



Effect of *Sargassum wightii* extracts on NaCl treated sorghum (*Sorghum vulgare* L.)

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ABSTRACT

The amelioration of *Sargassum wightii* SLF on NaCl treated *Sorghum* was studied. *Sorghum* plants were grown in pots and treated with different concentrations of NaCl supplemented with *S. wightii* extracts at different concentrations. The physical and biochemical parameters of *Sorghum* were studied on the 30th and 45th day after sowing. The results show a decrease in chlorophyll content and an increase in carotenoid content in the salt – treated plants. The carbohydrate content of the salt stressed plants was higher than the control plants. The amino acid content and total soluble sugar content increased across all treatments but the protein content was observed to have decreased in the treated plants. The amino acid content increased in response to the salt stress. The application of *S. wightii* SLF affects the mechanism of salinity tolerance in *Sorghum*.

Introduction

Soil is the medium for supporting plant growth and development. Soil properties influence fertility, water relations, gas exchange and physical support of plant roots. Soil rarely provides ideal conditions for plant growth. The productivity of crops is adversely affected by high salt content in most of the soils (Alam *et al.*, 2000). Environmental stresses such as high and low temperature, drought and salinity are potentially harmful to the plants (Van Breusegem *et al.*, 2001). Salt stress is certainly an absolute stress limiting crop production and affecting agriculture worldwide (Ashraf, 1994). Salinity included osmotic stress causes specific ion toxicity and affects the activity of major cytosolic enzymes by dissovving intracellular K⁺ homeostasis. It also causes oxidative stress in plant cells (Cuin and Shabala, 2007; Chen *et al.*, 2007). Conventional selection and breeding techniques have been used to improve salinity tolerance in crop plants (Ashraf, 2002).

Seaweeds have long been used as soil fertilizers. Several advantageous effects of spraying crude extracts of seaweeds on plant growth such as improved germination, higher yields and increased resistance to diseases are reported (Abdel-Mawgoud *et al.*, 2010). *Fucus* spp., *Padina* spp., *Laminaria* spp., *Sargassum* spp., and *Turbinaria* spp. are used as bio fertilizers in agriculture (Hong *et al.*, 2007). The seaweed

components such as macro and micro nutrients, amino acids, vitamins, auxins, cytokinins and ABA like growth substances affect cellular metabolism in treated plants leading to enhanced growth and crop yield. They serve as a source of organic matter, metabolic enhancers and biostimulants.

Materials and Methods

The seaweed *Sargassum wightii* was collected from Rastakadu coast in Kanyakumari District. The extract from the seaweed was prepared following the method of Bhosle *et al.* (1975). 500gm of the seaweed was taken and washed well to remove sand and other debris. 500 ml of distilled water was added and they were boiled separately for one hour and filtered. The filtrate was taken as 100 % concentration of the SLF. From these different concentrations of the extract needed for the experiment was prepared by dilution. The seeds of *Sorghum* were surface sterilized with 1% HgCl₂ and soaked in distilled water for 12 hours. These seeds were sown in seven pots (grown in triplicates) and the treatments (100mM, 200mM and 300mM of NaCl and 1% *S. wightii* extracts) were given in triplicates to the plants. Salt stress was induced on the 15th day of sowing. To the SLF treated plants, along with the salt treatment, treatment with 1% *S. wightii* extracts was also given. The SLF extracts were poured into the soil as soil conditioners. The data were collected after the 30th and 45th day of sowing.

Table-1. Various treatments and supplementation followed in the experiments

Label	Treatments
C	Control
T1	NaCl (100mM)
T2	NaCl (200mM)
T3	NaCl (300mM)
T1+S	NaCl (100mM) +Sargassum Extracts @ 1%
T2+S	NaCl (200mM) +Sargassum Extracts @ 1%
T3+S	NaCl (300mM) +Sargassum Extracts @ 1%

The details of treatments are given in Table-1.

The physical and biochemical parameters were estimated on 30th and 45th day. The parameters studied are Leaf length, Shoot length, Leaf breadth, Fresh weight, Dry weight, Chlorophyll and Carotenoid pigments (Arnon, 1949), Protein (Lowry *et al.*, 1951), Amino acids (Moore and Stein, 1948) and Carbohydrates (Miller, 1972).

