



Improve the salinity stress by using ascorbic acid on Germination, Growth Parameters, Water Relations, Organic and Inorganic Components of Sweet Pepper (*Capsicum annum*, L.) Plant

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ABSTRACT

The relationship between compatible solutes (osmolytes) and antioxidants are the strategies that plants have developed to tolerate salt stress. A greenhouse experiment was carried out to study the response of presoaked Sweet Peppers (*Capsicum annum*, L.) seeds in freshly prepared ascorbic acid (50 ppm \approx 0.3 mM ASC) or distilled water (control) for 12 h at natural environmental conditions, to reduce the effect of salinity stress. Generally, the sweet pepper seeds germination occurred after 4 days, while, the germination rate (%) were more faster after soaking the seeds in ascorbic acid compared with control (soaked in distilled water). NaCl salt-stress treatments caused a reduction in all growth parameters (fresh and dry weights of plant, leaf area and number per plant) compared control, particularly at high NaCl level (250 mM) more reduced. In the mean time, ascorbic acid had reduced the effect NaCl salinity stress on all growth parameters. Photosynthetic pigments (chlorophyll a & b and carotenoids) and chloroplast efficiency were increasing with salinity stress, but the response was more pronounced at 250 mM NaCl whether alone or combined with ascorbic acid. Also, salinity stress treatments tended to increased all of the total available carbohydrates (Monosaccharide, Disaccharides & polysaccharides), nitrogenous components (protein, amino acids & proline), antioxidase, (catalase, peroxidase & superoxide dismutases) enzymes activities and inorganic mineral elements (Na^+ , K^+ , N^{+3} , P^{+3} , Ca^{+2} , Mg^{+2} & Cl^-) but after soaked the seeds in ascorbic acid, these components tended to increased more. Application of NaCl salinity-stress on tomato plant induced the synthesis of nitrogenous components (protein, amino acids, proline), whereas, the tomato seeds soaked before planting in ascorbic acid which leads to remarkably increasing more for all nitrogenous components, antioxidase, carbohydrates and inorganic mineral elements content.

Keywords: Sweet Peppers (*Capsicum annum*, L.); ascorbic acid (ASC); NaCl salinity; growth parameters; Pigments; Organic Components, Antioxidase, Elements.

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1. Introduction

Sweet Peppers (*Capsicum annuum*, L.) Green, Yellow and Red Bell Peppers - What's the Difference? Bell peppers are members of the Nightshade family of vegetables along with potatoes, tomatoes and eggplants. Like chili peppers, bell peppers originated in South America where seeds of a wild variety are believed to date back to 5,000 B.C. The various colored Bell Peppers all come from the same plant, but differ in their level of maturity:

Green Peppers: Green bell peppers are harvested before they are fully ripe, one reason they are less expensive than other varieties. Green bell peppers will continue to first turn yellow and then red if they are left on the plant to mature. They have a slightly bitter flavor and will never have the sweet taste of their red, yellow and orange counterparts.

Orange and Yellow Peppers: More mature than green bell peppers, yellow and orange peppers have a fruity taste but are not as commonly found in local markets as green and red bell peppers.

Red Peppers: These are more mature than green, orange or yellow bell peppers. They are rich in carotenoid phytonutrients and contain almost eleven times more beta-carotene than green bell peppers as well as one and a half times more vitamin C. Red Bell Peppers have a sweet, almost fruity taste. Pimento and paprika are both prepared from red bell peppers. There are also other varieties that have a more tapered shape and do not have the lobes characteristic of the green, orange yellow and red varieties.

Sweet pepper plant one of the important and widespread crops in the world, is sensitive to moderate levels of salt in the soil. Generally, salinity problems in the world are due to sodium salts, particularly sodium chloride. Unfortunately, water supplies are often of poor quality (drainage water) as a result of new irrigation projects consequently a high rate of evapotranspiration with low rainfall leads to increased the levels of soluble salts in the soil. The roles of potassium and sodium in plant nutrition have occupied numerous investigators, potassium is the only monovalent cation which is essential for all higher plants, although sodium can have beneficial effects on plant growth (Epstein, 1972). Much salinity resulted from NaCl cause osmotic pressure of external solution become more than osmotic pressure of plant cells which is require to regulating osmotic pressure to preventing dehydration by plant cells. Uptake and transform of nutrition ions such as potassium and calcium, by excess sodium would make problems. High Na and Cl rates would cause to direct toxic effects on enzymatic and membranous systems (Nazarbeygi et al., 2011).

Sweet pepper is one of the most important economic crops in many countries. It belongs to fruit vegetables and its flesh is bapplied not only as salad, but also as fresh bfoods. The fruits contain a wide array of phytochemicals with well-known antioxidant properties (Hervert-Hernández et al., 2010); hence they have known to grow under stress conditions. The red pepper fruit has been used as a spice and a source of pigments to change the colour and flavor of foodstuffs (Mínguez-Mosquera et al., 2008).

Ascorbic acids is present in all living plant cells, the largest amounts being usually in the leaves and flowers, i.e., in actively growing parts (Smimoff et al., 2001; Ebrahim, 2005). The fact that it is very sensitive to reversible oxidation (ascorbic acid \leftrightarrow dehydro-ascorbic acid) suggests that it may be involved in cellular oxidation-reduction reactions, perhaps serving as a hydrogen-transport agent. Attempts have been made to employ active vitamins to overcome the drastic effects of salinity on seed germination and seedling growth as well as on some metabolic mechanisms (Khan and Zaidi, 1985; Ansari and Khan, 1986; Samiullah and Afridi, 1988). ASC is one of the most powerful antioxidants, the supply of ascorbic acid to tomato seedling might decrease the synthesis of active oxygen species and thereby increase resistance to salt stress. ASC is an essential cofactor for α -ketoglutarate-dependent dioxygenases (e.g. prolyl hydroxylases) important for formation of covalent adducts with electrophilic secondary metabolites in plants (Lopez-Munguia et al. 2011; Traber and Stevens 2011). The exogenous application of ascorbic acid could mitigate reduce the harmful effects of salinity in different crops (McKersie, et al., 1999; Al-Hakimi and Hamada, 2001; Prochazkova et al., 2001).

Many strategies used to combat salinity stress, exogenous application of plant growth regulators has received considerable attention (Afzal et al., 2005; Dolatabadian et al., 2008). Soaking seed before planting with growth regulators is beneficial in reducing negative effects of salinity on growth and physiological/biochemical responses of crops (Ashraf and Rauf, 2001; Afzal et al., 2005). Ascorbic acid also benefitted growth and may be due to the antioxidant activity of ascorbic acid protecting plants from damage due to abiotic stress (Beltagi, 2008). Priming typically affects germination time, leading to better growth and improved yield, especially in plants under stress (Halmer, 2004; Afzal et al., 2005; Piri et al., 2009).

The present work is an attempt to seeking means of improving the survival of horticulture Sweet Peppers (*Capsicum annuum*, L.) plant under experimental saline stress by soaking the seeds in Ascorbic acid (50 ppm ASC) for 12 h before. Applications of ASC for reduced the effects of NaCl salinity stress on germination, growth parameter, water relations, photosynthetic pigments and activity, antioxidant enzymes, organic and inorganic components.

