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ASCORBIC AND SALICYLIC ACIDS AS WELL AS SEAWEED AND YEAST EXTRACTS ALTERED STRESS-RELATED METABOLITES AND ENHANCED YIELD AND ITS QUALITY OF SALT- STRESSED SOYBEAN (GLYCINE MAX [L.] MERRILL).

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ABSTRACT

The alleviation effects of ascorbic (AsA) and salicylic (SA) acids as well as seaweed (SWE) and yeast (YE) extracts against salinity stress on soybean plant were evaluated. Two experiments were conducted at the greenhouse and labs. of the Agric. Bot. Dept., Fac. of Agric., Mansoura Univ., and Seed Technology Research Unit, Field Crops Research Institute, Agric. Research Center, during the two growing Seasons 2010 and 2011. Salt stress was imposed by dissolving natural salt crust in tap water to obtain saline irrigation water at 5000,6000,7000,8000 and 9000 mgL^{-1}.

Results indicated that all tested substances partially restored the salinity induced decrease in seed yield, with AsA and YE were the most effective in this respect. Total phenols, proline and endogenous ascorbic acid contents were increased in response to either salinity stress , stress alleviators or their interaction. The ratio between Na and K in both shoots and roots was decreased in response to stress alleviators. In addition, seed oil and protein contents were increased in salt–stressed , stress alleviators- treated plants compared with salt-stressed only plants. In addition, results indicated that the alleviative effect of the applied stress mitigators was evident not only on salt-stressed plants but also on their descendant seeds where their germination, vigour, and seedling establishment were enhanced. It is concluded that AsA, SA, SWE and YE, preferably the first and the latter, could be used to maintain yield of soybean growing in salt-affected soils.

Keywords: soybean, *Glycine max*; salt- stress tolerance; ascorbic acid; salicylic acid ; seaweed extract; yeast extract.

INTRODUCTION

Soybean (*Glycine max* [L.] Merrill) is a legume species native to East Asia, widely grown for its edible seeds which has numerous uses. Soybeans produce significantly more protein per acre than most other legumes. Soybean protein is an ingredient in many meat and dairy analogues .The oil is used in many industrial applications. Seeds contain significant amounts of phytic acid , alpha-linolenic acid and isoflavones. Abiotic stress including salinity, drought, excess irrigation, high or low temperature, and heavy metals are common sources of stress which affects plant growth, development ,

structure , as well as physiological and biochemical processes. Salinity is one of the major stresses and can severely limit plant growth and productivity (Chernane *et al.*, 2015). In Egypt, salinity is a serious problem particularly in the Delta. Sixty percent of the cultivated lands of Northern Delta region are salt-affected, twenty percent of the Southern Delta and Middle Egyptian region and twenty five percent of the Upper Egypt region are salt-affected soils (Gehad, 2003).

Most of the crop species are glycophytes, and generally show limited growth and development due to salinity. Salt stress affects major plant processes such as photosynthesis, protein synthesis and lipid metabolism (Parvaiz and Satyawati, 2008). During salt stress, generation of reactive oxygen species (ROS) is accelerated, leading to oxidative stress. ROS interact with many cellular components, causing significant damage to membranes and other cellular structures and induce oxidative stress (Parvaiz and Satyawati 2008). In most studies concerning the effects of salinity on plants salinity stress is generated through the use of NaCl as the Salinization agent, which represent an artificial salinized medium. Authentic effects of more naturally saline medium resembling sea water and containing multiple cations and anions is less conducted and poorly-understood.

Plants have evolved a ROS-scavenging network, composed of enzymatic and non-enzymatic antioxidants for maintaining the levels of ROS under tight control. However, under stressful conditions, generation of ROS surpass the plant antioxidant system's ability to scavenge them. In such situation, external measures are needed to strengthen the defense mechanisms and maintain plant growth and productivity. Several compounds with different biochemical effects have been tried to mitigate stress effects on various plant species and mostly applied either as seed soaking , foliar spray or both combined. From these compounds AsA (Dehghan, *et al.*, 2011) , SA (Arfan, *et al.*, 2006; Arfan 2009 ; Kabiri *et al.*, 2014) , seaweed extract (Abdel Aziz *et al.*, 2011; Mansori *et al.*, 2014; Chernane *et al.*, 2015) , and YE (Hammad and Ali, 2014; Ibrahim, 2014), have been used to induce plant tolerance to various environmental stressors. However, Effects of these compounds on salt-stressed soybean is less understood. In most studies conducted to evaluate the effects of stress alleviators on plants, their effects were invariably evaluated through monitoring some features of F0 plants including their seed yield. As seeds are the antecedents to the F1 generation, their quality attributes that indicate the degree by which they give a healthy F1 generation are invariably ignored which represent a gap in the previous studies. Therefore, the present investigation aimed to study the effects of some stress modulators (SMs) on:

1. The metabolic adjustment of salt-stressed soybean (cv. Giza 111) that contribute to the plant's salt stress tolerance.
2. The establishment of the F1 generation via estimating certain quality parameters of F0's seed germination and seedling growth.

The hypothesis on which the study stands is that under salinity stress, these SMs will modulate salinity-induced responses towards acquiring salt stress tolerance.

MATERIALS AND METHODS

Experimental conditions

Experiments were carried out at the Agricultural Botany Dept., Fac. of Agric., Mansoura Univ. and Seed Technology Research Dept., Agric. Res. Center, Ministry of Agric., Egypt during 2010 and 2011 seasons. Seeds of Giza 111 cultivar were obtained from Seed Production Station's Central Administration, Dakahlia Governorate, Egypt. In the two growing seasons, rhizobium inoculated seed sowing was carried out on May, 15th in pots (40 cm inner diam.) containing 15 kg of air dried soil at the rate of 6 seeds/pot. Physical and chemical characteristics of the experimental soil are shown in Table (1). Thinning was made 20 days after sowing (DAS) to leave 4 uniform seedlings/ pot. The pots were arranged in a randomized complete block design. Each pot received calcium superphosphate (15.5% P_2O_5), at a rate of 150 kg/fed before sowing; nitrogen in the form of urea (46.5% N) at the rate of 50 kg/fed, divided into two equal doses and supplied at 20 and 30 DAS and potassium sulphate (48% k_2O) at the rate of 50 kg/fed, applied at the beginning of the flowering stage.

Six levels of salinity i.e : 1) Tap water as a control (320 mgL^{-1}), 2) 5000 mgL^{-1}, 3) 6000 mgL^{-1}, 4) 7000 mgL^{-1}, 5) 8000 mgL^{-1} and 6) 9000 mgL^{-1} were prepared by dissolving known weight of natural salt crust in tap water (artificial seawater).

Table (1)*: Physical** and chemical*** characteristics of the experimental soil

Sand	Silt	Clay	$CaCo_3$	pH	EC (dsm^{-1})	O.M.	N (ppm)	K (ppm)	P (ppm)	Na ($meqL^{-1}$)	Cl $meqL^{-1}$)
22.1	33	44	2.5	8.1	0.43	1.92	14.4	227	11.76	0.83	1.1

* **Values are the means of the analyses of the experimental soils of the two growing seasons; ** Mechanical analysis followed the pipette method using sodium hydroxide as a dispersing agent (Piper, 1950); *** chemical analyses were carried out according to Jackson (1967).**

Seeds were soaked for 3 hours in the solutions of the stress modulators (SMs) used before inoculation and sowing. In addition, the plants raised from soaked seeds were sprayed with the same SMs at two physiological stages (30 and 60 DAS). The used SMs treatments were 1) Tap water, as a control; 2) Ascorbic acid (AsA), 250 mgL^{-1}; 3) Salicylic acid (SA), 250 mgL^{-1}; 4) Seaweed extract (SWE), 1000 mgL^{-1} and 5) yeast extract (YE), 1000 mgL^{-1}. Each treatment had four replications. Components of YE are shown in Table (2) according to Mahmoued (2001).

Investigated parameters

Biochemical constituents

Samples were taken at 75 DAS to estimate the following biochemical parameters:

Total phenols:

The assay was based on the method of Toivonen and Stan, (2004). A sample of 10 mg was extracted in 1.2 M of HCl and 50 % Me-OH by heating at 80 ^{o}C for 3h. After centrifugation at 18000 g, 0.1 mL of

supernatant was mixed with 0.1 mL of Folin- ciocalteu reagent and 0.5 mL of 20 % Na_2 CO_3 and allowed to stand in the dark for 15 min. Absorbance was measured at 725 nm with gallic acid as a standard and the total phenolic content was calculated as gallic acid equivalent.

