

## Biochemical Efficiency of *Chondrus crispus* Aqueous Extract for Alleviating Salinity Stress in *Vicia faba* L. cv. Masr-1

Walaa A. Elshalakany

Department of Biochemistry, Faculty of Agriculture, Ain Shams University, Cairo, Egypt.

Received: 25 Oct. 2020 / Accepted 15 Dec. 2020 / Publication date: 30 Dec. 2020

### ABSTRACT

One of the most significant abiotic factors that restrict the development and yield of crops is saline stress. It can have a considerable impact on a plants biochemical characteristics and, in turn the economic output of crops. Since faba bean (*Vicia faba* L. cv. Masr-1) are sensitive to salt, my study looked into the effects of applying red algal seaweed extract (15 and 30% red algae extract) on the growth, biochemical and tolerance features of seedlings cultivated under saline stress conditions (3000, 6000, and 12000 ppm NaCl). Plants treated with saline treatments had considerably less fresh, dry weight and chlorophyll content. All faba bean seedlings died at the greatest salinity level 12000 ppm NaCl. However, seaweed treatment (15% red algal extract) reduced the effects of salt stress and markedly increased the contents of phenolic and proline, pigments, and enzyme activity. While the treatments for seaweed extraction reduced electrolyte leakage (EL) and malondialdehyde (MDA) contents. In conclusion, salt stress may be greatly lessened by treating faba bean seedlings with red algal extracts at concentration 15%.

**Keywords:** Biochemical traits, Enzymes activity, Red algae, *V. faba*, Saline stress, Seaweed extract.

### 1. Introduction

Regarding the use of fresh and dried seeds yield for human food and animal feeding, the faba bean (*V. faba* L.) is regarded as one of the most important legume crops in the world due to its high protein content (up to 35%) and good source of other nutrients and bioactive compounds, such as K, Ca, Mg, Fe, Zn, polyphenols, carotenoids, and carbohydrates (Longobardi *et al.*, 2015; Landry *et al.*, 2016). Faba bean plants is a good suitable crop for enriching the soil nitrogen content for symbiosis with rhizobium that can enhancement increasing reclamation rate of the marginal lands (Cazzato *et al.*, 2012), however, faba bean is more sensitive to salt stress than other legume plants like cowpea (*Vigna unguiculata* L.) (Shaddad *et al.*, 2014).

Salinity is considering one of the most environmental adverse that has direct and indirect reduction impedes plant growth and agricultural economic yield production around world by 50 - 70% (Osakabe *et al.*, 2014). In addition, extreme soil salinity has additional detrimental effects on plants, including the suppression of enzyme activity and the production of lipids, proteins, and nucleic acids due to an increase in the rate at which reactive oxygen species (ROS) are accumulating (Parvaneh *et al.*, 2012); decreasing on cellular integrity, respiration, mitochondrial electron transport chain and photosynthesis (Apel and Hirt, 2004); premature plant leaf senescence; and disturbance the nutrient uptake by the plants due to the soil ionic imbalance (Perveen and Nazir, 2018). One of the negative impacts of salinity is the reduction in chlorophyll concentration, which inhibits translocation and the absorption of photosynthetic products, which has a limiting influence on plant leaf area development (Netondo *et al.*, 2004; and Ehsanzadeh *et al.*, 2009).

Plants can overcome the damage of salinity throughout various mechanisms that including the positive feedbacks of external biotic or abiotic stimulators for increasing plant tolerance against salinity adverse (Bose *et al.*, 2014). There are several stimulators used to ameliorate the plant ability for salinity drawback such as amino acids, organic acids and, extracts of seaweed and microalgae (Chiaiese *et al.*, 2018). Similarly, by boosting antioxidant activities such superoxide dismutase (SOD), catalase (CAT), peroxidase (POD) and ascorbate peroxidase (APX), furthermore antioxidant molecules such ascorbic acid (AA) and reduced glutathione (GSH), the seaweed can induce the defense systems in parallel with

under saline abiotic stress in order to reduce the harmful effects of free radicals and for plants to survive under stressful conditions (Sharma *et al.*, 2012).