2. Materials and Methods:

Plant material, germination and growth conditions:

Most other vitamins and minerals are comparable for the two varieties. All quantities and % daily values (DV) are based on one cup (92g) of raw bell peppers. A one cup measurement for other varieties is not currently available on the USDA database. No daily values for beta-carotene are currently available.

Table (1): Comparison Between Green, Red and Yellow Bell Peppers.

Nutrient	Green	Red	Yellow
Vitamin A	12% DV	105% DV	3.6% DV
Vitamin C	137% DV	292% DV	282% DV
Beta Carotene	340 mcg	841 mcg	110 mcg



<http://www.whfoods.com/genpage.php?tname=foodtip&dbid=68>

A homogenous of clean-healthy from Sweet Peppers (*Capsicum annuum*, L.) was obtained from the Crop Institute, Agricultural Research Center, Giza, Egypt. The Seeds of Sweet Peppers (*Capsicum annuum*, L.) were surface sterilized with 2% sodium hypochlorite solution for 3minutes and then thoroughly rinsed with sterile deionized water. The seeds were then soaked in freshly prepared ascorbic acid (50 ppm \approx 0.3 mM; El-Tayeb, 1995) or distilled water (control) for 12 h at natural environmental conditions. Batches of seeds were germinated in Petri dishes containing ash less filter paper moistened with a solution identical to that they had originally been soaked (distilled water and 25 mL⁻¹ Ascorbic acid). The percent of germination was recorded after 3 days. Seedlings were selected on homogeneity of height and developmental stage. Transfer the seedling tomato plants to plastic pots (5 seedlings/pot) containing clay-sand soil ($\frac{1}{2} \approx$ v/v) was used, the soil was mixed thoroughly to assure complete and uniform distribution (40 cm diameter, 60 cm depth, 7.5Kg soil pot⁻¹) at 10°C/17°C, with supplementary lighting (red electric bulb, 20 w at night), at relative humidity of 75-80%. Two pots group experiments were conducted in the screen greenhouse condition during winter season (2011/2012) treated tomato plant seedling with NaCl concentrations (50, 100, 150, 200 & 250 mM) and distilled water. The sand culture technique and nutrient solution were similar to those adopted by Hewitt (1952). One group from the pots were irrigated twice a week interval with normal tap water needed with full strength Hoagland nutrient solution (Hoagland and Arnon, 1950), while the other pots were irrigated with different concentrations from NaCl with full strength of Hoagland nutrient solution, and allowed to grow for about 60 days (flowering stage) post sowing. Plant have been harvested after 60 days for determined the all growth parameters, five replicates for each treatment, were used to calculate the mean of each growth parameters. The plants of each treatments were washed with distilled water, the length plants were measured (cm/plant), Number of leaves were recorded, Leaf area (cm²/leaf) assessed using the leaf No. 4 from the lower, by a Portable Area Meter Leaf Area Meter (*Laser Leaf Area Meters CI-202*), CID, Bio-Science, USA. The fresh after weighing, then dried at oven for drying at a temperature of 80- 85°C for 72 h (g/plant).

Water Relations:

Succulence and Dry matter content (%): The percentage of the Succulence content and dry matter content (D.M.C. %) was determined after drying the shoot and root samples in air – circulation oven at 80°C after constant weight, and calculated as the following equation:

$$\text{Succulence} = \text{F. Wt.} / \text{Oven D. Wt} \dots\dots\dots(1)$$

$$\text{D.M.C. \%} = (\text{Oven D. Wt.}/\text{F. Wt.}) \times 10 \dots\dots\dots(2)$$

Measurement of Relative Water Content: The relative water content (R.W.C. %) was measured according to a modification of the method of Weatherly (1950); Slatyer (1957); Weatherly and Barr (1962). Detached leaf samples were weight immediately and floated on distilled water in a darkened refrigerator (5°C). Saturation of the leaves was attained after 24 h. and the leaves were rapidly and thorough blotted and weighed immediately. The leaves were then dried at 80°C to constant weight in an air – circulation oven to constant weight. The relative water content of leaves was expressed according to the following equation:

$$\text{R W C \% (S. Wt. \%)} = \frac{(\text{F. wt.} - \text{Oven D. Wt.}) \times 100}{(\text{Saturated Wt.} - \text{Oven D. Wt.})} \dots\dots(3)$$

Physiological Studies

Photosynthetic Pigments: Chlorophyll a, chlorophyll b and carotenoids of leaves were determined spectrophotometrically as the method described by Metzner et al. (1965). An 85% aqueous acetone extract of a known



F.W. of leaf was assayed *Spectrometrically* (LKB NOVASPEC) at 664, 645, 420 nm. The following equations were used to determine the concentration of the pigment fractions as $\mu\text{g/ml}$.

$$\text{Chlorophyll a} = 10.3 E_{664} - 0.918 E_{645} \dots\dots\dots (4)$$

$$\text{Chlorophyll b} = 19.7 E_{645} - 3.870 E_{664} \dots\dots\dots (5)$$

$$\text{Carotenoids} = 403 E_{452} - (0.0264 \text{ Chl. a} + 0.426 \text{ Chl. b}) \dots\dots (6)$$

The pigment fractions were calculated as $\mu\text{g Chl./mg D.W.}$

Photosynthetic Activity: Chloroplasts were prepared by the method of Aronoff (1949) and Osman, *et al.* (1982). Fresh leaves were shredded, ground for one min in a blender, using a buffered solution of 0.4 M sucrose, 20 mM HEPES-KOH (pH 7.8), 3 mM MgCl_2 , 4 mM sodium ascorbate and 0.1% bovine serum albumin (BSA). The much was strained through cheese-cloth, filtered and the suspension centrifuged (1 min at 8,000 X g). The pellet was re-suspended in the isolation medium, centrifuged (5 min at 300 X g) and the supernatant re-centrifuged (10 min at 1,000 X g). The sediment was re-suspended in a 2 ml buffer solution at pH 6.8 and the aggregates dispersed (Osman *et al.*, 1982). The levels of chlorophyll a & chlorophyll b were determined by the method described by Mackinney (1941). An aliquot of 0.2 ml of the chloroplast suspension was extracted with 3.8 ml of 85% cold aqueous acetone and the density of the extract measured at 652 nm. The chlorophyll content was calculated according to the following equation :

$$C = E_{652} \times 1,000 / 34.5 \text{ mg chl./L} \dots\dots\dots (7)$$

Where C = Chlorophyll a & b.

The photosynthetic activity of the isolated chloroplasts was measured using potassium ferricyanide ($5 \times 10^{-4}\text{M}$) as an electron acceptor. Reduction of ferricyanide was monitored spectrophotometrically (LKP NOVASPEC) at 420 nm at room temperature. The reduction mixture contained 0.2 ml of chloroplast suspension, (0.2-0.8 mg chl. ml^{-1}), 3.8ml HEPES buffer (pH 7.8), and $5 \times 10^{-4}\text{M}$ potassium ferricyanide. The mixture was illuminated at 300 Wm^{-2} using a slide projector provided with a heat filter with a 24 v, 250 w quartz halide bulb, 15-45 cm from the well. The photosynthetic activity of the isolated chloroplasts was calculated from the standard curve and expressed as $\mu\text{mol fericyanide mg chl}^{-1} \text{ h}^{-1}$ (Arnon and Shavit, 1963).

