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ALLEVIATION OF SEA WATER STRESS ON TOMATO PLANTS BY FOLIAR APPLICATION OF ASPARTIC ACID AND GLUTATHIONE

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Abstract

Tomato plants (*Lycopersicon esculentum* Mill.) treated with 8 and 16% of sea water reduced growth parameters and chemical constituents. Both aspartic acid and glutathione increased plant growth, anthocyanin, α-tocopherol, ascorbic acid and enzymatic activities. Increased endogenous amino acids led to positive changes in protein electrophoresis and caused obvious changes in anatomical features of the stems. The effect of aspartic acid was superior to that of glutathione in increasing plant growth. Under low saline conditions, maximum plant growth was obtained from plants treated with aspartic acid and 8% of sea water, followed by 4%. Data revealed that antioxidants could partially alleviate the harmful effects of salinity.

Introduction

Soil salinity is an abiotic stress and limiting factor for plant production. Increasing NaCl concentration in nutrient solution adversely affected tomato shoot and plant height, K concentration and K/Na ratio (Garcia *et al*. 2007). Salt stress causes partial swelling of endoplasmic reticulum, decrease in mitochondrial cristae and swelling of mitochondria, fragmentation of tonoplast and degradation of cytoplasm by the mixture of cytoplasmic and vacuolar matrices in leaves of sweet potato (Mitsuya *et al*. 2000). The important response of salt stress is reduction in the rate of leaf surface expansion leading to cessation of expansion as salt concentration increases. Salt stress causes water deficit as a result of osmotic effects on metabolic activities of plants and this water deficit results in oxidative stress because of the formation of reactive oxygen species. The reactive oxygen species that are by products of hyperosmotic and ionic stresses cause membrane disfunction and cell death. The most common stress responses in plants is overproduction of compatible organic solutes which are low molecular weight, highly soluble compounds that are usually nontoxic at high cellular concentrations. The plants defend against these reactive oxygen species by induction of activities of certain antioxidative enzymes such as catalase, peroxidase, glutathione reductase, and superoxide dismutase, which scavenge reactive oxygen species. Activities of antioxidative enzymes increase under salt stress in wheat (Hernandez *et al*. 2000**)**.

The asparagine and glutamine connect the two important metabolic cycles of the plant, the carbon and nitrogen cycles, and they have an influence both on sugars and proteins. In plants, aspartate is the precursor to several amino acids, including methionine, threonine, isoleucine (Rawia *et al.* 2011). Glutathione (GIT) is non-protein thiol present in plants. The alleviating effect of glutathione might be through scavenging active oxygen species under salt stress.

The aim of present work is to increase the salinity tolerance by treatment with glutathione and aspartic acid in tomato plants grown under higher level of salinity.

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Materials and Methods

Tomato-super strain B- (*Lycopersicom esculantum* Mill., Castle rook cultivar) seeds were obtained from the Vegetables Research Center, El-Dokkey, Giza, Egypt. The seeds were surface sterilized by immersing in 70% ethanol for 2 min followed by 0.2% sodium hypochlorite (NaOCl) for 3 min. They were washed for several times with sterile distilled water, and then grown in large clay pots (40 cm diam) containing loam based garden soil and were divided into two groups. The 1st group of pots was irrigated with tap water to serve as control. The 2nd group was irrigated with tap water until the 3-leaf stage appeared then they were divided into three sets and irrigated with three salinity treatments. The sea water salinity was prepared by diluting sea water brought from the Mediterranean Sea, near Alexandria city (the major ions of sea water were: 487 mM Na^+, 10 mM K^+, 54 mM Mg^{2+}, 586 mM Cl, plus other less concentrated macro- and micro-nutrients) to obtain three concentrations (4, 8 and 16%) using electrical conductivity meter. Plants were watered with equal amounts of the suggested concentrations of the sea water to maintain soil moisture near field capacity. At 5-leaf stage, some pots from each group was sprayed with 20 ml/plant of two different solutions namely aspartic acid in concentration of 100 mg/l and glutathione in concentration of 50 mg/l glutathione. Spraying treatments were carried out two times with two weeks interval for both spraying treatments. Plant samples were harvested 10 weeks after sowing date (70 days old). Leaves, stems and roots were separated for analysis.