Seaweeds comprise a provenance of many substances such as; polysaccharides, vitamins, amino acids, auxins, cytokinins, abscisic, macro and micro-elements. These components are crucial for plant physiology to adapt to stressful situations, for increasing plant vegetative growth and crop yield, and for increasing plant immunity to disease infection (Elshalakany, 2016). In addition to the aforementioned, it is possible to recover the effects of salt on plant development by using the extract of some marine algae as bio-fertilizers in seed soaking treatments (Shaddad *et al.*, 2013). The current study intends to assess the effects of red algal aqueous extract treatment on salt-sensitive *V. faba* cv. Masr-1 seedlings growing under saline stress in terms of growth, biochemical, and tolerance features.

## 2. Material and methods

### 2.1. Source of seaweed:

The samples of red seaweed algae were taken at Abu-Qir Beach in Alexandria, Egypt coastal region, and were immediately recognized as *Chondrus crispus* (Rhodophyceae) at the National Institute of Oceanography and Fisheries (NIOF), Alexandria, Egypt. According to Bhosle *et al.*, (1975), the morphologically unique thallus of red algae was packed individually in the new polyethylene bags, following which it was stored in an icebox with slush ice and brought right to the lab for aqueous extraction processes. To remove the salt from the samples surfaces, they were rinsed with distilled water, and the surplus water from the drained thallus was spread out on blotting paper. One kilogram of frozen fresh red algal culture in liquid nitrogen were crushed and macerated in 1 L of dist. H<sub>2</sub>O for 24 hr under continuous 150 rpm shaking at 20 °C. The crude aqueous extract was filtered through muslin cloth and then sterilized through Whatman number 41 (pore size 20-25 µm) filter paper and directly the concentration was adjusted as 15 and 30%, and stored at 4 °C for the following studies. The chemical components of *C. crispus* were evaluated and presented according to Elshalakany (2016).

### 2.2. Plant materials and experimental proceeding:

From the genetic breeding program of the Agricultural Research Center (ARC), Dokky, Cairo, Egypt, seeds of the *V. faba* L. cv. Masr-1, sensitive to salt stress, were obtained. Design; the experiment was created utilizing a two-way analysis of the Randomized Complete Block Design as 3 concentrations of red algal crude aqueous extract (zero, 15 and 30%) under for 4 treatments of salinity stress of NaCl (zero, 3000, 6000 and 12000 ppm)/each concentration of algal extract, and 3 replicates/each NaCl concentration, distributed 30 polyethylene bags (4×8×13, Cm) total, each containing 700 g of acid-washed sand soil. Application of red algal crude aqueous extracts; through seed soaking, the seeds were soaked for 24 hrs before planting and divided into 3 groups (1<sup>st</sup> group soaked in dis. H<sub>2</sub>O as control, 2<sup>nd</sup> and 3<sup>rd</sup> group are 15 and 30 % of crude aqueous red algal extracts receptively, and then transplanting as 2 seeds/bag, and all bags were kept and daily observed until completely germination. Throughout fertigation, after 20 days from planting all treatments are daily fertigating continuously 3 times for 15 days consist of individual separately 2 times fertigation alone by using nutrient solution according to Hoagland, (1950) for 1<sup>st</sup> group of red algal application. While, in the 2<sup>nd</sup> and 3<sup>rd</sup> groups were done using Hoagland nutrient solution prepared in 15 and 30% of red algal crude aqueous extracts. The 3<sup>rd</sup> time of irrigation was carried out in all application groups using dis. H<sub>2</sub>O to avoid the nutrients accumulation. Salinity treatment, the salinity stress was achieved at 35 days age stage of faba bean using 4 different concentrations of NaCl (zero, 3000, 6000 and 12000 ppm) for for all groups of red algal application dissolved individual in zero red algal extract in 1<sup>st</sup> group; while dissolved in 15% and 30% red algal extract for 2<sup>nd</sup> and 3<sup>rd</sup> group, respectively. All different individual NaCl concentrations were used for irrigating the plants every day for total time 7 days by poured rate 2 times while the 3<sup>rd</sup> time were carried out with using dis. H<sub>2</sub>O alone. Leaves sample collection, the plant leaves for vegetative parameter determination and biochemical analysed were harvested at 42 days after plantation for all different NaCl concentrations for 3 ways of red algal application except at salinity stress with 12000 ppm of NaCl that have harmful stress effects up to death of all plants, so there is not results data recorded on it. One part of individual harvested leaves samples was divided twice into 2 groups, one of them freshly weighted (FWt) and reweighted again after drying in oven 72 °C/72 hrs to get dry weight (DWt). While the other group were freshly weighted and then distributed to 2 groups, one used or another stored at -20 °C according for the following farther vegetative or biochemical analyses.