I- Organic Components:

Carbohydrate: 300 mg of oven dry plant material was extracted with 5 ml of borate buffer (28.63 g boric acid + 29.8 g KCl + 3.5 g NaOH in a liter of hot distilled water), left for 24 hr, then centrifuged and filtered. The filtrate was used for the determination of the direct reducing value (DRV-including all free monosaccharide) and total reducing value (TRV-including sucrose), while the residue was dried at 80°C for determination of polysaccharides (Naguib, 1963 & 1964).

Direct Reducing Value (DRV), was carried out by evaporation, 0.1 ml of extracted cleared borate buffer was reduced to dryness and then mixed with 1 ml of modified Nelson solution (Naguib, 1964). The mixture was maintained on a boiling water-bath for 15 min, after which it was cooled rapidly using running tap water. Thereafter 1 ml of arsenomolybdate (Nelson, 1944) was added, the mixture was diluted to a definite volume, and its intensity measured at 700 nm, using colorimeter (LKP NOVASPEC Surplus Model 4049 Spectrophotometer).

Total Reducing Value (TRV): For determination of total reducing value (TRV), 0.2 ml of cleared extract was mixed with deionized water up to 5 ml then 0.2 ml of the diluted extract was mixed with 0.1 ml of 1% invertase enzyme solution and the mixture maintained at 37°C for 0.5 hr. Thereafter, the reducing value was determined as described before for DRV (Naguib, 1963 & 1964). The difference between the value obtained from this step and that of the DRV is an estimated of **sucrose**, in terms of glucose made up to 3 ml left overnight at 28°C and then centrifuged.

Polysaccharides: 10 mg of the remaining residue was mixed with 0.2 ml of 1% taka diastase enzyme and 0.1 ml acetate enzyme and ml acetate buffer (6 ml acetic acid 0.2N+4 ml sodium acetate buffer 0.2 N). The reducing value of 1ml of filter was estimated as above (Naguib, 1963).

Proteins contents: Dry samples collected during the growth study were analyzed for protein content, after precipitating the protein with 15% TCA at 4°C according to Lowry *et al.* (1951).

Total Free Amino Acids contents: These were determined by the method described by Ya and Tunekazu (1966). An aliquot of 0.1 ml plant extract was heated in a test tube with 1.9 ml of ninhydrin citrate buffer-glycerol mixture in a boiling water bath for 12 min, and cooled at room temperature. Then the tube was well shaken and the optical density read at 570 nm. A blank was determined with 0.1 ml of distilled water and a standard curve obtained with 0.005 to 0.2 mM g Glycine.

Proline contents: This was estimated using the acid ninhydrin method described by Bates *et al.* (1973). Two ml of water extract were mixed 10 ml of 3% aqueous sulfosalicylic acid. Two ml of this mixture was allowed to react with 2 ml acid ninhydrin-reagent and 2 ml of glacial acetic acid in a test tube for 1 h at 100°C ; the reaction was terminated by cooling the mixture in an ice bath. The reaction mixture was extracted with 4 ml toluene, and mixed vigorously for 15-20s. The chromatophore - containing toluene was aspirated from the aqueous phase, warmed to room temperature, and the absorbance read at 520 nm using toluene as a blank. The proline concentration was determined from a standard curve.

Total indole: as described by Larsen *et al.* (1962) and **total phenol**, as described by Malik and Singh (1980) were estimated in the fresh shoots.



Determination of antioxidant enzyme activities: The **catalase** (CAT, EC 1.11.1.6) activity was assayed from the rate of H₂O₂ decomposition following the method of Cakmak and Horst (1991).

Peroxidase: (POD, EC 1.11.1.7) following the method of Macheix and Quessada (1984) and **superoxide dismutases:** (SOD, EC 1.15.1.1) as described by Dhindsa et al. (1981).

II- Inorganic Components:

Mineral Elements: Ions content measurements were carried out after extraction with 0.1 nitric acid of the ashed (powdered) milled samples at 500°C obtained after combustion in a muffle furnace, the milled samples were estimated following the "wet ashing procedure" (Richards, 1954); the acid digests of the oven dried samples were analysed. Oven dried plants were subjected to acid digestion and sodium (Na⁺) and calcium (Ca²⁺) estimated photo-metrically using a corning-400 flame photometer (Johnson and Ulrich, 1959; Allen et al., 1974). Phosphorous (P⁺³) was estimated using the method of Sekine et al. (1972) by the Molybdenum-blue method (Allen et al., 1974), while nitrogen was estimated by the Automatic MicroKjeldahl method (Allen et al., 1974). Determination of potassium (K⁺) content by Miller (1998) methods, the plant parts were dried in a ventilated oven for approximately 78 h at 60°C to a constant weight and then ground, the samples were digested in a nitric-perchloric acid mixture and analyzed with Atomic Absorption Spectrometer (*Carl Zeiss Jena, Germany*). Chlorides were determined by the AgNO₃ titration method as described by Jackson and Thomas (1960).

Statistical Analysis: All data were subjected to *F* test ANOVA and the means were compared using Duncan's multiple range (*P*<0.05). Where relevant, the experimental data was subjected to analysis of variance. Percentage values were transformed into arcsines according to Bliss (1973) and analysis of variance was carried out according to Snedecor and Cochran (1967)

3. Results and discussion

Germination Rate: Results presented in Table (2) indicated that the germination rate (%) of Sweet Peppers (*Capsicum annuum*, L.) seeds increased with seeds soaked in ascorbic acid (ASC) more than the seeds soaked in distilled water. Generally, the sweet pepper seeds germination occurred after 4 days, but the germination rate were more faster after soaking the seeds in ascorbic acid (ASC compared with control (distilled water). Germination is a crucial stage in seedling establishment and plays a key role in crop production. The germination process comprises two distinct phases the first is imbibitions, mainly dependent on the physical characteristics of the seeds and the second is a heterotrophic growth phase between imbibitions and emergence (Khajeh-hosseini et al, 2003; Akbari ghogdi et al., 2012). The ascorbic acid plays a remarkable role in case of seed germination, cell growth under salinity due to its antioxidants properties (Netondo et al., 2004).

Table (2): Impact of Ascorbic Acid (ASC) on germination rate of Sweet Peppers (*Capsicum annuum*, L.) seeds.

Germination Rate (%)	Time / Days									
	1	2	3	4	5	6	7	8	9	10
Ascorbic acid (50 ppm ≈ 0.3 mM)										
(- ASC)	----	----	----	9	18	32	59	72	89	100
(+ASC)	----	----	----	38	63	86	92	100	100	100
<i>F</i> Values	----	----	----	***	**	**	*	*	*	N.S.

