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MoringaStenopetala Leaves Extract Improve Antioxidant Defense Abilities and Salt Tolerance of Wheat Plant Irrigated with Seawater

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ABSTRACT

Effect of irrigation bread wheat plants (*Triticumaestivum L.*, cv. Giza 168) with sea water (10 and 20% v/v), spraying with *Moringastenopetala* leaves extract (5 gL^{-1} dry weight in 0.1% Tween solution) cultivated under normal and stress conditions were studied. Plant bioregulators (Oxalic acid at 200 ppm) at the vegetative growth stage on photosynthetic pigments, antioxidant components, activity of some antioxidant enzymes, lipid peroxidation products, growth parameters, mineral content and economic yield were estimated. Irrigation of wheat plants with sea water led to an increase in Na^+ ion, activities of antioxidant enzymes, superoxide dismutase, ascorbate peroxidase and total peroxidase and TBARs components. In contrast, the contents of photosynthetic pigments and yield components were reduced. Furthermore, the overall growth of wheat plants was interrupted by irrigation with sea water (10 and 20%) and the effect was pronounced at higher level (20%). Application of Oxalic acid had a slight effect on plant growth, antioxidant behavior and activity of antioxidant enzymes in plants irrigation with sea water compared with that in stressed wheat plants. Application of algal extracts significantly increased the contents of total chlorophyll and antioxidant phenomenon. In additional, application of *Moringastenopetala* leaves extract exhibited strong positive correlation with increase in fresh weight (FW), grain weight and yield components. It is concluded that productive purpose of wheat crop by mean of brackish water (at 20 v/v level) is possible under a level of economical value through its application of *Moringastenopetala* extracts.

Keyword:*Moringastenopetala***,** wheat plants, antioxidant enzymesand irrigated with seawater

INTRODUCTION

Maringa stenopetala is commonly known Moringa species that are grown in the tropics and sub-tropics. It is reported that the edible parts of Moringa tree are exceptionally nutritious Melesse*et al.,* (2012). All parts of the tree except the wood are edible, providing a

*Moringa*species are one of the most useful trees in the tropics and subtropics of Asia and Africa, with a multiple of uses. These species are the most widely cultivated of the Moringaceae family and was utilized by the ancient Egyptians. Traditionally, almost all parts of Moringa flower, fruits, leaves and roots are edible, and have long been consumed as vegetable and used to treat many diseases (Rahman *et al.,* 2009). The leaves are highly nutritious, which contain more vitamin A than carrots, more calcium than milk, more iron than spinach, more vitamin C than oranges and more potassium than bananas and more protein than milk and eggs (Sreelatha*et al*., 2011). Moreover, leaves of Moringa species are rich in various phytochemicals like carotenoids, amino acids, sterols, glycosides, alkaloids, flavonoids, moringine, moringinine, phytoestrogens caffeoylquinic acids and phenolics (Anwar *et al.,* 2007). Fruits and seeds have been reported as a rich source of protein, essential elements (Ca, Mg, K and Fe) and vitamins (vitamin A, C and E). A recent study conducted by Melesse and Berihun (2013) indicated that the leaves of M. stenopetala are rich in protein (28.2%) and contain reasonable amounts of essential amino acids of which some are comparable with those found in soybean meal. In stomata closure, osmotic stress leads to leak in CO_2 availability for photosynthetic carbon assimilation, thereby causing high accumulation of superoxide in chloroplast which can cause photo inhibition and photo-oxidation damages (Ashraf *et al.,* 2008). Under pathogens, drought and salinity stress condition; ROS are generated through pathways such as photorespiration, mitochondrial respiration and from the photosynthetic apparatus (Pei *et al.,* 2000). However, under these conditions, the cellular electron transport within the different subcellular compartments is impairs and lead to generation of ROS compounds (Ali and Alqurainy, 2006). Therefore, ROS could be considered as cellular biomarker for stresses and as secondary messenger in the stress response signaling pathways. However, plants have the high ability to scavenge ROS radicals by producing two types of antioxidants action includes: Enzymatic and non-enzymatic systems (Abd El Baky*et al.,* 2010; Gupta *et al.,* 1999). However, many studies indicated that some natural compounds might play an important role in enhancing plant tolerance to some abiotic stresses such as salt, drought and extreme temperatures (Ashraf, 2010; Abd El Baky*et al*.; 2010; Abd El Baky*et al*., 2014).