Plant height, number of leaves, leaf area, fresh weight and dry matter of shoot and root were determined for each treatment. Anthocyanin content was estimated after Krizek *et al*. (1993). The content of α-tocopherol was measured following Philip *et al.* (1954). Ascorbic acid was determined as described by Mukherjee and Choudhuri (1983). Catalase (CAT), peroxidase (POX) and polyphenol oxidase (PPO) activities were assayed according to Kumar and Khan (1982). Electrophoretic protein profile of tomato leaves was analyzed according to SDS-PAGE technique (Laememli 1970) which relates polypeptide maps, molecular protein markers, percentage of band intensity using gel protein analyzer version 3 (MEDIA- CYBERNE TICE, USA). Acid hydrolysis and HPLC determination of amino acids were carried out after Gehrke *et al.* (1985). Anatomical studies were carried out by using the method of Nassar and El-Sahhar (1998). The sections were examined to detect histological manifestations of noticeable responses resulting from the treatment of the plants.

The results were subjected to one-way ANOVA and the differences between means at the 5% probability level were determined using DMRT.

Results and Discussion

Irrigation of tomato plant with sea water revealed that plant height, number of leaves/plant, root length, leaf area, as well as fresh and dry weights of the different plant parts declined with increasing sea water (SW) concentrations as compared with control (Table 1). However, foliar spraying of plants with aspartic acid or glutathione increased these growth parameters of plants grown under either non saline or saline conditions compared with control. The inhibitory effects of salt stress on these growth parameters were also recorded by other investigators in various plant species (Hassanein *et al*. 2008, Wahba *et al.* 2002)**.**

Salt stress had an inhibitory effect on the ancothyanin, α-tocopherol and ascorbic acid contents in the tomato plants, these contents were significantly decreased in the stressed plants (Table 2). On the other hand, data shows that foliar spraying of the plants with AA and/or GlT significantly increased the contents of anthocyanin, α-tocopherol and ascorbic acid particularly under saline conditions (4 and 8%) as compared with control. Salt stress accelerates the formation of Reactive Oxygen Species (ROS). Plants maintain complex systems of multiple types of

Table 1. Effect of foliar spraying with aspartic acid and/or glutathione on growth parameters of tomato plants irrigated with different concentrations of diluted sea water.

Treatments		Plant height (cm)	Root length (cm)	No. of leaves/ plant	Leaf area (cm^2)	Fresh weight (g)		Dry weight (g)	
						Shoot	Root	Shoot	Root
	Tp (Control)	31.0^{bcd}	20.80^{de}	8.0^{cde}	81.6^{g}	28.68^{ef}	4.31^{g}	2.86^{c}	0.57^{de}
Sea water (SW)	4% SW	33.10^{abc}	21.25^{de}	8.5^{bcde}	88.3^{f}	29.35^{ef}	6.1^{ef}	2.99^{bc}	0.69^{d}
	8% SW	26.45^{de}	18.25^{ef}	7.5^{de}	70.6^{h}	25.22^{g}	4.39^{g}	1.64^{d}	0.4^{ef}
	16% SW	24.50^{e}	16.75^{f}	7.0^{e}	69.4^{h}	19.35^{h}	4.15^{g}	1.64^{d}	0.35^{f}
	AA + Tp	35.20^{ab}	22.3^{bcd}	9.5^{abc}	101.2^{e}	31.05^{de}	7.55^{d}	3.24^{bc}	0.89^{c}
Aspartic acid (AA)	AA+ 4% SW	37.00^{a}	25.00^{b}	10.0^{ab}	135.7^{b}	37^{ab}	10.87^{ab}	3.6^{ab}	1.39^{a}
	AA + 8% SW	37.65^{a}	28.95^{a}	10.5^{a}	142.6^{a}	38.8^{a}	11.33^{a}	3.97^{a}	1.42^{a}
	AA+ 16 % SW	32.55^{cde}	22.00^{de}	8.0^{cde}	105.1^{g}	30.22^{fg}	5.87^{f}	2.96^{d}	0.59^{ef}
Glutathione (GlT)	GlT +Tp	35.00^{ab}	22.00^{cd}	9.0^{abcd}	97.5^{e}	30.91^{e}	6.56^{e}	3.16^{bc}	0.76^{cd}
	GlT + 4% SW	36.00^{a}	24.45^{bc}	9.5^{abc}	119.1^{d}	33.71^{cd}	9.48^{c}	3.31^{abc}	1.11^{b}
	GlT + 8% SW	36.20^{a}	25.00^{b}	10.0^{ab}	130.5^{c}	35.93^{bc}	10.62^{b}	3.47^{abc}	1.26^{ab}
	GlT +16% SW	26.80^{de}	19.0^{efj}	8.0^{cde}	72.4^{h}	25.93^{g}	4.67^{g}	1.77^{d}	0.43^{ef}

Tp = Tap water. Data are reported as means (n = 5). Means were separated by DMRT, different letters indicate a significant difference at $p \leq 0.05$.