-ASC = Soaking Seeds in distilled water before germinated, Without Ascorbic Acid

+ASC = Soaking Seeds in Ascorbic Acid before germinated, With Ascorbic Acid

Values are expressed as the mean of five samples ± Standard Deviation (±SD)

Statistical Analysis treatments, where relevant, the experimental data were subjected of One – Way analysis of variance (ANOVA). Note: *F* values * = *P* < 0.05, ** = *P* < 0.01, *** = *P* < 0.001 and N.S. = Not Significant.

The role of ascorbic acid (ASC) in seed germination and cell growth under salinity is remarkable its anti-oxidant activity, rather than its possible utility as an organic substrate for respiratory energy metabolism. So, after soaking the Sweet Peppers seeds in Ascorbic acid (ASC), the rate of germination increased with times. The effect of ascorbic acid on plant survival is associated with the partial inhibition of a few interactions in active oxygen species production. An artificial increase in cellular level of an antioxidant such as ascorbic acid should be beneficial in improving stress tolerance at germination level (Shalata, and Neumann, 2001; Khan et al., 2006a).

The similar results on Sweet Peppers seeds germination were reported also by treatment the seeds with exogenous ascorbic acid increasing the level of ascorbic acid uptake by different tissues under salinity (NaCl) stress (Shaddad et al., 1990; Arrigoni and De Tullio, 2000; Arab and Ehsanpour, 2006).

Growth Parameters: Generally, salinity stress in Sweet Peppers plants retards all major growth processes that have been examined, the growth parameters decreased significantly with increasing NaCl salinity concentrations (50, 100, 150, 200 & 250 mM), whereas, the level of growth increased more in the presence of ascorbic acid (+ASC), compared as



control (-ASC and +ASC) without salt-stress, as shown in Table (3). Sweet peppers plant tended to increased significantly the growth parameters (plant height, leaf number and area, fresh and dry weight) in the presence of ascorbic acid (+ASC) compared to control (sweet pepper) plant. Overall, Sweet Peppers plant height tended to decreased with increasing NaCl salinity concentrations while after soaked the seeds of sweet peppers in ascorbic acid (+ASC) tended to increased plant height. So, added ascorbic acid (+ASC) decreased the effects of salinity stress with all NaCl salinity concentration compared with control. The results agree with Ejaz et al. (2012) they found that the leaf area per plant was significantly reduced under salt stress, while ascorbic acid (ASC) applications markedly improved the inhibitory effects of salt on plants. The fresh weight in four tomato cultivars decreased with salinity stressed (Ashraf and Harris, 2004; Okhovatian-Ardakani et al., 2010; Ali et al., 2011).

Hence, it is assumed that exogenous ascorbic acid (ASC) improves seed tolerance to salinity significantly. Salinity reduces plant productivity first by reducing plant growth during the phase of osmotic stress and subsequently by inducing leaf senescence during the phase of toxicity when excessive salt is accumulated in transpiring leaves (Munns, 2002 b). Water deficits may exert their effects directly on cell extension and division (Greenway and Munns, 1983). Ascorbic acid (ASC) influence mitosis and cell growth in plants, affects phyto-hormone-mediated signaling processes during the transition from the vegetative to the reproductive phase as well as the final stage of development and senescence (Smirnoff and Wheeler, 2000; Barth et al., 2006). The beneficial effect of ascorbic acid (ASC) on plant height may be attributed to the fact that ascorbic acid (ASC) is involved in the regulation of shoot and root elongation, cell vacuole, Leaf area and cell expansion (Smirnoff, 1996; Sumalan and Carmen, 2002; El Hariri et al., 2010; Farahat et al., 2013). Although ascorbic acid is one of the most important and abundantly occurring water soluble antioxidants in plants, relatively little is known about its role in counteracting the adverse effects of salt stress on plant growth (Beltaji, 2008; Athar et al., 2008).

Table (3): Interactive effects of Ascorbic Acid (ASC) and NaCl Salinity Treatments on Plant Height, Leaf Area (cm²) and Number, Fresh and Dry Weight of Sweet Peppers (*Capsicum annuum*, L.) Plant.

The leaf area was estimated in the 6th leaves from the shoot top. Plants were grown, under glasshouse conditions, for 90 days after germination and irrigated at 80% field capacity.

Growth Parameters	NaCl Salinity Concentrations (mM)					
	0.0	50	100	150	200	250
Ascorbic Acids (50 ppm ≈ 0.3 mM)						
Plant Height (cm/plant)						
(- ASC)	34.1±0.39	32.6±0.41	28.3± 0.40	23.1± 0.34	20. 7± 0.52	18.2± 0.22
(+ASC)	39.3±0.58	44.5±0.35	45.4± 0.25	41. 3± 0.45	38.5± 0.41	28.3± 0.41
F Values	**	*	**	*	*	*
Leaf Number						
(- ASC)	11.5±0.11	10.6 ±0.31	10.1 ±0.26	8.6±0.43	7.3±0.18	5.6±0.128
(+ASC)	14. 7±0.23	17.5± 0.24	13.7±0.48	11.3± 0.33	10.6±0.48	8.4±0.38
F Values	*	**	**	**	**	**
Leaf Area (Cm ² /Leaf)						
(- ASC)	32.37±0.24	38.57±0.58	34.88±0.25	30.70±0.51	27.37±0.46	20.31±0.46
(+ASC)	55.20±0.47	52.50±0.35	46.03±0.45	42.64±0.40	36.23±0.69	30.23±0.69
F Values	**	**	**	**	**	**
Plant Fresh Weight (g/Plant)						
(- ASC)	26.75±0.19	34.67±0.07	30.38±0.21	22.40±0.06	15.69±0.02	13.69±0.02
(+ASC)	38.98.±0.26	47.35±0.30	42.71±0.29	34.95±0.09	21.56±0.13	16.56±0.13
F Values	N.S.	N.S.	*	*	*	*
Plant Dry Weight (g/Plant)						
(- ASC)	1. 34±0.02	1. 71±0.03	1.31±0.02	1.26±0.03	1.18±0.01	1.08±0.01
(+ASC)	1. 70±0.03	1.61±0.04	1.81±0.03	1.31±0.02	1.29±0.02	1.12±0.02
F Values	N.S.	*	N.S.	N.S.	N.S.	N.S.

-ASC = Soaking Seeds in distilled water before germinated

+ASC = Soaking Seeds in Ascorbic Acid before germinated

Values are expressed as the mean of five samples ± Standard Deviation (±SD)



Statistical Analysis treatments, where relevant, the experimental data were subjected of One – Way analysis of variance (ANOVA). Note: F values * = $P < 0.05$, ** = $P < 0.01$, *** = $P < 0.001$ and N.S. = Not Significant.

Water Relations: Data presented in Table (4) indicated that the succulence and relative water content (RWC %) decreased significantly with increasing NaCl salinity concentrations, especially in the absence of ascorbic acid (-ASC) more than in the presence of ascorbic acid (+ASC) in Sweet Peppers plant. Whereas, dry matter content % (DMC %) increased significantly with increased NaCl salinity concentration in the absence of ascorbic acid (-ASC) more than in the presence of ascorbic acid (+ASC). So, increasing NaCl salinity concentration tended to reduced the absorption of water leading to a drop in water content, the inhibitory effect of NaCl on growth parameters could be attributed to the osmotic effect of NaCl salinity, in addition, the changes in water status under NaCl stress may cause a reduction in meristem activity as well as cell elongation (Salter et al., 2007; Shah, 2007; Chookhampaeng, 2011). These suggestions are in a good agreement with present results, which showed that the increase of WC and RWC was associated with a decrease in transpiration rate.