Table (2): Chemical analysis of yeast extract

Amino acid mg/100g dry weight		**Vitamins and Carbohydrates mg/100g dry weight**	
Arginine	1.99	Vit.B1	2.23
Histidine	2.63	Vit.B2	1.33
Isoleucine	2.31	Vit.B6	1.25
leucine	3.09	Vit.12	0.15
Lysine	2.95	Thimain	2.71
Methionine	0.72	Riboflavin	4.96
Phenyl alanine	2.01	Inositol	0.26
Threonine	2.09	Biotin	0.09
Tryptophan	0.45	Nicotinic acid	39.88
Valine	2.19	Panthothenic acid	19.56
Glutamic acid	2.00	P amino benzoic acid	9.23
Serine	1.59	Folic acid	4.36
Aspartic acid	1.33	Pyridoxine	2.90
Cystine	0.23	Total carbohydrates	23.20
Proline	1.53		
Tyrosine	1.49	Glucose	13.33

Proline content: according to the method of Bates, *et al.* (1973), approx. 300 mg of dry leaf tissues were homogenized in 10 mL of 3% (w/v) aqueous sulphosalicylic acid and filtered. To 2 mL of the filtrate, 2 mL of acid ninhydrin was added, followed by the addition of 2 mL of glacial acetic acid and boiling for 60 min. The mixture was extracted with toluene and free proline was quantified spectrophotometrically at 520 nm from the organic phase.

Nitrogen content: The improved Kjeldahl method of A.O.A.C. (1980) was employed.

Dry seed sample of 0.5 g weight was digested with sulphuric acid and hydrogen peroxide mixture. Distribution was carried out with 40% NaOH and ammonium was received in 4% boric acid solution. The distributes were then titrated with 0.02 N HCl using the mixed methyl red-bromocresol green indicator. Nitrogen concentration was calculated as percentage on the dry matter basis

Seed protein:

Protein percent was calculated by multiplying the percentage of total nitrogen by the factor of 6.25.

Oil seed content:

A samples of 5 g of dried seeds was extracted with Petroleum ether using Soxholet's apparatus according to the A.O.A.C. (1980) method.

Total ascorbic acid (AsA):

As A content was determined using the 2,6 dichlorophenol indophenol method as described by Ranganna (1979).

Potasium and sodium contents

Na and K percent in shoots and roots of soybean plant was estimated Flamephotometrically according to Peterburgski (1968) using Jenway Flamephotometer.

F0's seed quality parameters

For assessing quality of the seeds resulting from the pot experiment, a random sample of 400 seeds per treatment were sown on filter paper in sterilized Petri-dishes (14-cm diameter), 25 seeds/dish. Seed quality tests were done according the rules of International Seed Testing Association (ISTA,1985)

Statistical Analysis:

Data were subjected to ANOVA statistical analysis as described by Gomez and Gomez (1984). Treatment means were compared using the least significant difference (L.S.D.) at 0.05 level of probability. Similar results were recorded during the two growing seasons of the experiment so, only the data of the first season were presented.

RESULTS

Proline, total phenols and AsA contents:

Salinity levels above 5000 mgL^{-1} and all tested SMs as well as their interactions increased proline and phenols (Table 3) as well as ASA contents (Table 4). The highest increases were recorded in AsA-treated plants stressed with salinity at 9000 mgL^{-1}

Table (3): Effect of salinity stress levels and SMs as well as their interactions on proline (mg/g D.Wt) and total phenols (mg /100 g F.Wt) contents in shoots soybean plants.

