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Retrieval Efficacy of Seaweed Biomass on Cadmium Treated *Oryza sativa* L.

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ABSTRACT

Biosorption is an effective process for the removal and recovery of heavy metal ions from aqueous soAlutions. In the present investigation, naturally available plant based biosorbents *viz.*, powdered biomass of *Sargassum wighti* was examined to find out their efficiency on the biosorption of heavy metals from aqueous solution. Retrival efficacy of different concentrations of seaweed biomass on *Oryza sativa* seedlings was examined. Application of sea weed powder accelerated the growth of the plants. Addition of seaweed caused more accumulation of chlorophyll a, b, carotenoid and class of accessory pigments than the cadmium alone treated seedlings. The impact of seaweed on the biochemical constituents in terms of soluble sugar, free amino acid, soluble proteins and leaf nitrate was studied. Further, the scavenging enzymatic activities were decreased with the increased concentration of seaweed biomass reveals the stress relieving nature of plants.

Keywords : *Cadmium, Seaweed biomass, Sargassum wightii, Retrieval efficacy, Oryza sativa* L.

INTRODUCTION

Pollution of environment by toxic metals arises as a result of various industrial activities and has turned these metal ions into major health issues.Intensive crop cultivation requires the use of chemical fertilizers.The chemical fertilizers deteriorate soil quality and disturb the homeostasis of ecosystem. The toxic chemicals from the chemical fertilizers accumulate in the plants and plant products causing health problems to humans by biomagnifications. Plants growing in metal polluted locations exhibit altered metabolism,growth reduction,lower biomass production and metal accumulation and these functions are of human health concern. Conventional remediation methods of heavy metal contaminated soils are expensive and environmentally destructive (Bio-wise, 2003; Aboulroos *et al.,* 2006). It involves the removal of metals from polluted soils by transportation to laboratories, soil washing with chemicals to remove metals and finally replacing the soil at its original location. This decontamination strategy is an *ex situ* approach and can be very expensive and damaging the soil structure and ecology (Salt *et al.,* 1995). So, the development and implementation of eco-friendly and cost-effective process for removal / recovery of metal is essential to minimize the environmental hazard of toxic metals.

Biological methods such as biosorption / bioaccumulation for the removal of heavy metal ions may provide an attractive, eco-friendly alternative to physico-chemical methods. This involves the use of biosorbents prepared from naturally abundant and / or waste biomass. Due to the high uptake capacity and very cost effective source of the raw material, biosorption is a progression towards a perspective method. In the present investigation dried algal biomass of *Sargassum wightii* was utilized as biosorbents for the removal of heavy metal pollutants from synthetic metal solution.

The brown algae *Sargassum* has been shown to possess the required mechanical properties, chemical affinity and sorption capacity to bind metals such as cadmium in an effective, reversible and cost effective manner (Davis *et al.,* 2004). *Sargassum* is a brown seaweed that contains alginate with abundant carboxylic groups capable of capturing cations present in solution with a high sorption potential (Yang and Volesky, 1999;Mahajan and Kaushal,2018).

Seaweeds are the macroscopic marine algae found attached to the bottom in relatively shallow (coastal water) they grow in the intertidal, shallow and deep sea areas up to 1 meter depth and also in estuaries and back water on the solid substrate such as rook, dead corals and peoples, seaweed zone is one of the in conspicuous and wide spread biotype in the shallow marine environment the seaweeds are totally different from higher plants as they neither have the leaves, stems and roots or vascular systems none specialized sex organs. Their of seaweed is a manure in farming practices is very ancient and was prevalent among the Roman and also practiced in Britain, France, Spain, Japan and China.The global heavy metal pollution is increasing in the environment is due to increase in number of industries. Many industrial waste water contain heavy metals like cadmium, Lead, Zinc, Cobalt and Chromium. Cadmium (Cd) was the most abundant pollutant creating phyto toxicity while public health was mostly endangered by the presence of the toxic metals. Besides, Zinc (Zn), Nickel(Ni) Copper(Cu) and Lead(Pb) were also present in the contaminated sandy soil. The present study was aimed to assess the retrieval efficacy of seaweed biomass on cadmium treated *Oryza sativa*,L. seedlings.

MATERIAL AND METHODS

Oryza sativa L. Var. ASD - 16 of the family Poaceae was chosen to study the effect of various concentrations of cadmium on the growth and biochemical characteristics under stressful condition.The experimental soil for raising the cultivars was prepared by mixing red soil, black soil and sandy soil in the ratio of 1 : 1 : 1. The prepared soil was sterilised by solar sterilization method for 5 days. The analysed soil medium was taken in earthen pots of size 30×33 cm and filled in about two-third of its height (5 kg of soil per pot).