### 2.3. Biochemical analyses:

All biochemical analyses including, plant pigments content, proline content, total phenols content, malondialdehyde (MDA) content, and antioxidants activities were procedured and measured employing a UV-Vis Spectrophotometer UV 9100 B, Lab. Tech., in the Biochemistry Dept., Fac. of Agric., Ain Shams Univ., Cairo, Egypt.

### 2.4. Content of plant pigments:

For the chlorophylls and carotenoids analyses, 0.2 g of FWt leaves were extracted by crushing the tissue in a mortar using 10 mL N, N dimethylformamide (Wellburn, 1994). The resulting extracts were incubated in the dark fridge overnight and the total chlorophyll and carotenoids concentrations were measured at 470, 647 and 664 nm and then calculated as follows:

$$\text{Chlorophyll A conc.} = (12 A_{664}) - (3.11 A_{647}),$$

$$\text{Chlorophyll B conc.} = (20.78 A_{647}) - (4.88 A_{664}),$$

$$\text{Carotenoids conc.} = [(1000 A_{470}) - (1.12 \text{ Chl. a}) - (34.07 \text{ Chl. b})]/245.$$

### 2.5. Electrolyte leakage (EL%):

The plant cell membrane permeability was assessed throughout electrolyte leakage measurement in the solution of fresh sharp leaf discs using an electrical conductivity meter and calculated as the percentage of total ions released (Singh and Jha, 2016).

### 2.6. Proline content:

Troll and Lindsley (1955) used a ninhydrin colorimetric technique to determine the proline concentration, which Peters *et al.*, (1997) modified. Plant leaf tissues that had been frozen were crushed in a mortar and then homogenized with 100 mM sodium phosphate buffer, pH 6 (1:10, w:v). This procedure was followed by centrifugation for 10 min at 4500 rpm. 200  $\mu$ L of the extract were mixed with 1 mL of ninhydrin solution, which was made by dissolving 2.5 g of the compound in 100 mL containing orthophosphoric acid, acetic acid, and water (15:60:25, v:v:v). This procedure took place for 1 hour in boiling water. The extracted dye was then vigorously vortexed for 15 seconds while being extracted with 1 mL of toluene. The toluene phase was then detected at 515 nm right away. The proline concentration was calculated as  $\mu$ g proline g FWt<sup>-1</sup> from the standard curve of L-proline.

### 2.7. Content of total phenols:

The Folin-Ciocalteu technique, which uses the catechol standard curve, was used to quantify the total soluble phenolic content in the ethanolic extract of 80% of the FWt samples. The absorbance was measured at 650 nm after the reaction mixture of 0.5 mL of the extract, 2.5 mL of Folin-Ciocalteu's reagent, and 2 mL of sodium carbonate (7.5%) in test tube was mixed and left to stand for 30 min (Singleton and Rossi, 1965). Total phenol content was calculated using the catechol standard curve, the amount of total soluble phenols was determined as  $\mu$ g equivalents of catechol per g FWt of the sample.