Treatments / SMs (B)	Salinity levels (mg/l) (A)													
	320	5000	6000	7000	8000	9000	Mean	320	5000	6000	7000	8000	9000	Mean
	Proline content							Total phenols content						
Tap water	3.62	6.28	6.56	6.82	7.12	7.16	6.26	43.2	61.4	66.6	72.8	78.0	80.0	67.0
AsA	5.72	7.31	7.41	7.57	8.15	9.32	7.58	69.6	85.4	86.3	87.3	88.5	89.3	84.4
SA	4.68	6.42	6.75	6.92	7.20	7.93	6.65	47.3	71.0	73.6	77.5	80.6	82.6	72.1
SWE	4.85	6.64	6.90	7.10	7.32	8.35	6.86	56.8	79.6	81.3	82.5	83.9	85.7	78.3
YE	5.63	7.22	7.32	7.47	7.94	8.46	7.34	63.9	81.5	82.7	83.6	84.5	86.2	80.4
Mean	4.90	6.77	6.99	7.18	7.55	8.24		56.2	75.8	78.1	80.7	83.1	84.8	
LSD at 5%	A: 0.6		B: 0.8		AXB: 1.2			A: 2.4		B: 2.1		AXB: 3.3		

Table (4): Effect of salinity stress levels and SMs as well as their interactions on AsA content (mg /100 g F.Wt) in shoots of soybean plants.

Treatments / SMs (B) Antioxidant(B)	Salinity levels (mg/l) (A)						
	320	5000	6000	7000	8000	9000	Mean
Tap water	110	151	158	167	178	184	158
AsA	146	213	٢٢٢	257	298	310	241
SA	121	١٩١	٢٠٠	229	267	276	214
SWE	١٣٨	١٩٩	٢٠٨	٢٤٦	٢٨٦	٢٩١	٢٢٨
YE	١٤١	206	214	254	290	299	234
Mean	131	192	200	231	264	272	
LSD at 5%	A: 16		B: 12		AXB: 28		

Na and K contents as well as Na/K ratio:

All salinity levels increased Na while decreased K in both shoots and roots of soybean plants (Fig 1). On the other hand, all applied SMs generally increased K content whereas decreased Na content in both shoot and roots of soybean plant. Accordingly, Na/K ratio was increased due to salinity levels whereas decreased due to SMs treatments. Treatment with AsA was the most efficient treatment for decreasing Na/K ratio followed by the treatment with YE.

Yield and yield components

Salinity levels decreased soybean yield estimated as g plant^{-1}, and 9000 mg /l was the most damaging level in this respect (Table 5). In addition, seed yield of SMs-treated, salinity-stressed plants was significantly higher compared with untreated plants, though it is still below yield of control plants. So, applied SMs partially counteracted the harmful effect of salinity stress on seed yield of soybean plant.

Table (5): Effect of salinity stress levels and applied SMs as well as their interactions on seed yield (g plant $^{-1}$) of soybean plants

Treatments / SMs (B)	Salinity levels (mg/l) (A)						
	320	5000	6000	7000	8000	9000	Mean
Tap water	13.1	7.6	6.7	6.3	4.6	3.7	7.0
ASA	21.6	11.4	10.3	9.8	9.5	8.8	11.9
SA	14.6	9.4	9.1	8.9	8.3	7.9	9.7
SWE	١٦.٥	٩.٩	٩.٤	٩.١	٨.٦	٨.٣	١٠.٣
YE	١٩.٨	10.8	9.9	9.5	9.3	8.5	11.3
Mean	17.1	9.8	9.1	8.7	8.1	7.4	
LSD at 5%	A: 1.2 B: 0.7 AXB: 3.0						

Oil and protein in seeds :

Salinity levels decreased while applied SMs increased both oil (Table 6) and protein (Table 7) percentage in soybean seeds. The highest

protein percent was obtained in response to AsA treatment followed by YE. Moreover, SMs counteracted the damaging effects of salinity on oil as well as protein percent. The most prominent counteracting effect was due to AsA. Treatment with YE ranked the second in this context.

Table (6): Effect of salinity stress levels and applied SMs as well as their interactions on oil percent in seeds of soybean plants

Treatments / SMs (B)	Salinity levels (mg/l) (A)						
	320	5000	6000	7000	8000	9000	Mean
Tap water	17.83	١٣.١٦	١٢.٨٦	١١.٦٤	١٠.٤٥	١٠.٤٠	١٢.٧١
ASA	١٨.٩٦	١٧.٦٠	١٧.٤٢	١٧.١٠	١٦.٨٨	١٥.٨٠	١٧.٢٩
SA	١٨.٣٢	١٦.٧٨	١٦.٢٠	١٥.٩٦	١٤.٧٢	١٤.٣٥	١٦.٠٦
SWE	١٨.٤٥	١٧.٣٢	١٦.٧٣	١٦.٥٦	١٥.٦٤	١٤.٤٣	١٦.٥٢
YE	١٨.٦٢	١٧.٥٠	١٦.٩٤	١٦.٧٠	١٥.٨٧	١٤.٦٠	١٦.٧١
Mean	١٨.٤٤	١٦.٤٧	١٦.٠٣	١٥.٥٩	١٤.٧١	١٣.٩٢	
LSD at 5%	A: 1.44 B: 0.92 AXB: 2.32						