Results

In the present study, *Sorghum* plants were treated with three different concentrations of NaCl solution. Two sets of treatment were given along with control, one without 1% of SLF (*S. wightii* extracts) and the other with the SLF treatment. The salt treatment of *Sorghum* plants caused symptoms of phytotoxicity. The T3 plants were unable to survive in 300mM of salinity and they wilted on the 42nd day after sowing of the seeds. However the T3 plants treated with SLF did not wilt and survived till the end of the experiment. The results show that salt stress significantly affects the physical parameters (Table-2). The Shoot length, leaf length, leaf breadth; fresh weight and dry weight are affected. The length of the shoot was more in salt stressed plants than in SLF treated plants and control in 30th and 45th day. It ranged between 5 cm-12 cm. Leaf length was more in T3+S. The plant height varied from 17 to 37 cm. The length of the leaf was more in salt stressed plants and control than SLF treated plants on 30th day. On 45th day, the length of the leaf was more in SLF treated plants than salt treated and control. Leaf breadth was more in control for both 30th and 45th day. Leaf breadth was more in T1+S in 30th day and T2+S in 45th day. It ranged between 0.38-1.26 cm. Fresh weight was more in

control on 30th day and T3+S on 45th day. Compared to salt treated plants, fresh weight was more in SLF treated plants and it ranged between 0.11-0.27 gm. The dry weight of SLF treated plants was more when compared to salt stressed plants. The control plants had increased amount of dry weight which ranged between 0.021-0.071gm.

The results of the analysis of pigment parameters are given in Table-3. The amount of Chlorophyll a and total chlorophyll were more in control in both 30th and 45th day. The chlorophyll a content ranged between 1.73-2.22 and 1.68-2.35 mg g⁻¹FW on 30th and 45th day respectively in salt stressed plants and in SLF treated plants it ranged between 1.74-2.24 and 1.68-2.25 mg g⁻¹FW on 30th and 45th day respectively. Chlorophyll b was more in salt stressed plants than SLF treated plants. Salt stressed plants ranged between 0.52-0.73 mg g⁻¹FW and 0.67-0.87 mg g⁻¹FW in SLF treated plants on 30th day. Total chlorophyll in salt stressed and SLF treated plants were 2.45-3.34 and 2.45-2.91 mg g⁻¹FW respectively on 30th day. With an average of 2.54 and 0.63 mg g⁻¹FW total chlorophyll were present in SLF treated and salt treated plants on 45th day. The amount of carotenoids was more in T2+S and less in control. It ranged between 800-1200 and 800-950 mg g⁻¹FW in salt stressed and SLF treated plants respectively on 30th day. Both in salt treated and SLF treated plants, carotenoid ranged between 160-170 mg g⁻¹FW on 45th day.

The results of the biochemical parameters are given in Table 4. The carbohydrates ranged between 24-27 and 20-27 mg g⁻¹FW in salt treated and SLF treated plants on both 30th and 45th day. Carbohydrate was more in T2 and T2+S in 30th day and less in T3+S. On 45th day, it was more in T3+S and less in T1+S. The amount of amino acid ranged between 15-30 mg g⁻¹FW on both 30th and 45th day. On 45th day, it ranged between 10-25 and 30-40 mg g⁻¹FW in SLF treated and salt treated plants. The maximum amount of amino acid was seen in T2. On 30th day, protein was more in control and it ranged between 150-180 mg g⁻¹FW and 120-160 mg g⁻¹FW in salt stressed plants and SLF treated plants respectively. Protein content in salt stressed plants ranged between 80-103 mg g⁻¹FW and 50-120 mg g⁻¹FW in SLF treated plants on 45th day and more in T1+S.

Table-2. Physical Parameters of Salt Stressed *Sorghum vulgare* on 30th and 45th Day

	Shoot Length (cm)		Leaf Length (cm)		Leaf breadth (cm)		Fresh Weight (g)		Dry Weight (g)	
	30 th day	45 th day								
Control	5.74	11.44	21.64	27.25	0.82	1.2	0.16	0.22	0.031	0.071
T1	6.21	10.28	25.33	26.65	0.38	1.04	0.14	0.24	0.031	0.064
T2	6.16	12.76	22.48	22.35	0.77	1.15	0.11	0.098	0.022	0.041
T3	5.84	—	22.2	—	0.49	—	0.13	—	0.031	—
T1+S	6.02	9.19	23.9	26.6	0.69	1.18	0.15	0.26	0.030	0.066
T2+S	6.03	8.29	22.4	23.9	0.65	1.26	0.14	0.27	0.021	0.063
T3+S	5.9	8.12	17.5	37.8	0.45	1.25	0.11	0.22	0.027	0.054

