achieved the highest values of Chl a (3.45 mg/g) and Chl b (2.35 mg/g) with application of ASA under control, while Giza 2 has the lowest values of Chl a (1.43 mg/g) and Chl b (1.04 mg/g) under salinity without ASA application. It is noted that pretreatment of seeds with ASA led to significant increases of Chl a and Chl b under control and salinity stress compared to control and salinity without ASA. For example, Chl a and Chl b increased from 1.43 mg/g and 1.04 mg/g to 1.98 mg/g and 1.45 mg/g, respectively with ASA under salinity in Giza 2. Decreased seedling pigments (Chl a and Chl b) with salinity stress by NaCl may be due to reduction in photosynthesis (Zhang *et al*., 2003). Also, NaCl causes the increased activity of the chlorophyllase enzyme which in its turn breaks down the chlorophyll by removing the phytol group (Santos, 2004). ASA has a great role in counteracting the negative effects of salinity stress by protecting photosynthetic apparatus and photosynthetic pigments from oxidation by increasing enzymatic antioxidants activity to provide protection against ROS (Hamada, 1998 and Sairam *et al*., 2005). These results are in line with the results of EL-Afry *et al*. (2018) who revealed that application of ASA at different concentrations (200 and 400 mg/L) exhibited increases in photosynthetic pigments under salinity in flax.

Ascorbic acid and total phenolics

Data in table (2) showed that total phenolics contents (TP) in leaves of seedling lupine cultivars were significantly decreased under salinity at 50 mM NaCl as compared to control. Under salinity stress without ASA, Giza 3 has the highest values (40.20 mg/g) as compared with

other cultivars. Pretreatment with ASA enhanced TP in all leaves of seedling lupine cultivars under control and salinity. The highest value of TP (62.86 mg/g) was found in Giza 3 under control with ASA followed by (52.50 mg/g) in Giza 1 under salinity with the application of ASA. Plants differ in their phenolics contents, that are related to environmental and genetic factors (Awika and Rooney, 2004). Our results showed that decreasing of TP under the influence of salinity in leaves of seedling lupine cultivars may be due to enzymatic activity disturbance and reduction in photosynthesis (Wong *et al*., 2006). ASA has a positive effect on photosynthesis and the synthesis of carbohydrates, which are the main substrates for the biosynthesis of phenolics and flavonoids (Shazly *et al*., 2013). Increasing of TP with the application of ASA prevented oxidative stress of plant cells caused by salinity and thus protected plants from damaging lipids, proteins, RNA and DNA (Apel and Hirt, 2004). These results agree with El-khamissi *et al*. (2018) and Allahveran *et al*. (2018).

Table 2. Effect of ascorbic acid on chlorophyll pigments and total phenolics in seedling leaves of lupine cultivars under salinity stress.

Cultivars	Treatments	Photosynthetic pigments (mg/g fresh wt.)				TP (mg/g F.wt.)
		Chl a (mg/g F.wt.)	Chl b (mg/g F.wt.)	Chl (a+b)	Chl (a/b)	
	0+0 ASA	2.65	1.82	4.47	1.45	39.42
Giza 1	0+200 ASA	3.26	2.14	5.40	1.52	44.27
	S+0 ASA	1.93	1.42	3.35	1.35	37.60
	S+200 ASA	2.67	1.76	4.43	1.51	52.50
	0+0 ASA	2.75	1.48	4.23	1.85	42.18
Giza 2	0+200 ASA	2.83	1.99	4.82	1.42	46.66
	S+0 ASA	1.43	1.04	2.47	1.37	35.52
	S+200 ASA	1.98	1.45	3.43	1.36	47.81
	0+0 ASA	3.25	2.13	5.38	1.52	44.47
Giza 3	0+200 ASA	3.45	2.35	5.80	1.46	62.86
	S+0 ASA	2.15	1.55	3.70	1.38	40.20
	S+200 ASA	2.40	1.70	4.10	1.41	48.09
LSD		0.05	0.045	0.085	0.01	1.58

S= 50 mM salinity by NaCl, 0 and 200 ASA= 0 and 200 mg/L ascorbic acid, Chl a= Chlorophyll a, Chl b= Chlorophyll b, TP= Total Phenolics