However, a positive association between the high accumulation of antioxidants and degree of salt tolerance has been drawn in different plant species. As examples, guaiacol Peroxidase (POX), SOD, POX and CAT enzyme activities play a significant role in the protection and recovery of several plants against oxidative stress induced by salt stress (Ashraf, 2010; Turhan*et al.,* 2008). Moreover, plants contain a variety of nonenzymatic molecules (ascorbic acid, tocopherols, carotenoids, flavonoids, glutathione) which play a substantial role in counteracting oxidative stress caused by stresses (Tausz*et al* ., 2000; Schafer *et al*., 2002). Abd El Baky*et al*., (2010), reported that microalgae could be enhanced plant salt tolerance through increasing the production of non-antioxidant compounds and elevate the activities of antioxidant enzyme system. Since, evaluation of functioning of all natural components in salt tolerance including that of oxidative stress tolerance is necessary. Therefore, this study was aimed to understand the impact of treated wheat plants irrigated with 10 and 20% seawater with moringa extracts as bio regulator to enhance salt tolerance.

MATERIALS AND METHODS

Plant Source:*MoringaStenopetala* Tree Cultivated In Egypt

Preparation of *MoringaStenopetala*Leaves Extracts and Oxalic Acis

Maringa stenopetala leaves (5 g) were homogenized in deionized water (100 ml) containing 1% Tween-20, and the volume was competed to one liter with deionized water (5 g leaves L^{-1} in 0.1% Tween-20 solution). Oxalic acid at 200 ppm in 0.1% Tween-20 solutions. The *Moringastenopetala* leaves extract and oxalic acid were applied as a spray at the vegetative stage of wheat plants (40-days-old).

Wheat Grains

The grains of wheat cultivar named Giza 94 were obtained from Wheat Department, Agriculture Research Centre, Ministry of Agriculture, and Giza, Egypt. The wheat grains were surface sterilized in 0.1% $HgCl_2$ solution for 3 min, washed thoroughly with distilled water before cultivation.

Wheat Cultivation

A pot experiment was conducted in the greenhouse of National Research Centre, Dokki, Giza, Egypt, during the winter 2010/2011 in order to evaluate the effect of spraying of *Maringa stenopetala* leaves extract as source of Amino acid and of antioxidant compounds to enhance salinity tolerance of wheat plants irrigated with 10 and 20% (v/v) sea water and tap water as a control. The experiment included 12 treatments and repeated three times. The experimental was conducted in plastic pots (40 cm in diameter) filled with 20 kg soil (sand and clay 2:1 v/v), each one contained forty grains and irrigated with tap water as required. Also, the plants were received the optimum fertilizer levels (PO^{-4} , as super-phosphate; and N as ammonium nitrate) as recommended by Ministry of Agriculture in Egypt. After 40 days from cultivation (vegetation stage), the plants were sprayed (first treatment) with algal extracts (5g L^{-1} dry weight in 0.1% Tween solution) and oxalic acid (200 mg L^{-1}). Then, plants of each set were divided into three groups, which irrigated with 0, 10 and 20% sea water, respectively. Then, the plants were irrigated with sea water and tap water alternatively every 7 days until the end of experiments. After 14 days the second treatment with algal extracts and bio regulators were done. Samples from each treatment at 70 days-old were uprooted and washed with distilled water, dried with filter paper and used for chemical analysis.

Estimation of Growth and Grain Yield

Nine plants from each treatment (three plants per replicate) were collected and immediately rinsed with iso-osmotic solution, blotted on filter paper and weighed to obtain the fresh weight (FW). For determination of dry weight (DW) the plant parts from each treatment dried to constant weights at 65^{o}C. Leaves and stems were separated and weighed. After harvest, the weight of 100 grains and grain yields were calculated.

Extraction and Estimation of Chlorophyll

Wheat leaves (1 g, FW) dried on filter paper were ground in acetone (5 ml, 80 %) and allowed to stand overnight in dark at 4° C followed by centrifugation at 10,000 × g for 5 min. The contents of total chlorophyll (T-Chl), chlorophyll a (Chl-a) and chlorophyll b (Chl-b) in the supernatant were determined according to Lichtenthaler (1987).