Seeds of *Oryza sativa* L. ASD-16 was procured from Agricultural Extension Centre,Tirunelveli. Healthy seeds of *Oryza sativa* L. were surface sterilized with 0.1% mercuric chloride (w/v) for one minute and washed with running tap water followed by distilled water.

The seaweed used in the present study is *Sargassum wightii*, and were collected from Rasthakad coast (Lat.8 6 3 N) in Kanniyakumari of south Tamilnadu. They were hand picked and washed thoroughly with sea water and finally with fresh water to remove all unwanted impurities. The seaweed biomass was shade dried and finally powdered.

The dry powder of various concentrations of seaweed biomass was inoculated separately to the synthetic solutions of cadmium (6mM), the concentration at which the toxicity was found to be at optimum level. The concentrations of plants biomass used were 2, 4 and 8 g in 100 mL of 6 mM heavy metal solution prepared separately in a conical flask. It was kept under mechanical shaking using rotary shaker for 48 hours. After that, all the plant biomass was filtered through filter paper. The filterate was applied in the experimental pots separately at the time showing and on every alternate day of plants growth. Five replicates along with suitable control (water) and Control (6mM Cadmium chloride) plants were also maintained. The 30 days old plants were analysed for its stress relief with the aid of growth and biochemical changes.

Growth parameters such as seedling length, Fresh weight, dry weight and leaf area, photosynthetic pigments (Wellburn&Lichtenthaler,1984) Glucose (Jayaraman,1981) Protein (Lowry,*et.al*.,1951) Amino acid (Jayaraman, 1981) Proline (Bates, *et.al*.,1973) Leaf nitrate (Cataldo, *et.al*., 1978) and enzymatic activities such as Nitrate reductase(Jaworski, *et.al.*, 1971) Catalase(Kar and Mishra,1976) and Peroxidase(Addy and Goodman, 1972) were analysed. Statistical analysis (One way ANOVA – Tukey test) was done by using the statistical package, origin – version 7.0

RESULT AND DISCUSSION

The result obtained on the present study indicated that, increase in concentration of cadmium chloride results in decrease in the shoot length and root length. Foy *et al.*, (1978) and Alam and Adams (1979) proved that the dry matter yield of tops and roots of oats decreased with increased zinc chloride level. Zinc chloride reduces the possibility of successful seedlings establishment was observed in germinating grass seeds during the revegetation in acid soil in which concentration of zinc and other metals very high (Winter halder, 1985). After the application of various concentration of plant biomass, the growth and biochemical characters were enhanced when compared to control plants. Amendment materials have always been recognized as important for improvement and restoration of physical condition of alkaline soil (Mahajan and Kaushal,2018). Application of plant biomass to the soil, increases the soil micro organisms, which in turn, the polluted soil in to a usable one. These organisms produce CO_2 in the soil atmosphere, which dissolves in water to form weak carbonic acid. This acid dissolves most of the insoluble soil materials, there by making them available to plants as nutrients. Thus, phycobial consortium makes the soil fertile (Swarnalata Mishra and Tiwari, 1993).

There is an increase in shoot, root length and biomass accumulation. It showed a significant increase than the control plants. These amendments also caused increase in pigment contents than the control plants. The increase in seedling growth may be due to phenyl acetic acid (PAA) and other closely related compounds present in the seaweed extract as well as the presence of some growth promoting substances. Micro and macro elements present in the seaweed extract were responsible for the enhancement of the growth of wheat plants (Beckett and Van staden, 1990). In the present study, both the leaf area and total chlorophyll are found to increase with the concentrations of amendments. This indicated that, the phycobial consortium enhanced not only the leaf area but also the content of photosynthetic pigments which finally led to increase in the plant biomass. After amendment application, there is an increase in the chlorophyll and carotenoid content and reduction in the anthocyanin content. The impact of seaweed extract on the biochemical constituents was studied of free amino acids, soluble protein and starch content. The level of free amino acid content was high in treated seedlings. The accumulation of soluble protein in leaves are considered to be an indicating of efficient metabolic status. The Phycobial consortium caused significant increase in the photosynthetic pigment. Their increase in chlorophyll content may be due to a decrease in chlorophyll degradation. Yield enhancement effects due to improved chlorophyll content.