Isozymes expression

Isozyme expression of Esterase (EST), Peroxidase (POX) and Catalase (CAT) in seedling leaves of the three lupine cultivars treated or non-treated with salinity and ascorbic acid are represented in (Tables 3, 4 and 5). EST isozyme analysis showed five bands at Rf 0.18, 0.35, 0.47, 0.63 and 0.85. The band at Rf 0.63 appeared in the three cultivars under all treatments. That means it was a distinctive band of lupine cultivars. EST isozyme band at Rf 0.47 was found in Giza 1 in all treatments, while it was missing in Giza 2 and Giza 3 cultivars which could mean that band distinguished Giza 1 cultivar. However, salinity stress caused the induction of new EST isozyme bands at Rf 0.18 and 0.35 whether in the presence or absence of ASA in all lupine cultivars as compared to control. Application of ASA led to the appearance of new EST isozyme band at Rf 0.85 in Giza 1 and Giza 3 under control and salinity, respectively (Table 3). On the other hand, POX isozyme band at Rf 0.27 was noticed in all cultivars of lupine with different treatments. Pretreatment with ASA also caused an increase of total POX isozyme bands in all cultivars under salinity stress. The band with Rf 0.52 was presented with the application of ascorbic acid under salinity as compared to salinity without ascorbic acid in Giza 2. Also, using ASA led to the induction of new POX isozyme bands at Rf 0.76 under salinity in Giza 1 and Giza 3, while this band disappeared in Giza 2 in all treatments (Table 4). CAT isozyme analysis displayed six bands at Rf 0.21, 0.32, 0.50, 0.65, 0.81 and 0.91. Giza 3 had the highest CAT isozyme bands (5 bands) under salinity with ASA. CAT isozyme band with Rf 0.32 disappeared under salinity in all cultivars and appeared again with ASA only in Giza 3 cultivar. The band at Rf 0.65 is found in all lupine cultivars in all treatments. Giza 1 had a specific band at Rf 0.81 with ASA while it was absent in other cultivars. Also, Application of ASA caused the appearance of a new band at Rf

0.91 in Giza 2 and Giza 3 under salinity by NaCl, while this band was not present in Giza 1 (Table 5). Our results showed that the appearance of isozyme bands and disappearance of others under the influence of salinity by NaCl in all cultivars of lupine. The disappearance of some bands may be due to the suppression of the genes responsible for the synthesis of proteins by salinity stress (Akladious and Abbas, 2014). The application of ascorbic acid led to the induction of new bands that may be related to the tolerance of cultivars to salinity or may help the cultivar in mitigating the harmful effects of salinity stress by its effective role against the oxidative stress resulting from salinity stress (Mohamed, 2005). (Gao *et al*., 2008) revealed that *Jatropha curcas* seedlings contain high peroxidase isozyme activity in salt tolerance cultivars as compared to sensitive cultivar. The results are consistent with Behairy *et al*. (2012) who found that changes in peroxidase and esterase isozymes under salinity with the application of ASA reduced the harmful effects on fenugreek plants under salinity stress.

Table 3. Effect of ascorbic acid on expression of esterase isozyme in seedling leaves of lupine cultivars under salinity stress.

Cultivars	Giza 1				Giza 2				Giza 3			
Rf of visible bands	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA
0.18	-	-	+	+	-	-	+	+	-	+	+	+
0.35	-	-	+	+	-	-	+	+	-	-	+	+
0.47	+	+	+	+	-	-	-	-	-	-	-	-
0.63	+	+	+	+	+	+	+	+	+	+	+	+
0.85	-	+	-	-	-	-	-	-	-	-	-	+

S= 50 mM salinity by NaCl, 0 and 200 ASA= 0 and 200 mg/L Ascorbic acid.
Rf = retention factor, band present = +, band absent = -

Table 4. Effect of ascorbic acid on expression of peroxidase isozyme in seedling leaves of lupine cultivars under salinity stress.