Determination of Carotenoid Contents

The contents of total phycocyanin and carotenoids were spectrophotometrically determined by the method of Lichtenthaler (1987).

Determination of Total Tocopherols and Ascorbic Acid

These substances were spectrophotometrically determined as outlined by AOAC (1995) and Augustin *et al*. (1985), respectively.

Determination of Total Phenolic Content

Total phenolic content (TPC) was estimated as gallic acid equivalent using the Folin–Ciocalteu method (Li *et al.*, 2007).

Extraction of Cytosolic Fraction

A plant material (ca. 2g) was excised and homogenized in 10 ml of ice-cold grindingbuffer containing 0.4 M sucrose and 25 mMTris (pH 7.2). The homogenate was passed through 4 layers of cheat cloth and centrifuged at 12,000 × g for 15 min at 4°C. Theresulting supernatant was used for determination ofenzyme activities, lipid oxidation products and protein contents.

Enzyme Assays

The activities of wheat cytosolic superoxide dismutase (SOD; EC:1.15.1.1) was determined as described by Chance and Maehly (1955). The activity of ascorbate peroxidase (APX), (EC: 1.11.1.11) was assayed according to Nakano and Asada (1981). The activity of each enzyme was expressed on protein basis.

Determination of Lipid Peroxidation Products

The lipid peroxidation products in wheat cytosolic fraction were estimated by the formation of thiobarbaturic acid reactive substances (TBARS) and quantified in term of malonald-hyde (MDA) as described by Haraguchi*et al.*, (1997). The lipid peroxidation was expressed as micromoles of MDA calculated using the extinction coefficient of 1.56×10^5 mM^{-1} cm^{-1}.

Determination of Protein

The total protein content in wheat cytosolic fraction was determined at 595 nm, using Comassein blue G 250 as mentioned by Bradford (1979). Bovine serum albumin (BSA) was used as a protein standard.

Statistical Analysis

Results were statistical analyzed by as methods described by Snedecor and Cochran (1989).

RESULTS AND DISCUSSION

Total Antioxidant Content of Moringa Leaves

According to our previous study, moringa leaves have antioxtint activity (Abd El baky and El-baroty, 2013). Thus, antioxidant compuods content of moringaleavies extract was determined and the data are presented in Table 1. The results revealed that the moringa extract showed significant increase in antioxidant molecule. However, the concentrations of ascorbic acid (AA),total carotenoids (T-CAR), tocopherols, total flavonoids (TFC) and total phenolic (TPCs) in moringa were 1.76 mg/100g DW, 1.65 %, 130.65 mg/100g D.W, 1.53% and 3.65%, respectively. These values revealed that *Moringa*st. was characterizes by high content of AA, TPCs and TCO.

Table 1: Antioxidant compound contents of *Maringa stenopetala leaves*

Compounds	*Maringa stenopetala*
Ascorbic acid (mg/100g DW)	1.76±0.23
Carotenoids (%)	1.65±0.31
Tocopherols (mg/100gDW)	130.65±1.87
Total flavonoids (%)	1.53±0.12
Total phenolic (%)	3.65±0.54

Major and Trace Minerals Content of *M. Stenopetala* Leaves

As presented in Table 2, the contents of major and trace minerals in *M. stenopetala* leaves. The results revealed that the moringa leaves showed significant amounts of major and trace

minerals content. The major minerals concentrations of Ca, P, Na, K and Mg, were 3.44, 4.71, 0.69, 43.54 and 3.35 g/kg DW, respectively. However, the concentrations of Fe, Mn,Zn and Cu were 550, 17.6, 18.9 and 4.72 mg/kg DW these values revealed that *M. stenopetala* leaves was characterizes by high content of Major and trace minerals.