Table (7): Effect of salinity stress levels and applied SMs as well as their interactions on protein percent in seeds of soybean plants

Treatments / SMs (B)	Salinity levels (mg/l) (A)						
	320	5000	6000	7000	8000	9000	Mean
Tap water	37.4	33.9	33.3	32.8	31.6	30.2	33.2
ASA	39.8	36.3	35.8	35.6	35.3	35.0	36.3
SA	38.2	35.5	35.2	34.9	34.5	34.1	35.4
SWE	٣٩.٠	٣٥.٩	٣٥.٦	٣٥.٢	٣٤.٨	٣٤.٣	٣٥.٨
YE	٣٩.٤	36.0	35.7	35.4	35.0	34.5	36.0
Mean	38.8	35.5	35.1	34.8	34.2	33.6	
LSD at 5%	A: 2.9 B: 1.7 AXB: 5.3						

F0 seed's quality parameters

Salinity stress at all levels decreased whereas SMs increased all soybean seeds quality parameters tested; germination percentage (Table 8), speed of germination (Table 9), seedling dry weight (Table 10), and seedling vigor index (Table 11). The interaction effects between salinity and SMs on seed's quality parameters were significant, where all applied SMs enhanced all seeds quality parameters of plants grown under salinity stress compared with untreated plants hence, used SMs partially counteracted the harmful effect of salinity stress on seed quality parameters of soybean.

Fig (1): Na and K Contents as well as Na/K ratio in shoots and roots of soybean plants as affected by Salinity stress levels and applied SMs.

Table (8): Effect of salinity stress levels and applied SMs as well as their interactions on germination percentage of F0's soybean seeds.

Treatments / SMs (B)	Salinity levels (mg/l) (A)						
	320	5000	6000	7000	8000	9000	Mean
Tap water	80	41	39	35	31	29	42.5
ASA	٨٩	78	75	74	72	70	76.3
SA	٨١	52	50	47	44	43	52.8
SWE	٨٤	٧٥	٧٢	٧٠	٦٨	٦٥	٧٢.٣
YE	٨٦	76	73	71	70	67	73.8
Mean	84.0	64.4	61.8	59.4	57.0	54.8	
LSD at 5%	A: 5 B: 3 AXB: 8						

Table (9): Effect of salinity stress levels and applied SMs as well as their interactions on speed of germination of F0's soybean seeds.

Treatments / SMs (B)	Salinity levels (mg/l) (A)						
	320	5000	6000	7000	8000	9000	Mean
Tap water	40.6	23.6	22.4	21.6	18.7	15.9	23.8
ASA	٤٨.٥	39.5	37.6	36.2	35.2	32.2	38.2
SA	٤٣.٠	32.3	31.2	28.2	25.4	23.6	30.6
SWE	٤٥.٥	٣٦.٢	٣٤.٢	٣١.٣	٢٨.٠	٢٤.٦	٣٣.٣
YE	٤٧.٢	37.7	35.6	33.4	30.6	27.9	35.4
Mean	٤٥.٠	33.9	32.2	30.1	27.6	24.8	
LSD at 5%	Salinity: 5.6 Antioxidant: 3.2 Interaction: 7.8						

Table (10): Effect of salinity stress levels and applied SMs as well as their interactions on dry weight of 10 seedlings (g) resulting from germination of F0's soybean seeds.