Cultivars	Giza 1				Giza 2				Giza 3			
Rf of visible bands	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA
0.27	+	+	+	+	+	+	+	+	+	+	+	+
0.33	-	+	+	+	-	+	+	+	-	+	+	+
0.52	-	-	+	+	-	-	-	+	-	+	+	+
0.76	-	-	-	+	-	-	-	-	-	-	-	+

Table 5. Effect of ascorbic acid on expression of catalase isozyme in seedling leaves of lupine cultivars under salinity stress

Cultivars	Giza 1				Giza 2				Giza 3			
Rf of visible bands	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA
0.21	+	+	-	+	+	+	-	+	+	+	+	+
0.32	+	+	-	-	+	-	-	-	+	-	-	+
0.50	-	-	-	-	+	+	_	+	-	-	-	+
0.65	+	+	+	+	+	+	+	+	+	+	+	+
0.81	-	+	-	+	-	-	-	-	-	-	-	-
0.91	-	-	-	-	-	-	-	+	-	-	-	+

S= 50 mM salinity by NaCl, 0 and 200 ASA= 0 and 200 mg/L Ascorbic acid.
Rf = retention factor, band present = +, band absent = -

Protein electrophoretic pattern:

Sixteen protein bands with molecular weights ranging between 195.35 and 12.77 KDa were found in seedling leaves of lupine cultivars (Table 6). There are seven common bands that were observed in all lupine cultivars whether under control or salinity stress, with or without ASA or without ASA, which have molecular weights 181.64, 132.85, 71.13, 45.85, 38.15, 20.35 and 12.77 KDa. Salinity stress at 50 mM NaCl led to increasing the total number of protein bands in all cultivars. The application of ascorbic acid under salinity resulted in the induction of one new protein band with a molecular weight of 170.82 KDa in all cultivars, which was not present in treatments without ASA. Giza 1 had a specific protein band with molecular weight 177.45 KDa under control. Salinity stress stimulated the synthesis of three new protein bands at molecular weights 155.45, 140.33 and 110.48 KDa and the disappearance of one protein band with molecular weight 177.45 KDa as compared to control in Giza 1 cultivar. Also, two protein bands with molecular weights 155.45 and 60.57 KDa disappeared under salinity with ASA as compared to salinity without ASA. On the other hand, pretreatment with ASA led to the disappearance of one protein band with molecular weight 60.57 KDa in Giza 2 under salinity as compared to salinity without ASA. The results also showed the appearance of three protein bands at molecular weights 195.35, 155.45 and 85.64 KDa at salinity in Giza 3 as compared to control. Moreover, pretreatment with ascorbic acid stimulated the production of one new protein band at molecular weight 110.48 KDa, and the disappearance of two protein bands with molecular weights 155.45 and 85.64 KDa under salinity as compared to salinity without ASA.

The appearance of one protein band with a molecular weight of 170.82 KDa in all lupine cultivars under salinity stress with the application of ASA may be related to increasing tolerance of these cultivars to salinity stress. The most remarkable response of plants to salinity stress is the activation of many genes involved in respective salinity stress response (Rasul *et al*., 2017). Lu *et al*. (2015) found that salinity stress promotes changes in gene expression to adjust to adverse conditions. These results suggest that these proteins have specific functions to help different lupine cultivars in mitigating the harmful effects of salinity stress. According to the results of protein bands, using ascorbic acid helps new proteins and increases the accumulation of proteins that are involved in increasing the tolerance to salinity stress (Azooz and Al-Fredan, 2009). These results are in accordance with those of Bassuony *et al*. (2008) and Beltagi (2008).

Table 6. Effect of ascorbic acid on protein patterns separated by SDS-PAGE in seedling leaves of lupine cultivars under salinity stress.