Table 3. Effect of *Sargassum wightii* on the pigment parameters of NaCl treated Sorghum on 30th and 45th day

S.No.	Treatment	Chlorophyll a (mg/g FW)		Chlorophyll b (mg/g FW)		Total chlorophyll (mg/g FW)	
		30th	45th	30th	45th	30th	45th
1	C	2.48	2.35	0.86	0.76	3.34	3.11
2	T1	2.22	2.11	0.73	0.66	2.95	2.77
3	T2	1.96	2.07	0.61	0.63	2.57	2.71
4	T3	1.73	-	0.52	-	2.25	-
5	T1+S	2.24	2.24	0.67	0.57	2.91	2.75
6	T2+S	2.01	2.06	0.87	0.67	2.88	2.73
7	T3+S	1.75	1.68	0.70	0.63	2.45	2.31

Table 4. Effect of *Sargassum wightii* on the biochemical parameters of NaCl treated Sorghum on 30th and 45th day

Carotenoids (mg/g FW)		Carbohydrates (mg/g FW)		Amino acids (mg/g FW)		Proteins (mg/g FW)	
30th	45th	30th	45th	30th	45th	30th	45th
586.27	76.21	25.6	22.6	16	17	192	86.4
945.31	176.20	27.2	25.2	24	31	156	84.0
813.01	167.83	25.6	24.6	14	43	132	122.4
1192.90	-	23.2	-	26	-	180	-
855.20	107.23	25.6	20.6	22	21	156	120.0
1200.70	183.20	27.2	22.2	20	18	144	96.0
757.81	136.97	22.4	26.4	18	16	120	52.8

Discussion

Photosynthetic pigments are adversely affected by the high salt condition. Ion accumulation in leaves adversely affects the chlorophyll pigments. Parida and Das (2005) suggested that decrease in chlorophyll content in response to salt stress is a general phenomenon which leads to disordering of chlorophyll and appearance of chlorosis in plants. The observed decrease in chlorophyll content in the plants grown under saline conditions may be attributed to both the increased degradation and inhibited synthesis of the pigment (Garcia-Sanchez *et al.*, 2002). Carbohydrates play a lead role as osmoprotectants in carbon storage and radical scavenging. Salinity stress induced accumulation of total soluble sugars and sucrose in the leaves of salt sensitive cultivars without a concomitant increase in the activity of sucrose phosphate synthase. Protein induced by salinity may cause some alteration in cytoplasmic viscosity of the cell. Singh *et al.* (1987) reported that K plays an important role in plants in the transfer of nitrate from roots to shoots and leaves. Proteins that accumulate in plants grown under saline conditions may provide a storage form of nitrogen that is reutilized when stress is over and may play a role in osmotic adjustment. Amides such as glutamine and asparagine have been reported to accumulate in plants subject to salt stress. Among accumulated amino acids, proline may be of special interest because of its proposed role in plant salt tolerance (Mansour, 2000). This suggests that the amino acids are present in response to salt

stress. As stated by Munns (2003), suppression of plant growth under saline conditions may either be due to decreased availability of water or to the toxicity of sodium chloride. Also, the reduction in dry weight under salinity condition may be attributed to inhibition of hydrolysis of reserved foods and their translocation to growing shoots.

The ability to limit Na⁺ transport into the shoots and to reduce the Na⁺ accumulation in the rapidly growing shoot, is critically important for maintenance of high growth rates and protection of the metabolic process in elongating cells from the toxic effects of Na⁺. The increase in shoots characteristics might be due to the auxin content in the seaweed extracts which have an effective role in cell division and enlargement. This leads to an increase in shoot growth and plant fresh and dry weights (Gollan and Wright, 2006). The enhancing of vegetative growth can be due to seaweed components such as macro and microelement nutrients, amino acids, vitamins, cytokinins, auxins, and abscisic acid (ABA) like growth substances affecting cellular metabolism in plants leading to enhanced growth (Durand *et al.*, 2003; Stirk *et al.*, 2003). This positive effect might be due to the mineral content of Zn, Cu and B in the seaweed extracts, which have a great role in cell division and enlargement (Lopez *et al.*, 2008). It might also be due to the macronutrient content in seaweed extracts like nitrogen, potassium and phosphorous, which have a great role in plant nutrition (Attememe, 2009).