Treatments / SMs (B)	Salinity levels (mg/l) (A)						
	320	5000	6000	7000	8000	9000	Mean
Tap water	0.338	0.231	0.226	0.218	0.189	0.170	0.228
ASA	٠.٥٢٤	0.328	0.316	0.292	0.273	0.241	0.329
SA	0.361	0.290	0.283	0.266	0.250	0.224	0.279
SWE	٠.٣٨٢	٠.٣١٠	٠.٢٩٠	٠.٢٨٠	٠.٢٦٢	٠.٢٣٤	٠.٢٩٣
YE	٠.٤٦٨	0.321	0.304	0.288	0.270	0.239	0.315
Mean	0.415	0.296	0.284	0.269	0.249	0.222	
LSD at 5%	A: 0.039 B: 0.026 AXB: 0.057						

Table (11): Effect of salinity stress levels and applied SMs as well as their interactions on seedling vigor index of seedlings resulting from germination of F0's soybean seeds.

Treatments / SMs (B)	Salinity levels (mg/l) (A)						
	320	5000	6000	7000	8000	9000	Mean
Tap water	٢٧.٠	٩.٥	٨.٨	٧.٦	٥.٨	٤.٩	١٠.٦
ASA	٤٦.٦	٢٢.٣	٢٣.٧	٢١.٦	١٩.٧	١٦.٨	٢٥.١
SA	٢٩.٢	١٥.١	١٤.٢	١٢.٥	١١.٠	٩.٦	١٥.٣
SWE	٣٢.١	٢٣.٢	٢٠.٨	١٩.٦	١٧.٨	١٥.٢	٢١.٥
YE	٤٠.٢	٢٤.٤	٢٢.٢	٢٠.٤	١٨.٩	١٦.٠	٢٣.٧
Mean	٣٥.١	١٨.٩	١٧.٩	١٦.٤	١٤.٧	١٢.٥	
LSD at 5%	A: 2.7 B: 1.5 AXB: 5.1						

DISCUSSION

Salinity stress responses

Results of the current study indicated that salinity stress not only reduced soybean yield and altered the plant biochemical constituents of F0 plants, but also negatively affected germinability of its seeds as well as establishment and development of the F1 generation; indicating a resulting stress on soybean plants. Reduction of yield is the most significant aspect of salt stress in agriculture, to the extent that relative yield under salinity stress conditions was considered as an index for the plant's salinity tolerance (Hasanuzzaman *et al.*, 2013). Yield and its components was significantly reduced in salt-stressed *Vigna radiata* (Nahar and Hasanuzzaman, 2009). Salinity-induced reduction in plant yield may be due to a reduction in photosynthetically-active green foliage (Wahid *et al.*, 1997). Reduced pollen viability and consequently seed set under salinity stress conditions may also contribute to salinity-induced reduction in crop yield (Abdullah *et al.*, 2001).

Within salinity-induced metabolic alternations recorded in the present investigation, noteworthy was its effect on Na and K contents as well as the ratio between them in the plant organs (Fig 1) which may be considered a principal component of the stress imposed on soybean plants by salinity. Increasing Na at the expense of K has a dual effect of Na toxicity and malfunctions of various processes due to K deficiency. Na displaces K on binding sites essential for cellular functions (Munns 2002). Na-induced K deficiency interferes with stomatal functions which ultimately lead to water loss. Na also competes with NH_4 whereas Cl competes with NO_3 which ultimately lead to reduced N uptake which may account for the reduced seed's protein content recorded in the present investigation. On the other hand, reduced seed oil content may be due a reduction in photosynthetic activities induced by a reduction in either carbon metabolism or photophosphorylation under salinity (Sudhir and Murthy, 2004).

The accumulation of phenols, proline and ascorbic acid due to salinity was recorded in the present study. Induction of phenols (Chernane et al., 2015), proline (Abdel Aziz *et al.*, 2011; Farouk, 2011), and ascorbic acid

(Agarwal and Shaheen , 2007) was reported in plants exposed to salt stress. Several functions are proposed for the accumulation of proline in tissues submitted to salt stress: 1) osmotic adjustment, 2) detoxification of excess ammonia, 3) stabilization of proteins and/or membranes, 4) scavenger of free radicals and 5) stability of some cytoplasmic and mitochondrial enzymes (Ozdemir *et al.* 2004). Phenolic compounds retard or inhibit lipid autoxidation by acting as radical scavengers (Namiki, 1990) and, consequently, are essential antioxidants that protect against propagation of the oxidative chain. Ascorbic acid reacts with a range of ROS such as $^1O_2$, $O_2^{\bullet-}$, $HO^{\bullet}$ and H_2O_2 , which is the basis of its antioxidant action (Shigeoka *et al.*, 2002 ; Foyer, 2004) .