Table 2. Effect of foliar spraying with aspartic acid and/or glutathione on anthocyanin, α-tocopherol and ascorbic acid contents of tomato plants irrigated with different concentrations of diluted sea water.

Treatments		Anthocyanin (μM/g FW)	α-tocopherol (μg/g FW)		Ascorbic acid (μg/g FW)	
			Shoot	Root	Shoot	Root
	Tp (Control)	0.18 ± 0.01^{edf}	71.13 ± 0.79^{i}	13.78 ± 0.55^{ef}	13.10 ± 0.48^{de}	3.86 ± 0.25^{ef}
Sea water (SW)	4 % SW	0.21 ± 0.01^{bcd}	79.81 ± 1.12^{h}	14.91 ± 0.71^{def}	14.65 ± 0.56^{cd}	4.00 ± 0.40^{ef}
	8 % SW	0.18 ± 0.01^{def}	65.77 ± 0.80^{j}	13.11 ± 1.05^{ef}	14.00 ± 0.65^{cd}	3.92 ± 0.8^{ef}
	16 % SW	0.14 ± 0.00^{f}	61.13 ± 0.87^{k}	12.70 ± 0.97^{f}	10.17 ± 0.74^{e}	3.24 ± 0.56^{ef}
Aspartic acid (AA)	AA+ Tp	0.22 ± 0.01^{bcd}	105.11 ± 0.96^{e}	16.58 ± 0.42^{bcd}	16.54 ± 0.78^{bc}	5.00 ± 1.0^{bce}
	AA+ 4 % SW	0.29 ± 0.01^{a}	127.9 ± 1.34^{b}	18.70 ± 0.71^{ab}	19.23 ± 1.23^{ab}	6.81 ± 0.39^{ab}
	AA + 8 % SW	0.33 ± 0.01^{a}	138.26 ± 1.78^{a}	19.77 ± 0.84^{a}	22.01 ± 2.0^{a}	7.53 ± 0.53^{a}
	AA+ 16 % SW	0.19 ± 0.03^{cde}	93.05 ± 1.14^{f}	15.52 ± 0.59^{cde}	15.21 ± 1.04^{cd}	4.81 ± 0.6^{cde}
Glutathione (GlT)	GlT + Tp	0.21 ± 0.02^{bcd}	87.54 ± 0.87^{g}	15.30 ± 0.68^{cde}	14.33 ± 0.8^{cd}	4.27 ± 0.09^{def}
	GlT + 4% SW	0.23 ± 0.01^{bc}	111.49 ± 0.61^{d}	17.80 ± 1.56^{abc}	18.78 ± 2.17^{ab}	5.96 ± 0.85^{abc}
	GlT + 8% SW	0.24 ± 0.01^{b}	121.18 ± 0.82^{c}	18.91 ± 0.81^{ab}	19.32 ± 1.10^{ab}	6.22 ± 0.77^{abc}
	GlT +16% SW	0.15 ± 0.02^{ef}	70.03 ± 1.02^{i}	13.78 ± 1.42^{ef}	13.00 ± 0.4^{de}	3.11 ± 0.79^{f}

Tp = Tap water. Data are reported as means (n = 3). Means were separated by DMRT, different letters indicate a significant difference at $p \leq 0.05$.

antioxidants, such as anthocyanin, ascorbic acid (vitamin C) and tocopherol (vitamin E). Non-enzymatic antioxidant activity is represented by a series of antioxidant molecules that the plant uses against active oxygen species formation (Mittler 2002).

The changes in various enzyme activities of shoot and root of tomato plants in response to salinity stress either alone or in combination with aspartic acid and/or glutathione are shown in Figs 1 - 3. Results indicated that peroxidase (POX), catalase (CAT) and polyphenol oxidase (PPO) activities were significantly decreased in shoot and root of tomato plants irrigated with different concentrations of sea water (SW). On the other hand, tomato plants treated with AA and/or GlT exhibited more stress resistance by increasing their activities of CAT, POX and PPO as compared