Further, it could be suggested that the effectiveness of ASC depends on its mode of application, which may enhance the endogenous level of ASC and water status of treated plants (Azooz, 2004; Alqurainy, 2007; Athar et al., 2008). Search results consistent with the findings of the Sairam et al. (2002) and Ghoulam et al. (2002) they concluded that the salt treatment induced a reduction in leaves RWC. While, Mandhania et al. (2012) found relative water content (RWC) decreased progressively with increasing duration and levels of NaCl in the leaves of both salt-sensitive WH-542 and salt-tolerant KRL-19 cultivars. The adverse effects of NaCl on *Silybum marianum* L. plants the growth parameters, water content (WC) and relative water content (RWC) were mitigated by seed 100 ppm vitamin C (Summart et al., 2010; Ekmekçi and Karaman, 2012).

Table (4): Interactive Effects of Ascorbic Acid (ASC) and NaCl Salinity on Water Relations and Dry Matter Content of Sweet Peppers (*Capsicum annum*, L.) Plant.

Plants were grown, under glasshouse conditions, for 60 days after germination and irrigated at 80% field capacity.

Water Relations Ascorbic Acids (50 ppm ≈ 0.3 mM)	NaCl Salinity Concentrations (mM)					
	0.0	50	100	150	200	250
Succulence (Fresh Weight / Oven Dry weight)						
(- ASC)	19.96±0.12	20.34±0.23	23.19±0.26	17.78±0.23	13.30±0.36	12.68±0.36
(+ASC)	22.93±0.21	29.41±0.31	23.60±0.18	26.68±0.38	17.11±0.43	14.79±0.43
F Values	N.S.	*	N.S.	*	*	*
Dry Matter content (%)						
(- ASC)	5.01±0.09	4.93±0.31	4.31±0.23	5.63±0.61	7.52±0.56	7.89±0.56
(+ASC)	4.36±0.11	3.40±0.24	4.24±0.16	3.75±0.44	5.98±0.39	6.76±0.39
F Values	N.S.	N.S.	N.S.	**	*	N.S.
Relative Water Content % (Saturated Weight)						
(- ASC)	88.45±0.19	86.93±0.21	85.72±0.25	84.78±0.34	84.03±0.75	86.63±0.75
(+ASC)	89.82±0.26	88.43±0.37	89.79±0.28	89.27±0.81	88.15±0.93	89.55±0.93
F Values	N.S.	N.S.	*	**	*	*

Photosynthetic Pigments and Chloroplast efficiency:

Generally, the chloroplast pigments (chlorophyll a, chlorophyll b, and carotenoids) and photosynthetic efficiency exhibited marked significantly increased with salinity concentration, whereas more increased in the presence of ascorbic acid (+ASC) over control (-ASC) as presented in Table (5). Significant increases in the above mentioned characters were recorded from NaCl treatment plants compared to plants in the presence of ascorbic acid (+ASC). Salt stress (NaCl) has both osmotic (cell dehydration) and toxic (ion accumulation) effects on plants, impairing growth, ion homeostasis, photosynthesis and nitrogen fixation, among other key physiological processes (Zhu, 2001; Munns, 2002a; Tejera et al., 2004; Bartels and Sunkar, 2005).

Chlorophylls (a & b) and Carotenoids are main photosynthetic pigments and they play important role in photosynthesis, the changes in the amount of pigment were evaluated as the changes in photosynthesis. So, the changes of pigment contents under salt stress are used as parameter for selection of tolerant and sensitive cultivars in crop plants (Eryilmaz, 2007). Increasing sodium concentration in plant tissue can increase oxidative stress, which causes deterioration in chloroplast structure and an associate lose in chlorophyll. This leads to a decrease in chlorophyll, while increasing carotenoids content. (Khosravinejad and Farboondia, 2008; Pinheiro et al., 2008). The accumulation of soluble carbohydrates in plants has been widely reported as a response to salinity or drought, despite a significant decrease in



net CO₂ assimilation rate (Crowe et al., 2002; Murakeozy et al., 2003). The reduced water potential could be explained by the fact that during stress carbon allocation, osmotic adjustment and accumulation of soluble sugars compete with other sinks and can affect growth (Shi and Tadashi 2001; Balibrea et al., 2003; Jiang et al., 2007; Gama et al., 2007).

I- Organic Components:

Total Available Carbohydrates:

In these investigation, the total available carbohydrates (Monosaccharide, Disaccharides, polysaccharides) contents in Sweet Peppers plant increased significantly with salinity concentration at both in the presence (+ASC) or absence (-ASC) of ascorbic acid compared to control as shown in Table (5). The significantly increasing in total available carbohydrates, results from the close relationship between stomatal conductance and photosynthesis, thus lead to an increase in photosynthesis. The results of the study agrees with Farahat et al. (2013) they found that the combined treatment of ascorbic acid (100 ppm and 200 ppm) with salinity level at water salinity (3000 and 6000 ppm) gave significantly increased chlorophyll a & b, carotenoids content and total carbohydrates % of shoots and roots compared with control plants. The highest values were recorded with 200 ppm ascorbic acid. The substantial increase in carbohydrate contents may be due to the activation of photosynthetic machinery, as a result of the stimulatory effects of the used plant growth bio stimulators on photosynthetic process. Chlorophyll a & b contents and total carbohydrates % of shoots and roots were reduced as external salinity in irrigation water increased.

Table (5): Interactive Effects of Ascorbic Acid (ASC) and NaCl Salinity on Chloroplast Pigments (µg/mg Leaf F. Wt.) and Photosynthetic Activity (µmol fericyanide mg chl⁻¹ h⁻¹) of Sweet Peppers (*Capsicum annum*, L.) Plant.

The leaf was estimated in the 5Th leaves from the shoot top.

Plants were grown, under glasshouse conditions, for 60 days after germination and irrigated at 80% field capacity.