Cultivars		Giza 1				Giza 2				Giza 3			
No. of bands	MW	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA	0+0 ASA	0+200 ASA	S+0 ASA	S+200 ASA
1	**195.35**	**-**	**+**	**+**	**+**	**-**	**+**	**+**	**+**	**-**	**+**	**+**	**+**
2	**181.64**	**+**	**+**	**+**	**+**	**+**	**+**	**+**	**+**	**+**	**+**	**+**	**+**
3	**177.45**	**+**	-	-	-	-	-	-	-	-	-	-	-
4	**170.82**	-	-	-	+	-	-	-	+	-	-	-	+
5	**155.45**	-	-	+	-	-	-	-	-	-	-	+	-
6	**140.33**	**-**	**-**	**+**	**+**	-	**-**	-	**-**	**+**	**+**	**+**	+
7	**132.85**	+	+	+	+	+	+	+	+	**+**	+	+	+
8	**110.48**	-	-	+	+	-	-	-	-	-	-	-	+
9	**92.47**	+	+	**+**	+	-	-	-	-	+	+	+	+
10	**85.64**	-	-	**-**	-	-	-	-	-	-	-	+	-
11	**71.13**	+	**+**	+	+	+	**+**	+	+	+	+	+	+
12	**60.57**	+	**+**	+	-	+	**+**	+	-	+	+	+	+
13	**45.85**	+	**+**	+	+	+	**+**	+	+	+	+	+	+
14	**38.15**	+	**+**	+	+	+	**+**	+	+	+	+	+	+
15	**20.35**	+	**+**	+	+	+	**+**	+	+	+	+	+	+
16	**12.77**	+	**+**	+	+	+	**+**	+	+	+	+	+	+
Total number of bands		10	10	13	12	8	9	9	9	10	11	13	13

band present = +, band absent = -, MW = Molecular Weight.

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الاستجابات المورفولوجية والبيوكيميائية لتطبيق حمض الأسكوربيك على بعض أصناف الترمس تحت الاجهاد الملحي

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الملخص العربي

تعد الملوحة إجهاد حيوي وعامل مؤثر على إنتاج جميع المحاصيل، وخاصة في المناطق الجافة وشبه الجافة ويعتبر حمض الأسكوربيك مضاد أكسدة مهم في أنسجة النبات ويلعب دوراً كبيراً في تحمل الإجهادات المختلفة. يهدف هذا البحث إلى دراسة تأثيرات حمض الأسكوربيك على خصائص النمو وأصباغ التمثيل الضوئي في الأوراق والخصائص البيوكيميائية مثل الفينولات الكلية والتفريد الكهربي للمشابهات الأنزيمية والبروتين في ثلاثة أصناف من الترمس تحت الإجهاد الملحي. تمت دراسة تأثيرات حمض الأسكوربيك على نسبة الأنبات ومقاييس النم والمختلفة، وكذلك على أصباغ الكلوروفيل، والفينولات الكلية، والمشابهات الأنزيمية للإيستريز والبيروكسيديز والكتاليز، والتفريد الكهربي للبروتين في ثلاثة أصناف من الترمس المصري (جيزة 1 وجيزة 2 وجيزة 3) التي تنمو تحت الإجهاد الملحي بتركيز 50 ملمول من كلوريد الصوديوم. أظهرت النتائج إنخفاضا كبيرًا في جميع مقاييس النمو، وكذلك أصباغ الكلوروفيل (الكلوروفيل أ والكلوروفيل ب) والفينولات الكلية. أيضاً، أدى الإجهاد الملحي إلى تغييرات ملحوظة في حزم المشابهات الأنزيمية وزيادة العدد الإجمالي للحزم البروتينية في جميع الأصناف. تطبيق حمض السكوربيك تسبب في زيادة طول الريشة وطول الجذر والوزن الطازج للبادرات والوزن الجاف للبادرات في جميع الأصناف مقارنة بالتعرض للملوحة دون تطبيق حمض الأسكوربيك. أيضاً أدت المعالجة المسبقة بالأسكوربيك إلي زيادة كلوروفيل أ وكلوروفيل ب والفينولات الكلية وكذلك أدت إلى ظهور حزم جديدة من المشابهات الأنزيمية للايستريز والبيروكسيديز والكتاليز والتي قد تكون مرتبطة بزيادة تحمل أصناف الترمس للإجهاد الملحي. من ناحية آخري، حفز إستخدام حمض الأسكوربيك ظهور حزم بروتينية جديدة واختفت آخري عند أوزأن جزيئية مختلفة تحت الإجهاد الملحي في جميع الأصناف. أخيرًا، يمكن استنتاج أن نقع البذور في حمض الإسكوربيك بتركيز 200 مليجرام / لتر قبل تعرضها للملوحة قلل من التأثيرات الضارة للإجهاد الملحي على أصناف الترمس عن طريق تحسين الصفات المورفولوجية والبيوكيميائية.

الكلمات المفتاحية: ترمس، إجهاد الملوحة، حمض الإسكوربيك، الفينولات، ايزوزيم، التفريد الكهربائي للبروتين.