Effects of SM on alleviating salinity stress:

Results of the present investigation indicated that all SMs tested altered the metabolism of salt-stressed plants by elevating their contents from AsA, proline and phenols and maintained the Na/K ratio that has been disturbed due to salinity stress. These SMs effects mitigated salinity stress on plants hence, maintained their yield as well as its quality and contributed to a more vigorous F1 generation, substantiating the hypothesis on which the study is founded. Utilization of AsA, SA. SWE and YE as stress mitigators and the mode of action of their alleviative effects will be discussed as follows:

Effects of AsA

Utilization of AsA as a stress mitigator is well-established. It has been recorded that endogenous AsA content increases under salinity stress conditions (Agarwal and Shaheen, 2007). Accordingly, AsA was applied exogenously to alleviate drought stress on *Hibiscus esculentus* (Amin *et al.*, 2009) and to counteract salinity stress effects on *Glycine max* (Dehghan *et al.*, 2011). It functions as a reductant for many free radicals, thereby minimizing the damage caused by oxidative stress. AsA is an important antioxidant which reacts with a range of ROS such as $^1O_2$, $O_2^{\bullet-}$, $HO^{\bullet}$ and H_2O_2, constituting the basis of its antioxidant action (Shigeoka *et al.*, 2002 ; Foyer, 2004). On the other hand, AsA has been implicated in several types of biological activities in plants; 1) as an enzyme co-factor, 2) as an antioxidant and 3) as a donor/acceptor in electron transport at the plasma membrane or in the chloroplasts, all of which are related to oxidative stress resistance (Conklin, 2001). An essential route for ROS-scavenging by AsA occurrs near PSI, thereby minimizing the risk of escape and reaction of ROS with each other (Foyer and Noctor, 2000). Evidences from current study supported from relevant literature indicate that the alleviative effects of AsA against salt stress is based on enhanced antioxidant capacity (Athar *et al.,* 2008), especially its enzymatic component by elevating the activities of CAT and POD (Munir and Aftap, 2011) as well as CAT, POD and SOD (Dehghan *et al.*, 2011), preserving chlorophyll biosynthesis and marinating photosynthetic activities (Hamada and Al-Hakimi, 2009) and accumulation of compatible solutes including proline (Azzedine *et al.*, 2011).

Effects of SA

Previous studies revealed a significant role of SA in plant stress responses. Exogenous application of SA through rooting medium modulates

ion accumulation and antioxidant activity in salt-stressed spring wheat, conferring salinity tolerance (Arfan, 2009). In addition, supplementation of SA counteracted drought stress effects on *Nigella sativa* (Kabiri *et al.*, 2014). Stress alleviative effects of SA is mediated through enhancing both components of the plant's antioxidant system; the enzymatic component (El-Tayeb, 2005; Arfan, 2009) as well as the nonenzymatic component (Srivastava and Dwivedi, 1998) and accumulation of compatible osmolytes (Kabiri *et al.*, 2014), especially proline (Arfan, 2009). The accumulation of inorganic or organic osmolytes makes the surplus of water uptake possible as evidenced by increased relative water contents in tissues of SA-pretreated plants (Szepesi *et al.* 2005). Improved photosynthetic performance of plants under stress conditions due to SA application (Ananieva *et al.* 2004) is another element of its stress-mitigation effect, which suggest that SA-pretreatment may improve the gross rate of carbon assimilation during osmotic stress. SA-dependent hormonal regulation during salt stress conditions manifested by maintaining IAA and cytokinin contents, that otherwise diminish under stress, and thus reduce stress-induced inhibition of plant growth as well as preserving a high ABA level providing the development of anti-stress reactions (Shakirova *et al.* 2003).

Effects of SWE

In accordance with the results of the present investigation, SWE of *Ulva rigida* enhanced salt stress tolerance of wheat via activation of the enzymatic antioxidant system (SOD, CAT and APX) as well as accumulation of total phenolics, leading to enhanced growth of salt-affected, SWE-treated plants (Chernane *et al.*, 2015). In addition, extracts of two seaweeds, *Fucus spiralis* and *Ulva rigida* enhanced drought stress tolerance of *Phaseolus vulgaris* through modulating the plant's antioxidant system and the contents of some key plant osmoprotectants (Mansori *et al.*, 2014). SWE has been assigned an immune-modulating properties due to its constituents from antioxidants, vitamins, carotenoids, polysaccharides, phycobiliproteins (Abd El-Baky et al., 2008).