Table 2. The contents of major and trace minerals in *M. stenopetala*leaves

Minerals	*M. stenopetala* leaves Minerals contents
Major minerals (g/kg DW)	
Calcium (Ca)	3.44±0.34
Phosphorous (P)	4.71±0.23
Potassium	43.54±1.45
Magnesium	3.35±0.54
Sodium	0.69±0.11
Trace minerals (mg/kg DW)	
Iron	550±35.4
Manganese	17.6±0.55a
Zinc	18.9±1.50
Copper	4.72±0.55

Means between tree parts within Moringa species are significantly different ($p<0.05$)

Effect of *M. Stenopetala* Leaves Extracts on Photosynthetic Pigment Contents of Wheat Plants Cultivation under Seawater Stress

The concentration of photosynthetic pigment of wheat plants irrigate sea water (10 and 20%, SW) are shown in Table 3. In general, wheat plants irrigation with SW showed a significant decrease in the amount of T-Chl and Chl-a andChl-b contents when compared with wheat plants irrigated regular water (RW). For example, T-Chl contents in wheat plant irrigated 10 and 20% (v/v) SW were 0.657 and 0.457 mg/g FW, respectively. Whereas these value was 0.792 mg g^{-1} FW, in plants irrigated with RW only. Thus, chlorophyll degradation was dependant on water salinity level.

Table 3: Influences of Maringa stenopetalas extracts spraying onchlorophyll contents of wheat leavies irrigated by diluted sea water

Salinity	Treatments	Chl a mg/ g f.w	Chl b mg/ g f.w	Tatal Chl. mg/ g f.w
Tap water	Negative control (without only spraying)	0.552±0.05	0.24±0.01	0.792
	M. stenopetalas	1.23±0.12	0.48±0.04	1.71
	Positive control (Oxalic acid)	1.46±0.43	0.68±0.05	2.14
10 % sea water	Negative control (without only spraying)	0.347±0.02	0.31±0.02	0.657
	M. stenopetalas	0.987±0.65	0.45±0.05	1.437
	Positive control (Oxalic acid)	0.765±0.22	0.23±0.04	0.995
20 % sea water	Negative control (without only spraying)	0.347±0.02	0.11±0.02	0.457
	M. stenopetalas	0.789±0.01	0.21±0.02	0.999
	Positive control (Oxalic acid)	0.695±0.05	0.33±0.01	-
LSD at (=< 0.05)		0.11	0.01	0.21

Application of *M. stenopetala* leaves extractsto wheat plants irrigated SW led to significant increasing in the concentration of T-Chl, Chl-a and Chl-b as compared with the values in plants irrigated SW only. The T-Chl content was 1.437 to 0.999 mg g^{-1} in wheat plants irrigated with 10 % and 20% (v/v) SW and treated with *M. stenopetala* leaves extract. While, in non-treated plants, these value was 0.792 mg g^{-1} FW. In similar trend for photo-synthetic pigments, T-Chl, Chl-a andChl-b contents, were observed in plant treated with oxalic acid as plant growth factor. However, this increase was less pronounced, when compared with that induced in wheat stressed plant treated with algal extracts. Thus, the level of photosynthetic pigment was found to be restored in treated wheat plants irrigated SW due to application of

algal extracts. In general, the treated wheat plants with algae extract contained high amounts of antioxidant constituents (Table 3) was positive correlated with increasing of photosynthetic pigments restored in wheat plants irrigated SW. In other word, *M. stenopetala* leaves extract might improve the salt tolerance of wheat plants by restoring the main photosynthetic pigments. The similar results were reported by Abd El Baky*et al.*, (2010 and 2013), that chlorophylls inphotosynthetic membranes could be protects the photosynthetic apparatus from excessive ROS by quenching of singlet oxygen and other radicals.

Effect of *M. Stenopetala* Leaves Extract on Antioxidant Status of Wheat Plants Cultivation under Seawater Stress