### 2.8. Malondialdehyde (MDA) Content:

According to Heath and Packer (1968), the amount of lipid peroxide was determined in the context of MDA by using the thiobarbituric acid (TBA) test. After being homogenized in 0.1% (w/v) trichloroacetic acid (TCA), frozen tissues were centrifuged at 5000 rpm for 10 minutes. The reaction mixture consisted 1 mL of the supernatant and 4 mL of 0.5% (w/v) TBA that was dissolved in 20% (w/v) TCA and boiled in boiling water for 30 minutes before being immediately cooled at room temperature and centrifuged at 5000 rpm for 15 minutes. The supernatant was then measured at 535 nm and the MDA concentration was calculated as  $\mu$ m/g FWt using an extinction coefficient of 155 mM<sup>-1</sup>Cm<sup>-1</sup>.

### 2.9. Antioxidants activities:

#### 2.9.1. Soluble protein content:

In accordance with Cakmak *et al.*, (1993), fresh green leaves (500 mg) were crushed to a fine powder in liquid N and then extracted with 5 mL of cooled off extraction buffer (50 mM K-phosphate buffer, pH 7.6 and 0.1 mM Na<sub>2</sub>-EDTA). Based on the standard curve of bovine serum albumin, the

mixture was directly centrifuged at 20,000 g at 4 °C for 30 min, and then the supernatant was used to assess the content of soluble proteins and the activity of several antioxidant enzymes (Bradford, 1976).

### 2.9.2. Peroxidase activity (POD):

Their method was applied to measure the peroxidase activity by Hammerschmidt *et al.*, (1982). The sample used in the test mixture (1 mL) contains 0.8 mL of 50 mM phosphate buffer, pH 6.6, 0.1 mL of 0.3% H<sub>2</sub>O<sub>2</sub>, and 0.1 mL of 1% (v/v) guaiacol. For three minutes, the absorbance at 470 nm was measured every 30 seconds.

### 2.9.3. Catalase activity (CAT):

The technique developed by Chance and Maehly in 1955 and improved by Cakmak *et al.*, (1993) was used to measure catalase activity. By keeping an eye on the drop in absorbance at 240 nm after H<sub>2</sub>O<sub>2</sub> breakdown for one minute, CAT activity was calculated.

### 2.9.4. Phenylalanine ammonia-lyase activity (PAL):

The Lister *et al.*, (1996) approach was used to measure the activity of PAL. The amount of 100 µL crude enzyme, 1.9 mL of pH 8.8, 0.05 M Tris-HCl buffer, and 1 mL of 20 mM L-phenylalanine made up the reaction mixture. The addition of 0.2 mL of 6 M HCl brought the reaction to an end after it had been permitted to continue for one hour at 37 °C. The amount of enzyme that increased absorbance by 0.01 per hour at 290 nm is considered one unit of enzyme activity.

## 2.10. Statistical analysis

Using the Gomez and Gomez (1984) technique, data were statistically analyzed using Costas software (version 6.4, CoHort Software, USA). According to Snedecor and Cochran (1980), all data were subjected to a two-way analysis of variance, and the means were compared using the Duncan's multiple range test at a significant level  $P \leq 0.05$ .

## 3. Results

Compared to the non-stressed plants, the studied plants output of fresh and dry mass decreased significantly due to NaCl salinity. The faba bean died as a result of the harmful effects caused by the excessive saline level (12000 ppm NaCl). The strong stimulatory impact on fresh and dry weight without respect to salt concentration demonstrated the good benefits of seaweed water extracts on evaluated plants growing in the salinized circumstances, which somewhat mitigated the detrimental effect of salinity on growth (Table 1). Also, the highest values (26.78 and 27.35 g FWt/plant) was given at plants grown with algae extract of 15 and 30% red algal extract, with increase of 28 and 31% respectively over the control, followed by plants grown with 3000 ppm NaCl and algae extracts.

**Table 1:** Effects of red algae extract application on the fresh and dry weight (g plant<sup>-1</sup>) of faba bean growing under salt stress.

Red algae (%)	NaCl (ppm)	Fresh weight (g/plant)	Dry weight (g/plant)
Zero	Free	20.91b	1.62b
	3000	11.39d	1.11c
	6000	4.77e	0.42d
15	Free	26.78a	2.48a
	3000	21.36b	1.72b
	6000	15.74c	1.22c
30	Free	27.35a	2.51 a
	3000	20.78 b	1.68b
	6000	16.71c	1.17 c
LSD 0.05		0.728876	0.137

The same letters in each column indicate no significant differences among different treatments referring to the least significant difference LSD test ( $P \leq 0.05$ ).