Chloroplast Pigment & Photosynthetic Activity	NaCl Salinity Concentrations (mM)					
	0.0	50	100	150	200	250
Ascorbic Acids (50 ppm ≈ 0.3 mM)						
Chlorophyll a (µg/mg Leaf F. Wt.)						
(- ASC)	4.80±0.01	6.89±0.03	7.01±0.05	8.13±0.04	9.25±0.04	10.58±0.04
(+ASC)	9.33±0.01	10.42±0.04	11.49±0.01	14.58±0.02	16.44±0.03	18.34±0.03
F Values	*	*	*	*	*	*
Chlorophyll b (µg/mg Leaf F. Wt.)						
(- ASC)	2.24±0.042	3.35±0.015	4.39±0.032	6.52±0.035	7.16±0.021	8.16±0.021
(+ASC)	4.38±0.035	5.44±0.035	7.52±0.015	8.74±0.031	10.85±0.044	11.85±0.044
F Values	*	*	*	*	*	*
Carotenoids (µg/mg Leaf F. Wt.)						
(- ASC)	3.73±0.010	4.17±0.025	6.01±0.025	7.11±0.025	8.15±0.01	9.37±0.01
(+ASC)	5.51±0.025	7.72±0.006	8.84±0.006	9.94±0.006	10.05±0.047	11.05±0.047
F Values	N.S.	*	N.S.	*	N.S.	N.S.
Total Pigments (µg/mg Leaf F. Wt.)						
(- ASC)	10.77±0.18	11.41±0.49	17.41±0.79	21.76±0.65	24.56±0.75	28.11±0.75
(+ASC)	19.22±0.74	23.58±0.54	27.85±0.63	33.26±0.48	37.04±0.81	41.24±0.81
F Values	**	*	*	**	*	*
Photosynthetic Activity (µmol fericyanide mg chl ⁻¹ h ⁻¹)						
(- ASC)	67.32±1.21	71.34±1.32	75.31±0.87	78.31±2.01	80.28±1.75	80.28±1.75
(+ASC)	73.83±2.14	79.26±2.42	83.86±2.61	86.87±2.09	89.27±2.65	89.27±2.65
F Values	*	**	**	**	**	**

Nitrogenous Components:

Protein, Total Amino Acids, Proline Contents:

Results presented in Table (7) indicated that the effect of salinity concentration on sweet peppers plant in the presence (+ASC) or absence (-ASC) of ascorbic acid exhibited marked significant increase in total protein, total amino acids, proline contents compared with control tomato plants. Moreover, the protein, amino acids, proline contents increasing in sweet peppers plants in the presence (+AsA) more than in the absence (-ASC) of ascorbic acid may be responsible for the

stimulation of growth. Accumulation of amino acids as well as proline was ascertained in different halophytes and non-halophytes under salinity conditions (Greenway and Munn 1980; Harborne 1988; Dubey and Rani 1989). It was suggested that build up of proline and other organic solutes in shoots and roots of salinized plants either contributes to osmotic balance in cells (Stewart and Lee 1974; Zidan, 1991) or helps to maintain enzymes activities (Pollard and Wyn Jones 1979; Greenway and Munns 1980).

Campbell (1977) found that the chloroplasts have paramagnetic properties. Shabrangi and Majd (2009) concluded that, biomass increasing needs metabolic changes particularly increasing protein biosynthesis. Kapoor and Srivastava (2010) observed an increase in protein content when increasing salt concentration. Proline accumulation in response to lower salt concentration may contribute positively to salt tolerance, whereas the high concentration in leaf tissues under high salinity treatment may be partly due to leaf damage. The plants under salinity condition change their metabolism to overcome the changed environmental condition.

According to Ebrahimian and Bybordi (2012) the soluble protein content decreased on account of salinity stress, one of the mechanisms affected by salt stress in plants was protein synthesis. It is known that soluble protein content is an important indicator of physiological status of plants. Proline is synthesized in plants through two alternate pathways: L-ornithine and L-glutamate pathways (Parvaiz and Satyawati 2008). Proline is a typical adaptive response in plants and it may be a part of stress signal (Maggio et al., 2002; Yang et al., 2009).

Proline, which is an amino acid, is one such organic solute that plays a major role in this osmotic adjustment (Chookhampaeng, 2011; Loukehaich et al., 2011). Likewise, in tomato salt tolerance was attributed to the degree of plant to accumulate osmoprotectants, like proline (Patel and Pandey, 2008; Dasgan et al., 2009). Proline is one of well-known osmoprotectants and its accumulation is widely observed in various organisms under salt stress.

Table (6): Interactive Effects of Ascorbic Acid (ASC) and NaCl Salinity on Total Available Carbohydrates (mg/100g D. Wt.) of Sweet Peppers (*Capsicum annum*, L.) Plant.

Plants were grown, under glasshouse conditions, for 90 days after germination and irrigated at 80% field capacity.

Carbohydrates Content (Mg/100g D. Wt.)	NaCl Salinity Concentrations (mM)					
	0.0	50	100	150	200	250
Ascorbic Acids (50 ppm ≈ 0.3 mM)						
Monosaccharide (DRV)						
(- ASC)	10.70±0.79	11.87±0.81	13.13±1.01	14.98±2.01	15.79±1.31	16.79±1.31
(+ASC)	13.30±0.70	14.71±0.72	16.81±1.91	17.31±0.69	18.81±1.98	19.81±1.98
F Values	**	*	*	**	*	*
Disaccharide (Sucrose)						
(- ASC)	11.98±0.97	13.17±2.01	15.50±1.54	17.31±1.36	18.89±1.70	19.89±1.70
(+ASC)	14.01±1.03	19.80±1.80	20.70±0.79	21.31±1.03	22.31±1.31	23.31±1.31
F Values	*	**	**	**	**	**
Polysaccharide						
(- ASC)	4.98±0.03	7.31±0.06	8.62±0.07	10.71±0.07	11.31±0.03	12.31±0.03
(+ASC)	8.31±0.03	9.91±0.09	10.81±0.08	11.82±0.08	12.09±0.09	13.09±0.09
F Values	**	**	*	N.S.	N.S.	N.S.
Total Available Carbohydrates						
(- ASC)	27.66±1.02	32.35±1.04	37.25±0.98	43.00±2.01	45.99±2.04	48.99±2.04
(+ASC)	35.62±1.71	44.42±1.27	48.31±1.09	50.44±1.69	52.21±1.83	55.21±1.83
F Values	**	**	**	*	**	**

The amino-acid may play a role in protecting membranes and proteins against adverse effects of higher concentrations of inorganic ions and temperature extremes. Chookhampaeng (2011) noted that the salinity treatments caused the increased proline content in pepper plant. One mechanisms utilized by the plants for overcoming the salt stress effects might be via accumulation of compatible osmolytes, such as proline and soluble sugar. Production and accumulation of free amino acids, especially proline by plant tissue during drought, salt and water stress is an adaptive response.

Total Phenols and Total Indole:

Also, the effect of salinity concentration on sweet peppers seeds in the presence (+ASC) or absence (-ASC) of ascorbic acid exhibited marked significant increase in total phenols and total indole compared with control plants as shown in Table



(7). Hassanein et al. (2009) and Abd-El Hamid (2009) suggested that the ascorbic acid increased IAA content, which stimulates cell division and/cell enlargement and this in turn, improved plant growth.