As shown in Table 4, irrigation of wheat plants with 10% and 20% (in parenthesis) SW cause significantly increased in the accumulation of antioxidant compounds includes: TCAR, TOC, AA, TPCs and GSH in wheat plants over than that in plant irrigated RW only (Table 4). The levels of those compounds were about 0.631mg /g FW (0.795mg /gFW), 0.717 (1.33µmol/ gFW), 0.498 (0.576µmol/gFW), 1.78 (1.82 µmol/ gFW) and 0.411 (0.523 mg /gFW) and 0.421 (1.7) great as that in wheat plants irrigated with RW, respectively.Application of *M. stenopetala* leaves extractcontained high level of antioxidants constituents on wheat plants irrigated 10% SW, led to significant increase in antioxidant molecules including: T-CAR, TOC, AA, TPCs and GSH with values being about 1.33(1.25), 1.73 (1.42), 1.20 (1.03), 1.37 (1.34) and 1.32 (1.82) as greater as that in non treated plants (irrigated with 10% SW, only), respectively. In plant irrigated 20% SW, these values were about 2.12 (1.68), 3.4 (.84), 1.69 (1.46), 1.62 (1.58) and 2.94 (2.3) as high as that in plants irrigated with 20% SW, respectively. According to these values, could be demonstrated that the antioxidant contents in wheat plants irrigated with sea water were positively correlated with the levels of total antioxidant in algae extracts. As compared with application foliar of growth bio regulator oxalic acid (plant growth regulator), the both algal extract caused a great increase in antioxidant compounds in wheat plant irrigated sea wheat. The T-CAR and TOC content slightly increase under salt-stress their increased was significantly higher than that of negative control plant, but was similar that in plant treated with bio regulator. In contrast, foliar application of *M. stenopetala* leaves extract significant increase antioxidant content in wheat grown under low and high SW water levels over than that wheat treated with bio regulator. The similar trend was in the literature for increased levels AA, TPCs and GSH in plants grown under stress conditions. For instance, ascorbic acid as one of the two major soluble antioxidants in chloroplast, has a photoprotective function due to it is antioxidant capacity. Application of algae extract increased the level of phenolic compounds, ascorbic acid and α-tocopherol, in wheat plants irrigated sea water to protect the membrane by preventing or reducing oxidative damage by ROS (Abd El Baky*et al.*, 2009). However, it is hypothesized a cycle where H_2O_2 is scavenged by phenolic Compounds to produce phenoxyl radicals, this radicals is reduce the ascorbic acid into mono (OH)- dehydroascorbate (Abd El Baky*et al*., 2009 and Abd El Baky and El Baroty, 2013).

Effect of *M. Stenopetala* Leaves Extract on Antioxidant EnzymesActivitieof Wheat Plants Cultivation under Seawater Stress

The change in antioxidant enzyme activity of SOD, POD, APX and CAT (enzyme activity was expressed on a protein basis, specific activity) in wheat leaves was significantly ($P \leq 0.05$) affect by irrigation of SW (Table 5). The SOD enzyme activity in wheat plant was increased with an increase of 10% and 20% SW levels, with values of 40.21and 48.24 U/mg protein/min, respectively. While, this value was 33.45U/mg protein/min in plant irrigated RW. Treated 10 and 20% SW wheat plant with *M.stenopetalas*extract had high SOD activity with values 52.23 and 57.47 U/mg protein/min, respectively. Application of bioregulator significantly increased ($P \leq 0.05$) the SOD activity in both plant irrigated RW and SW. The results revealed that SOD activity in wheat plant irrigated SW exhibited markedly important

changes by treated with foliar application of *M.stenopetalas*extracts. As compared with foliar application of oxalic acid bioregulator, *M.stenopetalas*extracted was significant increase SOD activity in wheat plant irrigated with SW.

POD, APX and CAT activity in wheat plant also increased due to irrigation both 10% and 20% SW. However, the activities of POD, APX and CAT enzyme activities were higher in SW stress wheat plant treated with *M.stenopetalas.* Also, these activities were significantly increased in plant treated with oxalic acid. In general, induce enzyme activity of SOD, POD, APX and CAT in wheat plant treated with algae extracts behaved completely similar trend with significant differences. Such similar effect of algal extract on activities of antioxidant enzyme (SOD, POD, APX and CAT) in wheat plant exposed to salt-stress here has already been observed in some plants treated with some bioregulatores. If we looking for antioxidant constituents in algal and antioxidant activities of each enzyme, it is clear that the level of algal constituents may have a significant differential role in controlling activities of antioxidant enzyme of wheat plants when irrigated SW, which ultimately resulted in differential response to salt stress .