Regardless of the salt amount applied, salinity treatment had an inhibitory influence on the photosynthetic pigment content (Chlorophyll A, chlorophyll B, and carotenoids) as well as the overall pigment content in each of the examined plants. When compared to salinized plants, pre-soaking in 15% aqueous algal extract caused a noticeable increase in pigment content (Table 2). Regardless of the salinity level utilized, this stimulatory impact on photosynthetic pigments was noticeable in plants treated with *C. crispus* extract.

**Table 2:** Effects of red algae extract application on photosynthetic pigments of faba bean plants growing under salt stress.

Red algae (%)	NaCl (ppm)	Chlorophyll A (mg/g)	Chlorophyll B (mg/g)	Carotenoids (mg/g FWt)
Zero	Free	1.08b	0.31a	0.16c
	3000	1.06b	0.2b	0.14d
	6000	0.99c	0.19b	0.13e
15	Free	0.98c	0.36a	0.15d
	3000	1.28a	0.21b	0.19a
	6000	1.09b	0.23b	0.18b
30	Free	0.96 c	0.35a	0.15d
	3000	1.25 a	0.22 b	0.21 a
	6000	0.98 c	0.25b	0.17 b
LSD 0.05		0.018992	0.017429	0.007638

The same letters in each column indicate no significant differences among different treatments referring to the least significant difference LSD test ( $P \leq 0.05$ ).

Under various applications of salt stress and red algae extracts, the proline and total phenolic contents significantly varied (Table 3). The plants cultivated with the greatest salt stress levels of NaCl (6000 ppm) and algal extract application of 15% received the highest values (10.12 and 127.17  $\mu\text{g/g}$  FWt) of proline and total phenolic component contents respectively, followed by plants grown with 3000 ppm NaCl and algae extract of 15% (Table 3). On the other hand, as high salt stress rose in faba bean plants, the MDA contents and EI% considerably increased in values (48.04  $\mu\text{m/g}$  FWt and 65.15%, respectively) as presented in (Table 3).

**Table 3:** Effects of red algae extracts on stress markers (proline, total phenolic compounds, malondialdehyde content, and electrolyte leakage of faba bean plants growing under salt stress.

Red algae (%)	NaCl (ppm)	Electrolyte leakage (%EL)	Proline ( $\mu\text{g/g}$ FWt)	Phenolic ( $\mu\text{g/g}$ FWt)	MDA ( $\mu\text{m/g}$ FWt)
Zero	free	27.79d	5.12e	60.23e	27.93e
	3000	46.42b	7.28 d	77.64d	40.6b
	6000	65.15 a	8.57 c	100.4c	48.04a
15	free	21.42 e	4.85 e	125.83a	23.54f
	3000	31.03 c	9.26b	117.8b	31.18d
	6000	34.21 c	10.12 a	127.17a	33.54c
30	free	20.83e	4.91 e	111.56b	25.46
	3000	33.74 c	8.51c	121.33a	32.63 d
	6000	37.83c	7.32 d	102.72c	35.55 c
LSD 0.05		1.352626	0.672319	4.909781	0.717861

The same letters in each column indicate no significant differences among different treatments referring to the least significant difference LSD test ( $P \leq 0.05$ ).

The administration of algal extract while simultaneously raising salt stress levels greatly increased the activity of CAT, POD, and PAL. In faba bean plants cultivated under levels of 3000 and 6000 ppm salt stress, application of red algal extract dramatically boosted all antioxidant enzymes by a

factor of roughly 3-4 compared to the faba bean control value (Table 4). The plants grown with NaCl (3000 and 6000 ppm) in the presence of algal extract application (30%) had the highest POD activity (36.38 and 33.15  $\text{Umg}^{-1}$  protein, respectively), followed by the plants grown with NaCl (3000 and 6000 ppm) in the presence of algal extract application (15%). Additionally, as indicated in table 4, the plants grown with salt stress levels of NaCl and algal extract administration had the greatest CAT and PAL activity.