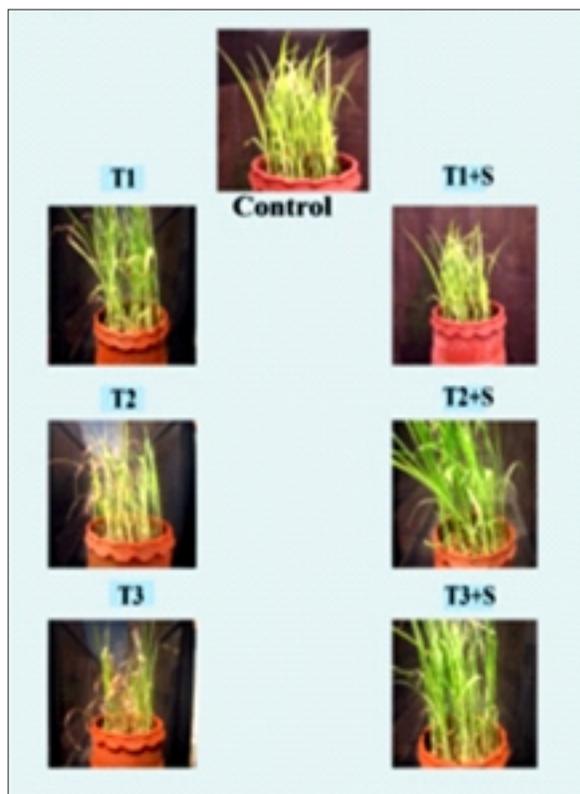


Plate-1. NaCl stressed Sorghum plant treated with Sargassum wightii extracts on 30th day

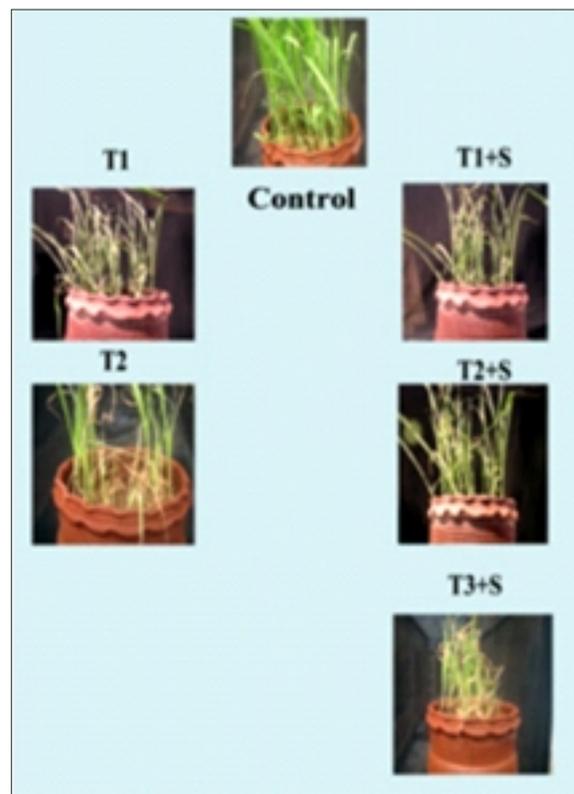


Plate-2. NaCl stressed Sorghum plant treated with Sargassum wightii extracts on 45th day

The effect of salinity on the biochemical parameters of *Sorghum vulgare* was studied in this investigation. The seeds were germinated in pots in garden soil. They were watered with distilled water till the 15th day of sowing. After that it was treated with NaCl on alternative days to induce salt stress. Along with the saline treatment, 1% of SLF treatment was also given. The T3 plants could not withstand the salinity after the 42nd day and are withered. However, T3 plants supplemented with SLF survived till the end of the experiment. The biochemical parameters such as pigment content, carbohydrates content, protein content, amino acid content were estimated on 30th and 45th day. A decrease in chlorophyll content and an increase in carotenoid content was observed in the salt stressed plants. The carotenoid content decreased at values closer to the control values in seaweed treated plants. Photosynthetic pigments were badly affected by the high salt condition. The carbohydrate content of the salt stressed plants was found to be higher than the control plants especially on the 45th day. The amino acid content was found to have increased across all treatments and all concentrations. However the protein content was observed to be highest in the control plants. It is evident from this study that the application of seaweed liquid extracts has some effect on the mechanism of salinity tolerance in *Sorghum*.