It has been know that, salt stress lead to result in extensive lipid peroxidation, which is an effective indicator of salt-induced oxidative damage at the cellular level (Hernández and Almansa, 2002). Also, Elstner (1991) reported that salt stress causes an oxidative stress due to induction high amount of ROS in plants such as superoxides ($^{\cdot}O2$), hydroxyl ($^{\cdot}OH$) and peroxy radicals ($^{\cdot}OOH$).

In this study, lipid peroxidation was determined by TBARs content, which is one of the decomposition products of polyunsaturated fatty acids (PUFA) of lipid membrane. As shown in table 6, the high level of TBARs in wheat plants irrigated with 10 % (4.13) and 20% SW (5.98 MDA mmol/mg protein fw) than that of plant irrigated RW (1.33 MDA mmol/mg protein fw). Whereas, these levels were decreased significantly in plants treated with *M.stenopetalas*extract (2.0 and3.22 MDA mmol/mg protein fw) when compared with untreated plants. Generally, these results revealed that *M.stenopetalas*extracts it may have protection action against oxidative damage cause by salt stress. Moreover, *M.stenopetalas*could improve protection defense in wheat plant with increase efficient either non-enzyme or enzyme antioxidative system, as marker by increased antioxidant compounds (CAR, AA, GSH, TOC and phenolic) and higher activity of SOD, CAT, POX and POX and GR antioxidant enzymes. Similar results obtained with Abd El Baky*et al*., (2010), demonstrated that significant decrease in TBARs level in leaves of wheat plant grown under salt stress and treated with algae extracts rich in antioxidant compounds appeared to be correlated with increase in activity of SOD and APO and induced activity of CAT, POX and APOX enzymes. In contrast, increased MDA content in wheat plants irrigated 10 and 20% SW may indicate a higher oxidative damage due to inadequate response of the endogens antioxidative systems. Hernandez *et al*., (2000) demonstrated that plants defend against reactive oxygen species by induction of activities of certain antioxidative enzymes such as CAT, PEX, GR, and SOD, which scavenge their reactive oxygen species. Also, Muthukumarasamy*et al.,* (2000) and Rios-Gonzalez *et al*., (2002) stated that salt tolerance is often correlated with increasing the activity of antioxidative enzymes such as APX, GR and SOD, in wheat grown under sea water stress. Similar findings also, have been reported in various plants.

The higher antioxidant enzyme activities of SOD, POD, APX, GR and GST were detected in tomato, barley, maize and sunflower plants grow under salt stress (Liang, 1999; Rodriguez-Rosales *et al.,* 1999). Raza*et al*., (2007) reported that the salt tolerance in many plants is correlated with a more efficient antioxidative system.

Table 4: Influences of *Maringa Stenopetalas* Extracts Spraying onAntioxidant Compounds of Wheat Leavies Irrigated By Diluted Sea Water

Salinity	Treatments	Carotenoids (mg/g)				Tecopherols (µmol/g)				Ascorbic Acid(µmol/g)				Phenolic(mg/g)				GSH(µmol/g)			
		Ratio[a]	Ratio[b]	Ratio[c]	F.W	Ratio[a]	Ratio[b]	Ratio[c]	F.W	Ratio[a]	Ratio[b]	Ratio[c]	F.W	Ratio[a]	Ratio[b]	Ratio[c]	F.W	Ratio[a]	Ratio[b]	Ratio[c]	F.W
Tap water	Negative control (without only spraying)	0.433	0	-	-	0.653	0	-	-	0.255	0	-	-	1.56	0	-	-	0.387	0	-	-
	M. stenopetalas	0.786	2.38	-	-	1.33	2.04	-	-	0.497	1.94	-	-	2.22	1.4	-	-	0.643	1.7	-	-
	Positive control (Oxalic acid)	0.522	1.21	-	-	0.921	-	-	-	0.335	1.31	-	-	1.98	1.27	-	-	0.421	1.08	-	-
10 % sea water	Negative control (without only spraying)	0.631	1.56	0	0.793	0.717	1.09	0	-	0.498	1.95	0	-	1.78	1.14	0	-	0.411	1.06	0	-
	M. stenopetalas	0.998	2.3	1.33	1.25	1.89	2.89	1.73	1.42	0.777	3.04	1.56	1.34	2.45	1.55	1.37	1.34	0.954	2.46	2.32	1.82
	Positive control (Oxalic acid)	0.751	1.73	0.75	0.944	1.41	2.16	1.96	1.06	0.598	2.34	1.2	1.00	1.79	1.14	1.00	0.98	0.765	1.97	1.9	1.46
20 % sea water	Negative control (without only spraying)	0.795	1.83	0.79	0	1.33	2.00	1.85	0	0.576	2.25	1.15	0	1.82	1.16	1.02	0	0.523	1.35	1.27	0
	M. stenopetalas	1.34	3.09	2.12	1.68	2.45	3.8	3.40	1.84	0.843	3.31	1.69	1.46	2.89	1.85	1.62	1.58	1.21	3.12	2.94	2.3
	Positive control (Oxalic acid)	0.976	2.25	0.977	1.22	1.78	2.72	2.48	1.33	0.711	2.78	1.42	1.23	1.85	1.18	1.00	1.01	0.876	2.26	2.13	1.67
LSD at (=< 0.05)		0.34				0.33				0.12				0.27				0.18			