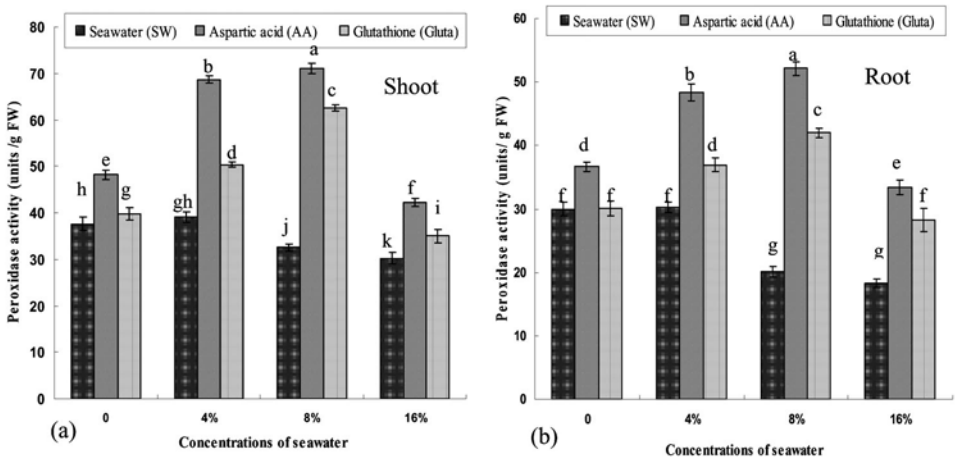

Fig.1a-b: Effect of foliar spraying with aspartic acid and/or glutathione on peroxidase activity of shoot (a) and root (b) of tomato plants grown under saline conditions. Bars represent the means ± Sd.

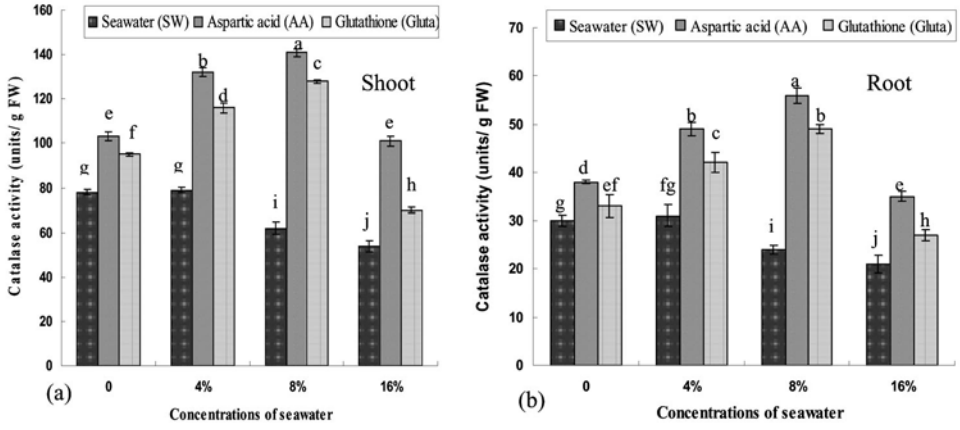

Fig. 2a-b: Effect of foliar spraying with aspartic acid and/or glutathione on catalase activity of shoot (a) and root (b) of tomato plants grown under saline conditions. Bars represent the means ± Sd.

with untreated plants. The higher activities in these enzymes were recorded in plants treated with AA and irrigated with 8% SW. The reduction in these enzymes activities as a result of salt stress demonstrated that these enzymes were unable to completely neutralize H_2O_2 resulted from the oxidative salt stress. Dash and Panda (2001) reported that higher activity of antioxidant enzymes

(CAT, POX and PPO) caused lower H_2O_2 production, lipid peroxidation and higher membrane stability. These enzymes are involved in reelimination of H_2O_2 from stressed cells. Catalase is the most effective antioxidant enzyme preventing oxidative damage. Exogenous application of either AA or GlT significantly increased the specific activity of phenol peroxidase in the stressed plants, which decreased the injurious effect of salt and it reacted with H_2O_2 and maintained the membrane integrity (Sairam *et al.* 2005).

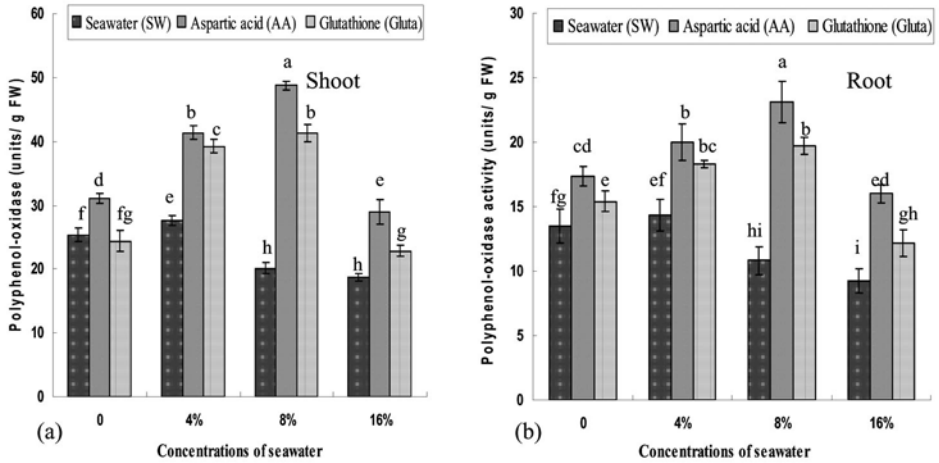

Fig. 3a-b: Effect of foliar spraying with aspartic acid and/or glutathione on polyphenol-oxidase activity of shoot (a) and root (b) of tomato plants grown under saline conditions. Bars represent the means ± Sd.