References

- Abdel-Mawgoud, A.M., F. Le'pine and E. De'ziel 2010. Rhamnolipids: diversity of structures, microbial origins, and roles. *Appl. Microbiol. Biotechnol.*, 86:1323–1336.
- Alam, S.M., A. Ansari and M.A. Khan 2000. Nuclear Institute of Agriculture, Tando Jam. *Industry and Economy*, 19-20: 8-2.
- Arnon, D.I. 1949. Copper enzyme polyphenoloxides in isolated chloroplast in *Beta vulgaris*. *Plant Physiology*, 24: 1-15. [2].
- Ashraf, M. 2002. Salt tolerance of cotton: Some new advances. *Crit. Rev. Plant Sci.*, 21: 1–30.
- Ashraf, M. 1994. Breeding for salinity tolerance in plants. *Crit. Rev. Plant Sci.*, 13:17-42.
- Attememe, J.Y.A. 2009. The effect of humic acid and seaweed extracts on the growth, chemical characteristics *Rosmarinus officinalis* L. The 6th scientific conference, Biology Dept., College of Education, University of Tikrit. *Plant Science*, 1-17.
- Bhosele, N. B., V.K. Dhargalkar and A. K. Untawale 1975. Effect of seaweed extract on the growth of *Phaseolus vulgaris* L. *Indian J. Mar. Sci.*, 4:208-210.
- Chen, C., C. Tao, H. Peng and Y. Ding 2007. Genetic analysis of salt stress responses in asparagus bean (*Vigna unguiculata* L. ssp. *Sesquipedalis* verdc.). *J. Hered.*, 98 (7): 655–665.
- Cuin, T.A. and S. Shabala 2007. Compatible solutes reduce ROS-induced potassium efflux in *Arabidopsis* roots. *Plant, Cell & Environment*, 30 (7): 875–885.

- Durand, N., X. Briand and C. Meyer 2003. The effect of marine bioactive substances (NPRO) and exogenous cytokinins on nitrate reductase activity in *Arabidopsis thaliana*. *Plant Physiol.*, 119: 489–493.
- Garcia-Sanchez, F., J.L. Jifon, M. Carvajal and J.P. Syvertsen 2002. Gas exchange, chlorophyll and nutrient contents in relation to Na⁺ and Cl⁻ accumulation in 'Sunburst' mandarin grafted on different rootstocks. *Plant Sci.*, 162: 705–712.
- Gollan, J.R. and J.T. Wright 2006. Limited grazing by native herbivores on the invasive seaweed *Caulerpa taxifolia* in a temperate Australian estuary. *Australia Estuary Marine and Fresh Water Research*, 57(7): 685-694.
- Hong, G., G. Heygster, J. Miao and K. Kunzi 2005. Detection of tropical deep convective clouds from AMSU-B water vapor channels measurements. *J. Geophys. Res.*, 110: D05205, doi:10.1029/2004JD004949.
- Lopez, R., F. Cabrera, E. Madejan, F. Sancho and M. Alvares 2008. Urban compost as an alternative for peat in forestry nursery growing media. *Dynamic soil-Dynamic plant*. 1: 60-66.
- Lowry, O. H., N.J. Rosebrough, A.L. Farr and R.J. Randall 1951. Protein measurement with the Folin phenol reagent. *J. Biol. Chem.*, 193:265–275.
- Mansour, M.M.F. 2000. Nitrogen containing compounds and adaptation of plants to salinity stress. *Biologia Plantarum*, 43(4): 491-500.
- Marschner, H. 1995. *Mineral Nutrition in Plants*. San Diego, CA (USA), Academic. 2nd Ed.
- Miller, J. H. 1972. *Experiments in molecular genetics*. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.
- Moore, S. and W.H. Stein 1948. Photometric ninhydrin method for the use in the chromatography of amino acid. *J. Biol. Chem.*, pp.176-367.
- Munns, R., 2003. Comparative physiology of salt and water stress. *Plant Cell Environ.*, 25: 239-50.
- Parida, A.K. and A.B. Das 2005. Salt tolerance and salinity effect on plants: a review. *Ecotoxicol. Environ. Saf.*, 60: 324–349.
- Singh, N.K., C.A. Bracken, P.M. Hasegawa, A.K. Handa, S. Buckel, M.A. Hermodson, F.P. Fankoch, F.E. Regniern and R.A. Bressan 1987. Characterization of osmotin. A thaumatin-like protein associated adjustment in plant cells. *Plant Physiol.*, 85:529-536.
- Stirk, W.A., M.S. Novak and J. Van Staden 2003. Cytokinins in macroalgae. *Plant Growth Regul.*, 41:13–24.
- Van Breusegem, F., E. Vranova, J.F. Dat and D. Inze 2001. The role of active oxygen species in plant signal transduction. *Plant Sci.*, 161: 405–414.