Ratio[a] :Treatments Negative control (without only spraying and irrigated by tap water)
Ratio[a] : Treatments Negative control (without only spraying and irrigated by 10 % sea water)
Ratio[a] : Treatments Negative control (without only spraying and irrigated by 20 % sea water)

Table 5: Influences of *Maringa Stenopetalas* Extracts Spraying onAntioxidant Enzymes Activitesof Wheat Leavies Irrigated

Salinity	Treatments	U/mg protein/min															
		SOD	Ratio[a]	Ratio[b]	Ratio[c]	POD	Ratio[a]	Ratio[b]	Ratio[c]	APX	Ratio[a]	Ratio[b]	Ratio[c]	CAT	Ratio[a]	Ratio[b]	Ratio[c]
Tap water	Negative control (without only spraying)	33.45	0	-	-	35.44	0	-	-	267.11	0	-	-	19.56	0	-	-
	M. stenopetalas	38.87	1.16	-	-	41.76	1.17	-	-	374.12	1.4	-	-	27.44	1.4	-	-
	Positive control (Oxalic acid)	36.45	1.08	-	-	37.78	1.06	-	-	330.9	1.2	-	-	23.58	1.2	-	-
10 % sea water	Negative control (without only spraying)	40.21	1.2	0	-	39.23	1.1	0	-	319.24	1.2	0	-	27.15	1.4	0	-
	M. stenopetalas	52.23	1.5	1.29	-	46.37	1.3	1.2	-	411.45	1.5	1.3	-	38.96	1.99	1.4	-
	Positive control (Oxalic acid)	42.56	1.27	0.81	-	43.25	1.22	1.1	377.2	1.4	1.2	1.2	-	30.65	1.7	1.1	-
20 % sea water	Negative control (without only spraying)	48.24	1.44	1.19	0	45.89	1.3	1.2	0	345.5	1.3	1.1	0	31.77	1.6	1.2	0
	M. stenopetalas	57.47	1.7	1.42	1.19	52.29	1.5	1.33	1.1	448.5	1.7	1.4	1.3	45.33	2.3	1.7	1.4
	Positive control (Oxalic acid)	52.88	1.58	1.31	1.09	48.11	1.35	1.22	1.04	410.6	1.6	1.3	1.2	37.65	1.9	1.4	1.2
LSD at (=< 0.05)		0.34				0.33				0.12				0.27			

Ratio[a] :Treatments Negative control (without only spraying and irrigated by tap water)
Ratio[a] : Treatments Negative control (without only spraying and irrigated by 10 % sea water)
Ratio[a] : Treatments Negative control (without only spraying and irrigated by 20 % sea water)

In general, the similar response of wheat to application both *M.stenopetalas*extract had protective effect on wheat plant due to differential by which amount of ROS was decreased in plant treated with higher quantity and quality of antioxidant compounds, there by resulting into high activities of antioxidant enzyme (APX, CAT, SOD and POD) in wheat plant irrigated sea water. Abd El Baky*et al*., (2012) reported that algal rich content antioxidant is *M.stenopetalas*know to scavenge hydroxy radicals and other reactive oxygen species in plants exposed to salt stress and their effect coupled with elevation in the activities of all antioxidant enzymes.