Different SDS-PAGE patterns of tomato plants irrigated with different levels of salinity (4, 8 and 16%) and foliar spraying with aspartic acid and/or glutathione are presented in Table 3 and Fig. 4. Irrigation of tomato plants with different sea water (8 and 16%) levels showed an increase in the total number of protein bands. In addition, tomato plants irrigated with different levels of salinity and foliar spraying with aspartic acid and/or glutathione exhibited an increasing in the total number of bands. These results indicate that the plant irrigated with different salinity levels plus application of glutathione and aspartic acid characterized by the appearance of new bands as compared with that of the untreated plants. Scanning of the gel showed that the protein band which has the molecular weight (MW) of 164.17 kDa disappeared under saline conditions. At the same time 4 bands with MWs of 54.12, 31.95, 28.50 and 19.28 kDa have developed in plants treated with different levels of salinity alone or with salinity levels plus sprayed by the two used antioxidants but these bands were not detected in the control plant. Foliar spraying of tomato with aspartic acid only induced the appearance of two newly protein bands at MWs of 202 and 17.96 KDa. Whereas, the protein band which has the MWs of 21.84 kDa characterized the plants treated with salinity levels and glutathione. These results confirmed the results reported by El-Bassiouny *et al.* (2008) who concluded that one of the important mechanism involved in the cell protection against salinity stress is the induction of de novo synthesis of a set of new protein. The salinity altered the protein patterns of two *Anabaena* strains by inducing the synthesis of a specific of proteins called the salt-stress proteins that are strain dependent **(**Apte and Bhagwat 1998).

Table 3. Effect of foliar spraying with aspartic acid and glutathione on electrophoretic pattern of the leaves of tomato plants irrigated with different concentrations of diluted sea water.

MW (kDa)	Tap water	Concentrations of SW (%)			AA + Tp	Aspartic acid + SW (%)			GIT + Tp	Glutathione + SW (%)		
		4	8	16		4	8	16		4	8	16
355	+	+	+	+		+	+	+	+	+	+	+
296			+	+	+	+					+	
202					+	+	+					
164.17	+				+			+	+	+	+	
132.19		+	+	+	+	+	+				+	+
106.44	+		+					+	+	+		
90.05		+		+	+	+	+				+	+
77.40	+	+	+						+	+		
62.51	+		+	+	+	+	+	+	+	+	+	+
54.12		+	+	+	+	+	+	+	+	+	+	+
48.06	+	+	+	+	+	+	+	+	+		+	+
44.39	+	+		+	+	+			+	+	+	
42.11			+				+	+		+	+	+
39.25	+	+	+	+	+	+	+	+	+	+	+	+
37.26	+	+	+	+	+	+	+			+	+	+
35.08	+			+	+		+	+	+	+		+
33.68	+	+	+	+		+	+	+	+	+	+	+
31.95		+	+	+	+	+	+	+	+	+	+	+
30.21	+		+	+	+	+	+	+	+	+	+	+
28.50		+	+	+	+	+	+	+	+	+	+	+
27.20	+	+	+	+	+	+	+	+	+	+	+	+
25.91	+	+	+	+	+		+	+	+	+	+	+
24.83	+	+	+	+	+	+	+	+	+	+	+	+
22.98	+	+	+	+	+	+	+			+	+	+
21.84			+	+					+	+	+	+
19.28		+	+	+	+	+	+	+	+	+	+	+
19.52	+		+	+	+	+	+	+	+	+	+	+
17.96					+	+	+					
16.61	+	+		+	+	+	+	+	+	+	+	+
14.60	+	+	+	+	+	+	+	+	+	+	+	+
10.16	+	+	+	+	+	+	+	+	+	+	+	+
7.26	+	+	+	+	+	+	+	+	+	+	+	+
Total No. of bands	21	21	25	26	26	25	26	22	24	26	27	25
No. of new bands		6	8	7	9	8	9	5	5	6	9	8

Table 4. Effect of foliar spraying with aspartic acid and/or glutathione on amino acids content of tomato plants irrigated with different concentrations of diluted sea water.