SWE can alleviate the harmful effect of salinity stress through its effects on: 1) activation of root cells and their cytokinins biosynthetic capacity (Schmidt, 2005), 2) enhancing leaf water status (Demir *et al.,* 2004), 3) altering hormonal balances and favor cytokinins and auxins production (Schmidt, 2005), 4) enhancement of antioxidant enzymes (Schmidt, 2005), 5) stimulating biosynthesis of tocopherols, ascorbic acid and carotenoids which protect PSII photosynthetic apparatus (Zhang and Schmidt, 2000) and 6) Stimulation of chloroplast development, enhancing phloem loading and delay senescence (Demir *et al.,* 2004).

Effects of YE

YE Extract is a biostimulant that had been used to sustain plant growth and yield in stressful conditions. It has been applied to enhance drought tolerance in *Phaseolus vulgaris* (Ibrahim, 2014) and wheat (Hammad and Ali, 2014). Stress alleviation effect of YE may be ascribed to its biostimulants, being a rich source of phytohormones (especially cytokinins), vitamins, enzymes, amino acids and minerals (Khedr and Farid, 2000 ; Mahmoud,

2001). Its cytokinins content have specially been implicated for its beneficial role during stress (Barnett, *et al.,* 1990).

CONCLUSIONS

Ascorbic acid, Salicylic acid, seaweed extract and yeast extract partially restored salinity-induced decrease in seed yield of salt-stressed soybean plants, with the first and the latter were the most effective in this respect. Results indicated that the alleviative effect of the these tested stress modulators was based on elevating the levels of endogenous ascorbic acid , total phenols and proline whereas decreasing the ratio between Na and K in salt-stressed plants. In addition, results indicated that the alleviative effect of the applied stress mitigators was evident not only on salt-stressed plants but also on their descendant seeds where their germination, vigour, and seedling establishment were enhanced

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تعديل نواتج التمثيل المرتبطة بالإجهاد وتحسين المحصول وصفاته فى نباتات فول الصويا المعرضة للإجهاد الملحى بإستخدام أحماض الأسكوربيك والسالسيلك ومستخلصات أعشاب البحر والخميرة

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أجرى البحث بهدف تقليل الأثر الضار للملوحة على نبات فول الصويا بإستخدام بعض مضادات الأكسدة حيث تم نقع البذور فى مضادات الأكسدة المختلفة لمدة ثلاث ساعات قبل الزراعة وتم رش تلك النباتات بعد نموها بنفس مضادات الأكسدة على فترتين ٣٠، ٦٠ يوم بعد الزراعة . وتم تقدير المحتويات الكيماوية فى المجموع الخضرى فى عمر ٧٥ يوم من الزراعة .

وكانت أهم النتائج المتحصل عليها أن مضادات الأكسدة المستخدمة يمكنها التغلب جزئيا على الآثار الضارة الناجمة عن مستويات الإجهاد الملحى المرتفع على المحصول وكان الأكثر تأثيرا فى هذا الشأن حمض الأسكوربيك ومستخلص الخميرة ، كما أدت مستويات الإجهاد الملحى والمواد المضادة للأكسدة والتفاعل بينهما إلى زيادة واضحة فى محتوى كلا من الفينولات والبرولين والأسكوربيك ، كما أدت معاملات مضادات الأكسدة لنقص واضح فى معدل الصوديوم إلى البوتاسيوم بينما لوحظ زيادة واضحة لمعدل الصوديوم إلى البوتاسيوم تحت مستويات الإجهاد الملحى فى كلا من المجموع الخضرى والجذرى لنباتات فول الصويا ، كما أدى التفاعل بين مضادات الأكسدة ومستويات الإجهاد الملحى لزيادة نسبة الزيت والبروتين مقارنة بالنباتات تحت مستويات الإجهاد الملحى ولكن ظلت هذه النتائج أقل من الكنترول .