**Table 4:** Effects of red algae extracts application on the antioxidant enzymes activities of faba bean plants growing under NaCl salt stress.

Red algae (%)	NaCl (ppm)	Soluble protein (mg/g FWt)	POD (Sp. activity)	CAT (Sp. activity)	PAL (Sp. activity)
Zero	Free	0.171 c	6.58e	25.73f	89.55e
	3000	0.173 c	20.24c	84.51d	219.56c
	6000	0.24 9a	21.65c	83.76d	222.91c
15	Free	0.184 b	8.12e	61.37e	170.24d
	3000	0.242a	24.6b	130.62b	245.99b
	6000	0.262 a	23.18b	135.14b	249.81b
30	Free	0.179 b	14.68d	118.06c	187.34d
	3000	0.215b	36.38a	140.31a	330.82a
	6000	0.261a	33.15a	144.16a	301.89 a
LSD 0.05		0.00446887	1.36253674	31.27729	24.64664

The same letters in each column indicate no significant differences among different treatments referring to the least significant difference LSD test ( $P \leq 0.05$ ).

#### 4. Discussion

Salinity stress can have a negative impact on a variety of agricultural characteristics, including yield, root and leaf attributes, water relations, ion absorption, and photosynthesis (Gama *et al.*, 2007). This may be because plants growing under salt stress have their metabolism inhibited (Munns, 2002; Gong *et al.*, 2013). Using red algae extracts might thus promote plant development and raise agricultural yield when it comes to crops cultivated under various salt stress conditions. This is caused by the presence of bioactive compounds such macro and micronutrients, amino acids, and growth hormones that stimulate cell metabolism in treated plants, promoting plant development and increasing production (Houssien *et al.*, 2011). In current investigaion, applications of salt stress dramatically reduced the quantity of plant pigments. Seaweed extracts benefit plants by promoting seed germination, stimulating growth, and enhancing photosynthetic pigments, according to Battacharyya *et al.*, (2015). The current study's findings showed that using red algae extracts under conditions of increased salinity (3000 and 6000 ppm NaCl) enhances the amount of chlorophyll A and chlorophyll B. Similar outcomes were attained by (Gireesh *et al.*, 2011), who demonstrated that the total chlorophyll content of cowpea (*Vigna unguiculata*) rose by 20% as a result of *Ulva lactuca* extracts when compared to the control treatment. The high content of N (1.0%) and Fe (0.20%) in algae extracts, which accounts for the high rate of photosynthesis in the plant treated with algal extracts, could increase cellular metabolic rate and delay plant senescence by protecting and preventing the senescence of chloroplasts, delaying the destruction of chlorophyll, and/or increasing the biosynthesis of the latter (Gharib *et al.*, 2014).

Except for un-stressed plants, where the application of algae extract at concentrations of 15 and 30% significantly increased electrolyte leakage when compared to other stressed plants, the results of this study showed that, there were distinct differences in electrolyte leakage between red algae extracts and salt stress treatments. According to Stevens *et al.*, (2006), a crucial component of the salinity tolerance mechanism is the maintenance of membrane integrity under salt stress. In order to prevent electrolyte leakage, red algae extract can preserve the membrane's integrity.

Lower proline concentrations under stress have been linked to improved crop cultivar stress tolerance in common bean plants. Proline accumulation has a beneficial function in the salt tolerance mechanisms of several crops by contributing to membrane integrity (Ashraf and Harris, 2004; Perveen and Nazir, 2018). It is a sensitive physiological measure of plant response to diverse abiotic stimuli.

Similar to my study's findings, which showed that employing red algae extracts together with a lot of salt stress significantly increased the concentration of proline. According to this study, (El-Sharkawy *et al.*, 2017) found that using algal extracts under salt stress increased the proline content of farmed alfalfa (*Medicago sativa* L.) by 24% in comparison to the control. By detoxifying ROS generated as a result of NaCl toxicity, free proline can lessen ROS damage, improve plant tolerance, and reduce NaCl saline stress. Additionally, it might physically put out singlet oxygen or specifically react with hydroxyl radicals (Howladar, 2014).