Major omponents (µg/ml)	Tap water	Concentrations of SW (%)			Tp + AA	Aspartic acid + SW (%)			Tp + GlT	Glutathione + SW (%)		
		4	8	16		4	8	16		4	8	16
Aspartic	13.15	13.52	14.53	16.14	17.74	31.16	44.67	15.85	16.74	29.41	38.65	14.32
Threonine	5.07	3.78	2.41	1.37	6.02	7.23	10.03	5.18	5.62	6.51	9.85	5.71
Glutamic acid	19.51	25.41	28.52	31.94	25.31	41.94	54.21	40.38	33.62	41.25	48.31	20.21
Proline	3.37	18.16	26.72	29.12	14.51	25.31	34.61	17.65	11.31	23.62	28.61	7.65
Glycine	5.08	4.46	5.02	4.11	5.66	13.31	15.78	5.18	6.23	10.96	13.21	4.44
Alanine	5.40	4.38	5.10	3.52	5.02	10.62	14.90	4.64	4.91	9.35	12.61	4.21
Cystine	-	-	-	0.0081	0.0048	0.0011	0.0041	0.0014	0.0032	0.0043	0.005	0.0012
Methionine	2.31	1.70	1.35	1.12	3.41	4.31	6.31	2.98	4.06	4.62	5.24	3.90
Isoleucine	3.57	2.93	3.15	1.96	4.15	5.30	6.45	3.25	3.96	4.82	6.21	3.14
Leucine	5.12	5.16	5.11	4.42	6.13	10.23	15.62	4.61	6.41	8.76	13.58	3.92
Tyrosine	2.32	5.00	8.10	3.67	8.5	3.21	10.21	1.93	7.3	4.15	9.25	1.14
Phenyl-alanine	6.21	3.69	5.31	4.27	5.31	10.21	13.21	4.32	4.35	8.65	11.45	4.06
Histidine	6.10	7.34	7.91	7.13	5.23	11.42	18.65	5.31	4.61	8.96	15.32	4.97
Lysine	5.17	5.46	7.23	6.34	7.08	11.21	16.37	4.13	5.54	10.53	14.14	3.81
Arginine	5.67	6.06	9.61	11.94	9.52	19.43	22.31	12.32	8.93	9.41	18.51	10.61
Serine	4.75	2.96	4.12	3.54	7.94	8.21	16.67	4.13	6.95	7.63	13.85	3.72
Valine	0.86	0.81	0.86	0.03	2.41	7.93	8.87	0.75	2.13	6.52	7.10	0.71

Salinity stress affected free amino acids in the shoot of tomato plants and the use of aspartic acid or glutathione showed a clear trend for amino acid constituent (Table 4). Results showed that different salinity levels led to reaccumulation of aspartic, glutamic, proline, histidine, and arginine, since these amino acids reached their maximum increasing under the highest saline concentration (16%) whereas, tyrosine and lysine were increased at 8% salinity level compared to that of the

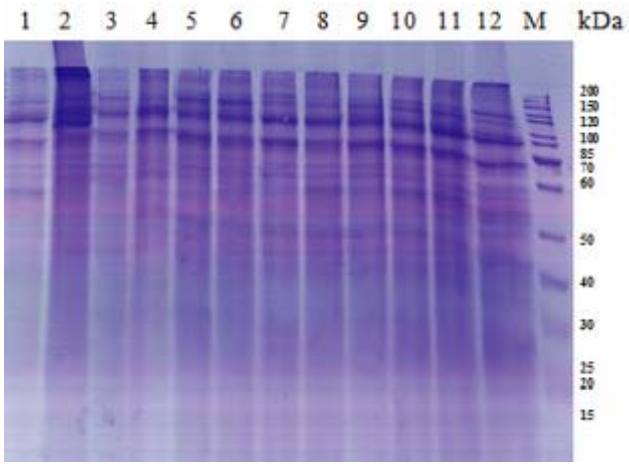

Fig. 4. Protein profile on SDS-PAGE of the leaves of tomato plants as influenced by foliar spray with aspartic acid or glutathione and grown under salinity stress. Where: (M) = Marker protein ; (1) = Control; (2) = 4% SW ; (3) = 8% SW ; (4) = 16% SW ; (5) = AA + Tp ; (6) = AA+ 4% SW ; (7) = AA+ 8% SW ; (8) = AA+ 16% SW ; (9) = GlT +Tp ; (10) = GlT + 4% SW ; (11) = GlT + 8% SW ; (12) = GlT + 16% SW.