The rate of photosynthesis is directly proportional to the amount of chlorophyll content. This may be due to the cadmium induced inhibition of electron transport system of the isolated chloroplast. Cadmium may directly affect the metabolism in shoot, (Schnabl, 1976). Cadmium induce mineral deficiency and blocks the reduction of Fe_3 to Fe_2 in the growth medium (Alams and Adams, 1979). The plants which were treated with Phycobial consortium shows high chlorophyll content then control observed in the present study can be attributed any one of the above reasons. High concentration of Cd may directly affect metabolism and translocation of carbohydrates. This may be due to cd which blocks the reduction of Fe^{3+} to Fe^{2+} in young leaves reflects the immobility of Fe in plants and its prime need for chlorophyll synthesis. In this experiment, level of Total soluble sugar was exhibit high when compared to $CdCl_2$ treated control plants. The result obtained on the protein content also revealed a increase in protein content in plants treated with increasing concentration of Phycobial consortium. The same result was observed by Ghilidiyal *et al*., (1986) in Linn seeds. The elevation in protein may be due to interference of Phycobial consortium with the enzyme activity, pigments synthesis, photo synthesis and nitrogen metabolism (Beanford *et al.*, (1977). The increase in sugar content might be due to increase in net photosynthesis rate at high concentration of macroalgae (Keul *et al*., 1979). Proline accumulates in the leaves of any plants, when the plants are subjected to stress (Paleg and Aspinal, 1981). It was observed in the present study also. The enhanced leaf proline content *viz.,* glutamate, through the activity of glutamate of protein (Greenway and Munns, 1980). The accumulation of proline is also considered as on adaptive response to stress (Stewart concentration of Phycobial consortium. Nitrate reductase (NR) is one of the cytoplasmic substrate inducible enzymes. The NR activity is decreased in cd treated plants. On contrary, high concentration of biostimulant, there was increase in the NR activity. Observed reduction of leaf nitrate content after various concentration of Phycobial consortium treatment indicates the increasing of NR activity.

Peroxidase are enzyme which utilize hydrogen peroxide to oxidize a wide range of oxygen donors such as phenolic substances, cytochrome etc., as reported by Scandalior (1974). In the present study, an enhanced peroxidase activity with the increase in the concentration of cadmium chloride was observed. where as, during Phycobial consortium treatment, activity of peroxide was reduced and it is revealed that, it rectify the stressful effect caused by cadmium chloride in experimental plants.

CONCLUSION

The effect of different concentration of seaweed biomass extract accelerated the growth of the plants. On the fresh weight basis, the addition of seaweed caused more accumulation of chlorophyll a, b, Carotenoid, class of accessory pigments was also high in Phycobial consortium treated seedlings than the cadmium chloride alone treated seedlings.

The impact of seaweed biomass on the biochemical constituents in terms of free amino acid, soluble proteins and glucose was studied. The level of free amino acid content was high in treated seedlings. The accumulation of soluble protein in leaves are considered to be an indication of efficient metabolic status than its counter parts. Further, the scavenging enzymatic activities were decreased in increased concentration of seaweed biomass reveals the stress reliving nature of plants. Thus an overall assessment of plant in terms of growth, pigment composition, biochemical constituents and enzymatic activities has exhibit the retrieval efficacy of plant based biosorbents.

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Table: 1 Effect of *Sargassum wightii* biomass treated cadmium (6 mM) on the growth of Oryza sativa L.

Sl. No.	Parameters	Con I (Water)	Con II (6 mM Cadmium)	2% (w/v)	4% (w/v)	8% (w/v)
1	Seed germination capacity (%)	98 ± 2.00	78 ± 2.00 [a]	86 ± 2.449 [a]	96 ± 2.449 [b]	98 ± 2.00 [b]
2	Root length (cm)	11.76 ± 0.107	8.340 ± 0.102 [a]	9.420 ± 0.128 [b]	10.18 ± 0.115 [c]	11.44 ± 0.102 [d]
		(100)	(71)	(80)	(86)	(97)
3	Shoot length (cm)	31.44 ± 0.307	28.36 ± 0.092 [a]	28.80 ± 0.137 [a]	30.24 ± 0.120 [b]	31.14 ± 0.136 [c]
		(100)	(90)	(92)	(96)	(99)
4	Leaf area (cm^2)	6.796 ± 0.108	5.258 ± 0.059 [a]	6.048 ± 0.094 [b]	6.342 ± 0.089 [bc]	6.650 ± 0.039 [c]
		(100)	(77)	(89)	(93)	(98)
5	Fresh weight (g)	2.838 ± 0.032	2.407 ± 0.020 [a]	2.572 ± 0.016 [b]	2.625 ± 0.007 [b]	2.785 ± 0.015 [c]
		(100)	(85)	(91)	(92)	(98)
6	Dry weight (g)	1.148 ± 0.009	0.831 ± 0.011 [a]	0.951 ± 0.017 [b]	1.105 ± 0.026 [c]	1.226 ± 0.019 [d]
		(100)	(72)	(83)	(96)	(107)

Values are an average of five observation Values in parentheses are percentage activity with respect to control. Mean ± SE
Superscripts of different alphabets of the same parameter indicate significant variations (P□□ 0.05 - Tukey test).

Table: 2 Effect of *Sargassum wightii* biomass treated cadmium (6 mM) on the pigment content of *Oryza sativa* L.