Antioxidant Enzymes:

Catalase, Peroxidase, and Superoxide Dismutase:

Data presented in Table (7) showed that the effect of salinity concentration on sweet peppers plant in the presence (+ASC) or absence (-ASC) of ascorbic acid increased significantly in the activities of the antioxidant enzymes (catalase, peroxidase, and superoxide dismutase), but the activity increased more in the presence of ascorbic acid (+ASC) compared with control. 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 (Mittova et al., 2003). Among them, ion homeostasis, osmotic adjustment, enhancement of antioxidant defense systems and increase of the photosynthetic ability are the most important ones (Zhu, 2000; Xiong and Zhu, 2002). It is newly clear that the antioxidant defense systems are very important for the determination of plant salt tolerance (Sairam et al., 2005;; Gossett et al., 1994; Mittler, 2002). Several studies demonstrated that salt-tolerant species increase their antioxidant enzyme activities and antioxidant content in response to salt stress, while salt-sensitive species do not (Ashraf and Harris 2004; Khan et al., 2002; Shalata et al., 2001). While, Mandhanian et al. (2012) found that the activities of Catalase (CAT) and Ascorbate Peroxidase (APX) increased with increasing the salt stress in both salt tolerant and salt sensitive wheat cultivars. Dolatabadian et al. (2008) they found the exogenous application of ascorbic acid induces activation of antioxidant enzyme system in canola (*Brassica napus* L.) resulting in reduction of detrimental effects of salinity. Antioxidative enzyme such as superoxide dismutase, ascorbate peroxidase and catalase play an important role against oxidative stress. Plants containing high activities of antioxidant enzymes have shown considerable resistance to oxidative damage caused by Reactive Oxygen Species (ROS) (Apel and Hirt, 2004; Khan et al., 2007; Gapinska et al., 2008; Frary et al., 2010). Indeed, several studies have shown that ASC plays an important role in improving plant tolerance to abiotic stress (Shalata and Neumann, 2001; Al-Hakimi and Hamada, 2001; Athar et al., 2008). Application of ascorbic acid can reduce the harmful effects of salt stress and may have stimulatory effects on plants; ascorbic acid is synthesized in the higher plants and improves plant growth. It is a product of D-glucose metabolism which affects some nutritional cycle activities in higher plants and plays an important role in the electron transport system (El-Kobisy et al., 2005).

Table (7): Interactive effects of Ascorbic Acid (ASC) and NaCl Salinity on Total Proteins; Total Amino Acids; Proline (mg/100g D. Wt.); Catalase Enzyme (H₂O₂/g F. Wt. Protein/min); Peroxidase and Superoxide dismutase (units mg⁻¹protein) Contents of Sweet Peppers (*Capsicum annum*, L.) Plant.

Plants were grown, under glasshouse conditions, for 60 days after germination and irrigated at 80% field capacity.

Nitrogenous components & Antioxidant enzymes	NaCl Salinity Concentrations(mM)					
	0.0	50	100	150	200	250
Ascorbic Acids (50 ppm ≈ 0.3 mM)						
Total Protein (mg/100g D. Wt.)						
(- ASC)	19.72±0.131	22.41±0.144	26.12±0.121	27.89±0.107	29.28±0.160	30.28±0.160
(+ASC)	36.48±0.067	47.19±0.185	50.17±0.172	54.24±0.110	56.39±0.175	59.39±0.175
F Values	*	*	*	**	**	**
Total Amino Acids (mg/100g D. Wt.)						
(- ASC)	17.97±0.086	18.72±0.079	19.83±0.124	20.52±0.090	20.76±0.289	21.56±0.289
(+ASC)	25.41±0.125	37.97±0.162	39.42±0.191	43.14±0.242	45.50±0.210	47.50±0.210
F Values	*	**	*	**	***	***
Proline (mg/100g D. Wt.)						
(- ASC)	10.13±0.98	12.91±1.01	14.38±1.19	15.19±0.81	17.89±0.38	19.89±0.38
(+ASC)	15.91±1.09	16.18±0.82	21.14±1.09	24.01±0.47	28.32±1.27	29.32±1.27
F Values	*	*	*	**	**	**
Catalase enzyme (H ₂ O ₂ /g F. W. Protein/min)						
(- ASC)	13.54±1.195	11.85±0.953	10.93±1.064	10.71±1.121	9.54±1.075	8.54±1.075
(+ASC)	17.02±0.922	20.76±0.879	22.98±1.107	23.16±1.032	25.28±1.246	26.28±1.246
F Values	*	*	*	**	**	**
Peroxidase (units mg ⁻¹ protein)						



(- ASC)	6.73±0.26	6.22±0.61	6.01±0.37	5.71±0.18	5.01±0.23	4.11±0.23
(+ASC)	9.02±0.47	10.13±0.52	11.38±0.51	12.13±0.22	13.70±0.53	13.70±0.53
F Values	*	*	*	*	*	*
Superoxide dismutase (units mg ⁻¹ protein)						
(- ASC)	2.68±0.28	2.81±0.52	2.62±0.09	2.35±0.19	2.18±0.28	2.07±0.28
(+ASC)	7.38±0.71	7.81±0.38	8.76±0.04	7.98±0.38	6.83±0.52	6.73±0.52
F Values	**	*	*	*	*	*

II- Inorganic Components

Mineral Elements:

Data presented in Table (8), indicated that the increasing NaCl salinity concentration tended to increase the inorganic mineral elements (potassium, Nitrogen, phosphorous, sodium, calcium, magnesium and chloride) contents compared with control sweet peppers plant. Whereas, mineral elements increased significantly more in the presence (+ASC) than in the absence (-ASC) of ascorbic acid. The deleterious effects of salinity on plant growth are associated with low water potential of the root medium which causes a water deficit within the plant; toxic effects of ions mainly Na⁺ and Cl⁻; nutritional imbalance caused by reduced nutrient (K⁺, Ca²⁺, Mg²⁺) uptake and/or transport to the shoot. Salinity mainly causes both hyper-osmotic stress and hyper-ionic toxic effects and the consequence can be plant demise (Munns and Termaat, 1986; Ashraf, 1994; Marschner, 1995; Serrano et al., 1999; Hasegawa et al., 2000).

Babu et al. (2012) found that the Potassium content was found in leaves and tomato fruits to be decreasing with increase in salt stress. Also, Khafagy et al. (2009) found that the significant decrease in K⁺ concentration occurred with increasing salinity levels in sweet pepper plants. In this respect, high salinity level (6000 ppm) NaCl (11.88 dSm⁻¹) was the most effective in this concern as compared to control. However, K⁺ concentration decreased in the root system more than shoot. Whereas, K⁺ concentration increased with application of ASC compared to non-salinized plants. In most cases, pre-soaking with ASC at 100 ppm was the most effective in increasing K⁺ concentration in the shoots and roots. Moreover, all interactions between salinity and AsA increased K⁺ concentration as compared to salinity stress.

Potassium may play a role on the synthesis of endogenous plant hormones (Haeder et al., 1981). Sucrose played a main role in the regulation of the root osmotic potential followed by K, glucose and Na this agree with the results (Eisa and Ali, 2003). However, exogenous application of ASC with varying levels (0, 50, 100 mg L⁻¹) in hydroponics also increased the accumulation of Na⁺ in the leaves of both wheat cultivars (Khan et al., 2006b). Despite its obvious importance, the low mobility of Ca²⁺ make the rates of its uptake and distribution limiting processes for many key plant functions. Furthermore, the general lack of recognition of the limiting role of Ca²⁺ is due in part to the fact that some important plant functions are controlled by changes in very small physiologically active pools of Ca²⁺ within the cytoplasm. As such, whole-leaf Ca²⁺ levels might not reflect any potential limitations (McLaughlin and Wimmer, 1999).