Table 6: Effect of *Maringa stenopetala* extracts on lipids peroxidation in wheat plants irrigated with sea water

Salinity	Treatments	TBARs (MDA mmol/mg protein)
Tap water	Negative control (without only spraying)	1.33±0.21
	M. stenopetalas	1.04±0.11
	Positive control (Oxalic acid)	1.21±0.05
10 % sea water	Negative control (without only spraying)	4.13 ±0.21
	M. stenopetalas	2.01±0.13
	Positive control (Oxalic acid)	2.67±0.15
20 % sea water	Negative control (without only spraying)	5.98±0.41
	M. stenopetalas	3.22±0.29
	Positive control (Oxalic acid)	2.87±0.21
LSD at (=< 0.05)		0.12

Effect of *M.Stenopetalas*Extracts on Dry Weight and Grain Yield of Wheat Plants Cultivation under Sea Water Stress

The irrigation wheat plant with 10 and 20% SW showed adviser effect on the overall growth parameters, that caused significant reduction of plant height, shoot fresh, spike length and spikelet's/ spike (data not showed). Dry weigh (PDW) and grain yield (GY) of wheat plants irrigated RW were significantly reduced 1.34 g and 3.89 g, respectively (Table 7). Treated wheat plant irrigated 10% and 20% SW with of *S M.stenopetalas*extracts showed markedly significant increase of both PDW and GY, with values ranged (3.86 g and 3.26) and (4.71g and 4.11g), respectively. Their elevation in PDW and GY of treated plant with algae extracts may be due to their improvement in all growth parameter. Similar effect was noted in plant treated with oxalic acid, but it was less than that in plant treated with algae extracts. However, low FW, DW and GY in wheat and barley plants irrigated SW has been reported in literature (Sairam and Srivastava, 2002; Francois *et al*., 1984; Abd El Baky*et al.,* 2010) stated that improve of PDW and SY as one of the parameters of salt tolerance in wheat irrigated SW treated with algae extract indicated that metabolic and photosynthetic processes was restored.

Table 7: Effect of *Maringa stenopetala* extracts on dry weight and yield in wheat plants irrigated with sea water

Salinity	Treatments	Whole plant dry weight(g)	100 grain weight(g)
Tap water	Negative control (without only spraying)	1.34±0.23	3.89±0.19
	M. stenopetalas	3.54±0.11	5.44±0.17
	Positive control (Oxalic acid)	2.65±0.13	3.37±0.21
10 % sea water	Negative control (without only spraying)	1.02±0.09	3.11±0.21
	M. stenopetalas	3.86±0.25	4.71±0.26
	Positive control (Oxalic acid)	2.89 ±0.32	3.87±0.76
20 % sea water	Negative control (without only spraying)	0.98±0.11	2.95±0.22
	M. stenopetalas	3.26±0.24	4.11±0.23
	Positive control (Oxalic acid)	2.76±0.26	3.13±0.19
LSD at (=< 0.05)		0.18	0.19

Here again,application of algal extracts seemed to reduce SW salinity stress of wheat plants by restored of photosynthetic pigments, and increase of antioxidant defense abilities included non-enzymatic and enzymatic antioxidant systems, which led to reduce of oxidative damage of

functional molecules and maintenance of many physiological processes of wheat plants such as photosynthetic activity and productivity (Abd El Baky*et al.*, 2010).

Finally, the*M.stenopetalas*extracts could be contain some bioactive components act as growth regulator substances such as auxine and cytokinins, in additional of antioxidant compounds (table 1) which lead to mitigate the effect of sea water salinity stress on wheat plant metabolic pathway. The results of Molnar and Ördög (2005) and Abd El Baky, *et al*., (2010) reported that some plant growth regulators found in microalgae possessed beneficial effects on tissue cultures of recalcitrant plants.

CONCLUTION

In conclusion, the results indicate that, treated wheat plant irrigated SW with *M.stenopetalas*extracts could be provide protection action against oxidative stress induce salt stress due to elevation the levels of non-antioxidant constituents and enzyme antioxidant protective systems, which involved as mean one of the factor responsible for salt tolerance of wheat plants. Therefore, the irrigation of wheat plants by mean of brackish water at 20% (v/v) is possible when treated with algal extracts.

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