Sl. No.	Pigments	Con I (Water)	Con II (6 mM Cadmium)	2% (w/v)	4% (w/v)	8% (w/v)
1	Chlorophyll *a* (mg/g LFW)	2.235±0.036	1.720±0.013 ^{a}	1.912±0.012 ^{b}	2.040±0.036 ^{bc}	2.160±0.041 ^{c}
		(100)	(77)	(86)	(91)	(97)
2	Chlorophyll *b* (mg/g LFW)	1.112±0.025	0.634±0.014 ^{a}	0.762±0.021 ^{b}	0.893±0.019 ^{c}	0.971±0.027 ^{c}
		(100)	(57)	(69)	(75)	(87)
3	Total Chlorophyll (mg/g LFW)	3.347±0.060	2.354±0.021 ^{a}	2.674±0.025 ^{b}	2.934±0.053 ^{c}	3.131±0.025 ^{d}
		(100)	(70)	(80)	(88)	(94)
4	Carotenoids (mg/g LFW)	1.325±0.034	0.916±0.014 ^{a}	1.012±0.024 ^{ab}	1.107±0.0178 ^{b}	1.241±0.024 ^{c}
		(100)	(69)	(76)	(84)	(94)
5	Anthocyanin (mg/g LFW)	1.456±0.025	1.938±0.013 ^{a}	1.781±0.040 ^{b}	1.663±0.025 ^{c}	1.528±0.021 ^{d}
		(100)	(133)	(122)	(114)	(105)

Values are an average of five observation..Values in parentheses are percentage activity with respect to control. Mean ± SE
Superscripts of different alphabets of the same parameter indicate significant variations (P □ 0.05 - Tukey test).

Table: 3 Effect of *Sargassum wightii* biomass treated cadmium (6 mM) on the biochemical characteristics of *Oryza sativa L*

Sl. No.	Biochemical Parameters	Con I (Water)	Con II (6 mM Cadmium)	2% (w/v)	4% (w/v)	8% (w/v)
1	Total Soluble Sugar (mg/g LFW)	42.58±0.177	35.54±0.156 ^{a}	38.76±0.249 ^{b}	41.18±0.126 ^{c}	42.94±0.211 ^{d}
		(100)	(83)	(91)	(97)	(101)
2	Total Soluble Protein (mg/g LFW)	29.20±0.294	23.86±0.246 ^{a}	25.67±0.144 ^{b}	27.17±0.178 ^{c}	28.63±0.154 ^{d}
		(100)	(82)	(88)	(93)	(98)
3	Amino acid (□ mole/g LFW)	5.320±0.149	7.386±0.044 ^{a}	5.972±0.045 ^{b}	5.684±0.058 ^{b}	5.327±0.042 ^{c}
		(100)	(139)	(120)	(107)	(100)
4	Proline (mg/g LFW)	159.8±0.990	178.98±0.321 ^{a}	170.40±0.245 ^{b}	165.39±0.367 ^{c}	161.15±0.398 ^{d}
		(100)	(112)	(107)	(103)	(101)
5	Leaf nitrate (mg/g LFW)	60.14 □ 0.181	68.13 □ 0.257 ^{a}	66.04 □ 0.218 ^{b}	63.97 □ 0.319 ^{c}	61.96 □ 0.271 ^{d}
		(100)	(113)	(110)	(106)	(103)

Values are an average of five observation. Values in parentheses are percentage activity with respect to control. Mean ± SE
Superscripts of different alphabets of the same parameter indicate significant variations (P□□ 0.05 - Tukey test).

Table: 4 Effect of *Sargassum wightii* biomass treated cadmium (6 mM) on the enzyme activities of *Oryza sativa* L.

Sl. No.	Enzymes	Con I (Water)	Con II (6 mM Cadmium)	2% (w/v)	4% (w/v)	8% (w/v)
1	Nitrate reductase activity (□ mole/g LFW)	19.36±0.039	15.30±0.016 ^{a}	16.46±0.0116 ^{b}	17.83±0.083 ^{c}	19.66±0.147 ^{d}
		(100)	(79)	(85)	(92)	(102)
2	Catalase activity (□ mole/g LFW)	0.539±0.008	0.941±0.008 ^{a}	0.844±0.016 ^{b}	0.754±0.010 ^{c}	0.534±0.016 ^{d}
		(100)	(175)	(157)	(140)	(99)
3	Peroxidase activity (□ mole/g LFW)	3.544±0.032	4.207±0.022 ^{a}	4.115±0.048 ^{a}	3.827±0.039 ^{b}	3.512±0.038 ^{c}
		(100)	(119)	(116)	(108)	(99)

Values are an average of five observation. Values in parentheses are percentage activity with respect to control. Mean ± SE
Superscripts of different alphabets of the same parameter indicate significant variations (P □ 0.05 - Tukey test).