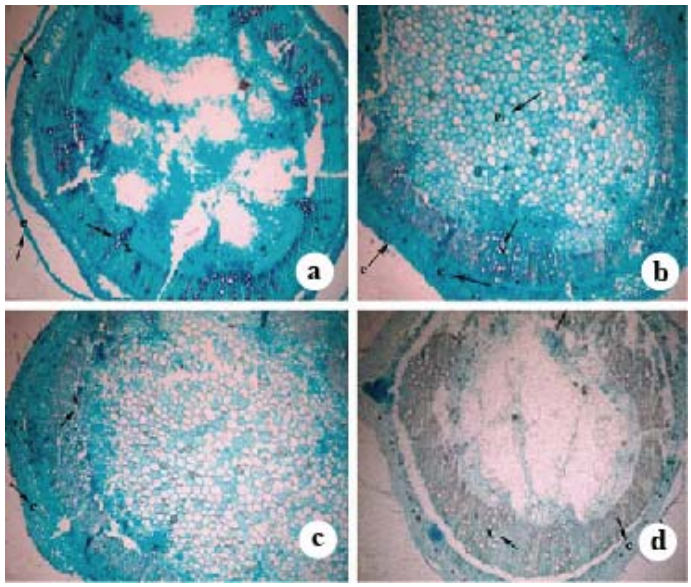

Fig. 5. Transverse sections of stem of tomato plants treated with diluted sea water concentrations; (a) Control (X40). (b) 4% SW (X40). (c) 8% SW (X40). (d) 16% SW (X40). (e) epidermis, (c) cortex, (x) xylem, (Pi) pith.

Table 5. Counts and measurements (µm) of certain histological features in transverse section through the stem of tomato plant treated with aspartic acid and/or glutathione and grown under different concentrations of diluted sea water.

Treatments		Stem diameter	Epidermis thickness	Cortex thickness	Xylem tissue thickness
Sea water (SW)	Tap water (Control)	3475.00	12.00	310.80	145.00
	4 % SW	3581.50	12.50	350.25	160.00
	8 % SW	3187.00	11.00	250.50	125.00
	16 % SW	3050.00	10.25	225.90	110.50
Aspartic acid (AA)	AA+ Tp	3940.5	13.00	430.25	187.50
	AA+ 4 % SW	4125.00	14.00	470.50	250.00
	AA + 8 % SW	4200.00	14.75	480.00	280.00
	AA+ 16 % SW	3825.00	13.00	410.00	170.50
Glutathione (GlT)	GlT + Tp	3662.00	13.00	400.00	165.00
	GlT + 4% SW	4050.00	13.50	450.90	225.00
	GlT + 8% SW	4125.00	14.00	462.50	225.00
	GlT +16% SW	3250.00	11.30	285.20	138.50

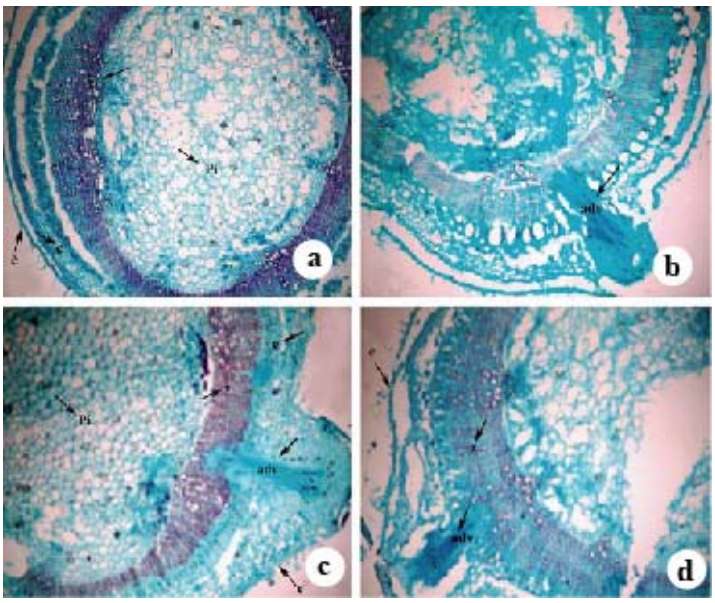

Fig. 6. Transverse sections of stem of tomato plants treated with aspartic acid and diluted sea water concentrations; (a) AA+Tp (X40). (b) AA+4% SW (X40). (c) AA+8% SW (X40). (d) AA+16% SW (X40). (e) epidermis, (c) cortex, (x) xylem, (Pi) pith, (adv) adventitious root.