Ascorbic acid has effects on many physiological processes including the regulation of growth and metabolism of plants under saline conditions and increasing physiological availability of water and nutrient (Barakat, 2003). The accumulation of nitrogen-containing compatible solutes including proline is known to function in osmotic adjustment, protection of cellular macromolecules from damage by salts, storage of nitrogen and scavenging of free radicals. The results of the present study agree with Farahat et al. (2013) where found the highest nitrogen in shoot resulted from ascorbic acid (200 ppm) and salinity (3000 ppm) level.

In the present data the results finding from Farahat et al. (2013) agree with they reported that the all of nitrogen (N); phosphorus (P) and potassium (K) contents in both shoots and roots increased gradually with increasing the levels of ascorbic acid. So, ascorbic acid (ASC) protect metabolic processes against H₂O₂ and other toxic derivatives of oxygen affected many enzyme activities, minimize the damage caused by oxidative processes through synergistic function with other antioxidants and stabilize membranes (Shao et al., 2008). Also, Bassuony et al. (2008) found the content of K⁺; Ca²⁺ and Mg²⁺ in *Zea mays* plant decreased significantly under salinity stress, compared with control. While, application of 100 ppm from vitamins C (ascorbic acid) resulted significantly increases of K⁺, Ca²⁺ and Mg²⁺ contents compared with controls. While, Flores et al., (2001) found that the salt stress inhibits the uptake and transport of potassium, calcium and phosphorus, we predict that sodium chloride will inhibit growth in sweet peppers plants. Soltani

Nezhad et al. (2011) found that the phosphorus content in tomato (*Lycopersicon peruvianum* L) plant decreased was increased NaCl at 150 mM. Whereas, with increasing salinity levels, the phosphorus content of roots decreased in all NaCl concentrations. In contrast, by increasing of salt concentration in the culture medium, phosphorus content decreased significantly in roots compared to untreated plants.

Abd El-Aziz et al. (2007) found that the N, P and K contents in *Synгонium podophyllum*, (L) increased gradually by increasing the concentration of ASC to 100 ppm compared with the untreated plants. The increment in N concentration due to ASC treatments could be explained by the findings of Talaat (2003) who showed that accumulation of nitrate by ASC foliar application may be due to the positive effect of ASC on root growth which consequently increased nitrate absorption. In this context the increase in P concentration by thiamine and AsA treatments may be attributed to the postulation. So, the present results not consistent with Farouk (2011) he reported that potassium, magnesium and calcium contents decreased with increasing salinity levels up to 11 dsm⁻¹. While, the present results agreed with where he found application of antioxidants, especially ascorbic acid, significantly increased potassium, calcium and magnesium wheat flag



leaf. The application of antioxidants, especially ascorbic acid, partially reversed the negative effects of salinity in this respect. While, our current findings agree with Taffouo et al. (2010a & b) they found that the effect of NaCl salinity on tomato he showed that the salt treatments increased significantly Na⁺ contents in roots, stems and leaves of plants. While no agree with K⁺ and Ca²⁺ concentrations of plants were decreased in all tomato cultivars.

These results agree with Hussein et al. (2011) they reported that the interaction effect between ascorbic acid (ASC) and rates of salinity water on nutrients uptake. Under water salinity irrigation, increasing the ascorbic acid spraying rate from 0 to 200 ppm increased the uptake of essential nutrients N, P, K, Ca and Mg of wheat, but did not agree with where observed decreased the Na and Cl uptake so the ascorbic acid played an important role of decreasing effects of saline conditions.

Conclusion

Results of the current study showed the positive impacts of ascorbic acid (ASC) on growth of sweet peppers plant as well as salinity treatment than control. So as a simple and safe method, soaking the seeds of sweet peppers plant before planting can be used to improvement plant growth and water efficiency. It appears that utilization of ASC can led to improve quantity and quality of Sweet Peppers (*Capsicum annum*, L.) plant by accumulated the organic and inorganic components. It suggests that ASC could stimulate defense system for salt-stress. Generally, using ASC treatment could be a promising technique for agricultural improvements but extensive research is required on different crops.

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Table (8): Interactive effects of Ascorbic Acid (ASC) and NaCl Salinity on Inorganic Minerals (mg/100g D. Wt.) of Sweet Peppers (*Capsicum annum*, L.) Plant.

Plants were grown, under glasshouse conditions, for 60 days after germination and irrigated at 80% field capacity.

Mineral Elements (mg/100g D. Wt.)	NaCl Salinity Concentrations (mM)					
	0.0	50	100	150	200	250
Ascorbic Acids (50 ppm ≈ 0.3 mM)						
Potassium (K ⁺)						
(- ASC)	6.78±08	8.39±0.09	9.31±0.03	10.73±0.04	12.98±0.07	14.18±0.07
(+ASC)	9.21±0.11	13.81±0.13	14.83±0.09	17.20±0.08	19.13±0.06	22.13±0.06
F Values	*	**	*	**	**	**
Nitrogen (N ⁺³)						
(- ASC)	12.10±1.21	13.70±1.17	14.78±1.17	16.10±1.84	17.71±1.91	18.01±1.91
(+ASC)	13.80±1.41	17.30±1.81	19.99±1.91	20.33±0.91	24.01±2.30	27.21±2.30
F Values	N.S.	*	**	*	**	**
Phosphorous(P ⁺³)						
(- ASC)	5.78±0.08	7.57±0.05	8.92±0.07	9.81±0.09	10.91±0.01	12.41±0.01
(+ASC)	8.21±0.06	9.31±0.07	12.01±0.09	14.30±0.13	16.80±0.08	18.80±0.08
F Values	**	*	**	**	***	***
Sodium (Na ⁺)						
(- ASC)	71.38±3.81	77.18±1.17	82.98±2.13	86.13±3.61	87.17±4.75	91.17±4.75
(+ASC)	70.19±2.83	73.13±4.18	76.81±3.31	78.01±1.21	78.81±1.83	81.81±1.83
F Values	*	***	*	***	**	**
Calcium (Ca ⁺²)						
(- ASC)	19.05±1.01	20.31±1.18	27.13±1.01	30.10±1.01	32.31±1.06	34.31±1.06
(+ASC)	26.57±0.87	28.09±1.01	32.01±0.91	36.09±1.07	39.01±1.21	41.01±1.21



F Values	*	*	**	*	*	*
Magnesium (Mg ⁺²)						
(- ASC)	27.30±1.13	29.98±2.07	30.98±1.48	35.01±0.91	38.12±1.03	43.12±1.03
(+ASC)	38.10±2.01	44.89±2.01	48.01±1.61	52.01±1.24	57.18±1.81	59.98±1.81
F Values	**	**	**	**	**	**
Chloride (Cl ⁻)						
(- ASC)	21.98±2.01	30.21±2.01	31.33±1.03	33.13±1.27	33.88±1.08	35.12±1.08
(+ASC)	27.33±0.97	32.88±2.01	34.01±2.13	35.84±1.83	37.17±1.07	39.87±1.07
F Values	*	N.S.	*	**	**	**

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