The primary sources of antioxidants in crops, phenolic compounds are regarded as key secondary metabolites of plants that play a significant role in plant growth and development (Elansary *et al.*, 2016). As a result, plants with high antioxidant content can demonstrate strong resistance to the oxidative damage brought on by salt stress (Meloni and Martnez, 2009). In plants growing under salt stress, the application of red algal extracts considerably boosted the total phenolic compounds, according to the results of the current study. According to Neffati *et al.*, (2011), variations in the synthesis of polyphenols responding to abiotic variables stimuli may be the cause of the rise in total phenolic compounds in plants.

Malondialdehyde (MDA), a biochemical stress indicator, was used in the current study to quantify lipid peroxidation. MDA prevents the growth of biomass and reduces the potential effects of a plants adaptation to stress (Hernández and Almansa, 2002). In contrast to the use of red algae extracts, MDA content was much higher in current work at doses of 3000 and 6000 ppm of NaCl. Lipid peroxidation causes a rise in MDA levels, and lipid peroxidase activity may be responsible for membrane degradation (Nisha *et al.*, 2013; Hassan *et al.*, 2017). The increase in MDA was also linked by Ebrahimian and Bybordi (2012) to ROS, which result in membrane lipid peroxidation, selectivity, and decreased membrane fluidity. The MDA concentration, which rose in response to salt stress, was dramatically reduced after treatment with seaweed water extracts. According to Elshalakany (2016), the foliar application of wheat led to improved growth, yield, and quality results. This was due to the presence of nutritional elements and plant growth regulators provided in the extract of *C. crispus*.

However, faba bean plant growth traits were promoted by the use of red algae extracts. In this regard, (Shaddad *et al.*, 2014) found that, plants exposed to 1% aqueous extract of *Sargassum dentifolium* or *Padina gymnospora* applied via seed soaking experienced stimulation in growth, photosynthetic pigment production, and antioxidant enzyme activity. The extract from *S. dentifolium* exhibited the highest level of activity. The ability of a plant to tolerate saline stress and the presence of an efficient antioxidant system within the plant are strongly correlated, and resistance to saline stress is consequently closely correlated with the effectiveness of such an antioxidant system (Raza *et al.*, 2007). In order to combat the effects of salt, plants that are exposed to saline stress attempt to develop a sophisticated and potent antioxidant system as well as ROS scavenging enzymes (Apel and Hirt, 2004). The current work findings are in accordance with those of Shaddad *et al.*, (2014), who discovered that plants subjected to NaCl salt stress exhibited considerably higher antioxidant enzyme activity. Additionally, Shaddad *et al.*, (2014) reported that saline stress increased the amount of antioxidant metabolites and antioxidant activity enzymes in *V. faba*. Therefore, our findings suggested that red algae extracts might aid in the detoxification of H<sub>2</sub>O<sub>2</sub> by increasing the activity of antioxidant enzymes during saline stress. Ultimately, saline stress was reduced by using red algae extracts, probably by scavenging ROS and safeguarding antioxidant enzymes.

## 5. Conclusion

The faba bean plants cv. Masr-1 cultivated under salt stress levels 3000 and 6000 ppm NaCl, responded better when red algal extracts were applied exogenously. The protective peroxidation-linked membrane deterioration and free radical scavenging for treated red algal extracts used exogenously in faba bean plants growing under salt stress may be responsible for the positive benefits. The use of 15% red algal extract is ultimately advised for plants growing in high saline environments 6000 ppm NaCl. Additionally, the findings of the present study not only allow us to draw the conclusion that treating seeds and seedlings with red algal extracts could reduce the negative effects of excess NaCl, also recommend that, the application of these extracts could be used as a biological amendment in soil reclamation techniques that could increase food production in cultivated lands and barren soils that have accumulated salt.

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