control. On the other hand, all the remaining amino acids were lowered by various salinity levels. In general, data in Table 4 indicated that all the amino acids were increased by spraying the plants with aspartic acid or glutathione under non saline or saline conditions compared with control. In addition, under 4 and 8% of saline condition, the treatment of AA or GlT increased all amino acids content particularly the contents of aspartic, glutamic acid, proline and arginine while the

concentration of 16% decreased amino acids content compared with control. Amino acids represent one of the important classes of metabolites in the cell because they are the building blocks of proteins, which form the chemical basis necessary for life and have a many of roles in metabolism. Amino acids such as proline, asparagines and aminobutyric acid, play an important role in the osmotic adjustment of the plant under saline conditions (Mittler 2002). The accumulation of amino acids in plants exposed to stress probably attributed to the disturbance in amino acid metabolism (Hemmat 2007). Hassanein *et al.* (2008) indicated that arginine was the effective compound in increasing proline, total amino acid and protein contents of wheat plants under normal or stressed condition.

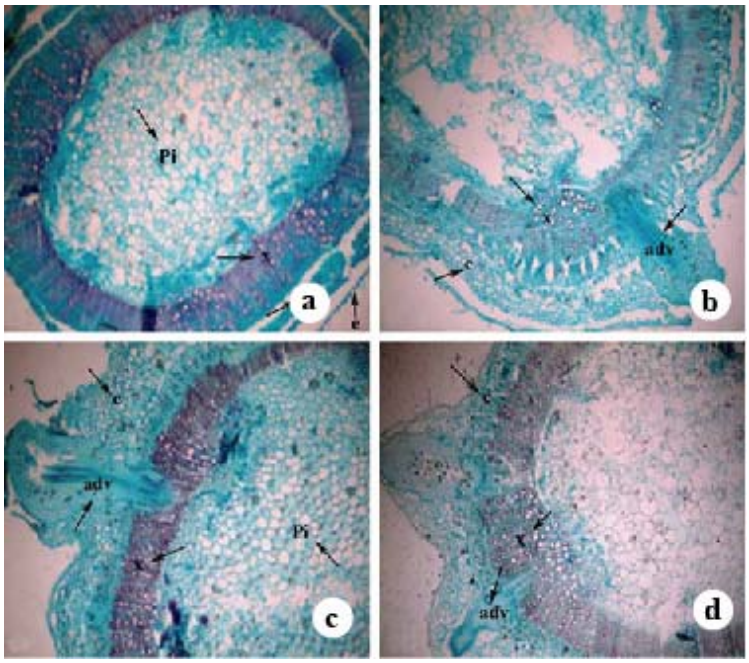

Fig. 7. Transverse sections of stem of tomato plants treated with glutathione and diluted sea water concentrations; (a) GlT+Tp (X40). (b) GlT+4% SW (X40). (c) GlT+8% SW(X40). (d) GlT+16% SW(X40). (e) epidermis, (c) cortex, (x) xylem, (Pi) pith, (adv) adventitious root.

Salt stress decreased the stem diameter, epidermis cell size, cortex zone thickness, number of xylem vessels in plants irrigated with the highest concentration of sea water (16%) in comparison with the control plants (Table 5 and Figs 5-7). On the other hand, spraying the plants with AA or GlT increased stem diameter, epidermis cell size, numbers of xylem vessels in comparison with the control plants. Moreover, application of AA and / or GlT at all different levels of salinity often caused obvious increase in the mentioned characters and the appearance of some adventitious roots which was observed only by using these amino acids accompanied by exposure the plants to salinity levels, since these adventitious roots were absent either in control plants or in plants treated with salinity only. Thus, amino acids treatment was mostly determined to have a successful performance in ameliorating the inhibitory effects of salinity on stem anatomy of tomato plants. The changes in stem anatomy of tomato plants caused by salinity stress appeared as a result of osmotic effect and the difficulty of water uptake from the saline soil. Reducing effects of salt stress on stem diameter, epidermis cell width, cortex zone thickness and xylem width were

reported previously (Pimmongkol *et al.* 2002). The present study revealed that cortex zone thickness and epidermis cell width increased under saline conditions by treating the plants with aspartic acid or glutathione could fit with those obtained by Ali (2001) who found that application of ascorbic acid induced anatomical changes in the stem of two cultivars of tomato. He also indicated that ascorbic acid applications increased stem diameter and cortical layer thickness. The presence of some adventitious roots which produced from the stem in plants treated with AA or GlT and grown under saline conditions confirm that these amino acids increase the tolerance capacity of plants to overcome the saline conditions by increasing their ability to absorb more amounts of water.

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