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NULLIFYING THE TOXICITY OF PLATE MAKING INDUSTRY EFFLUENT USING SEAWEED *ULVA LACTUCA*

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ABSTRACT

The Plate making industry effluent containing heavy metals that are toxic to plants, reduces the plant yield and even the soil fertility, although these toxic substances could be removed by plants and dried plant materials. The ameliorative effect of *Ulva lactuca* at different concentration viz., 2g, 4g and 6g/L in 60% concentrated Plate making industry effluent treated on *Cyamopsis tetragonoloba* Taub. was analyzed. Impact of effluent was understood by a steep decline in growth characters, pigment content and other biochemical characteristics. The powder of *Ulva lactuca* in 60% concentration of the effluent in different concentrations brought about considerable increase in germination percentage, growth and biochemical characteristics in *Cyamopsis tetragonoloba* Taub than when they were treated with Plate making industry effluent alone. Seaweed dry powder used in this study was found to be efficient in nullifying the toxicity of effluent on growth and biochemical characteristics of *Cyamopsis tetragonoloba* Taub.

Key Words: Bioadsorption, Seaweeds, Plant Stress, Plate Making Industry Effluent

INTRODUCTION

Water is indispensable and the most precious on the earth. Everything originated in water and everything is sustained by water. The disposal waste water from the industries has a serious havoc as receiving river stream has become coloured. Sivakasi is an industrial town in Virudhunagar district. The town alone has nearly 250 fireworks, 200 litho and offset, 700 match units and 150 other industries including dye, printing ink, chemical cardboard, metal plate making and chemical plate grinding industries. Quite a large number of chemicals have constantly used for manufacturing process (Ramasubramanian *et al.*, 2004). The industrial effluents contain especially inorganic content and colour, which impart objectionable colour to the water bodies. Presence of colour reduces the light penetration and photosynthetic activities of water bodies, some of the Plate makings used in the textile industries are found to be carcinogenic (Roshan Poi *et al.*, 2000).

These waste effluents can be treated by treatment methods like ozonation, chemical coagulation, adsorption and electrochemical technology. These chemical treatment methods require costly chemicals and also generable hazardous sludge, so use of plants to degrade, assimilate, metabolize or detoxify contaminants is cost effective and ecologically sounds for the restoration and management of our natural water resources. There is an urgent need to apply bioadsorption technology, using dried natural algal biomass to decontaminate the polluted water bodies in the world. This will be effective in bringing new resources and technology to solve environmental problems in India, generated by industries (Selvarathi, 2010).

This study aimed to analyze the characterization of Plate making effluent, the effect of Plate making effluents on morphometric, biochemical and enzymatic characteristics of *Cyamopsis tetragonoloba* Taub. and also study the effect of varying amount of dried natural biomass of *Ulva lactuca* with 60% effluent on the morphometric, biochemical and enzymatic characteristics of *Cyamopsis tetragonoloba* Taub.

MATERIALS AND METHODS

The Plate making industry effluent was collected from the Plate making industry Sivakasi. The sample for analysis was preserved as per the standard recommended procedure (Rainwater and Thatcher, 1963). The

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seaweed *Ulva lactuca* collected from Harberpoint coast near Tuticorin were shade dried and finally powdered by milling. Various concentrations of seaweed powder was prepared with 60% Plate making industry effluent.

Both control and experimental seeds were allowed to grow in uniform mixed red, black and sandy soil in 1:1:1 ratio. After ten days, seedling of *Cyamopsis tetragonoloba* Taub. were treated with various concentration of Plate making industry effluent (20%, 40%, 60%, 80% and 100% v/v). After ten days of effluent treatment, various morphometric, biochemical and enzymatic characteristics were analyzed. In another set 60% of effluent (the concentration at which toxicity was found to be optimum level based on LST analysis (Zar, 1984). was subjected to various concentration of seaweed (*Ulva lactuca*) powder (2g/L, 4g/L and 6g/L w/v), for 24 hours. Then filtered and the filtrate was used to treat plants. After ten days of treatment, various morphometric, biochemical and enzymatic characteristics were analyzed.

Twenty days old plants of *Cyamopsis tetragonoloba* Taub. were used for measuring the morphometric characters such as root length, shoot length, leaf area, fresh weight and dry weight were measured. The biochemical and enzymatic characters were analyzed by the following methods: chlorophyll and carotenoids (Wellburn and Lichtenthaler, 1984), anthocyanin (Swain and Hills, 1959), Total soluble sugar (Jayaraman, 1981), Protein content (Lowry et al., 1951), amino acid content (Jayaraman, 1981), leaf nitrate (Cataldo et al., 1978), *in vivo* nitrate reductase activity (Jaworski, 1971), peroxidase and catalase activity (Kar and Mishra, 1976).

RESULTS AND DISCUSSION

The results obtained on the analysis of the physico – chemical parameters of Plate making industry effluent, the effect of different concentration of effluent and the effect of effluent + seaweed powder treated plants were summarized and discussed below.

The physico – chemical characters, analysis were tabulated in Table 1. The morphometric characters such as root length, shoot length, leaf area, fresh weight and dry weight decreased (Table 2) with increasing in the concentration of Plate making industry effluent. Similarly, chlorophyll, carotenoids (Table 3), total soluble sugar, protein (Table 4) and nitrate reductase activity (table 5) also showed declining in trend. In contrary, the anthocyanin (Table 3), leaf nitrate, free amino acid, proline contents (Table 4) and the activities of antioxidant enzymes such as peroxidase, catalase (Table 5) were increased.

Plate making industry effluent contained 88.35 mg/L of total dissolved solids and 164 mg/L of total hardness, which leads to root and shoot length inhibition at higher concentration of the effluent. The pronounced inhibition of shoot and root growth and leaf area were the main cause for the decrease in fresh and dry weight of seedlings.

Table 1: Physico – Chemical analysis of Plate Making Industry Effluent

S.No.	Parameters	BSI Standards	Value Characters
1.	Temperature	-	29.5 ⁰ C
2.	pH	7 – 8.5	11.53
3.	Electric Conductivity (EC)	400	17.49
4.	Total Dissolved Solids (TDS)	500	88.35
5.	Total Hardness	300	164
6.	Biological Oxygen Demand (BOD)	-	15
7.	Dissolved Corbandioxide	22.8	79

The inhibition of biomass accumulation is directly related to the photosynthetic process. At higher concentration, the effluent showed inhibitory effect on both photosynthetic pigments and total soluble sugar. Similar result was observed by Kumar, (1999) after the irrigations with effluent.

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Table 2: Impact of various concentration of Plate Making Effluent on the Morphometric characteristics of *Cyamopsis tetragonoloba* Taub

Growth	Control	20%	40%	60%	80%	100%
Root Length (cm)	12.75 ± 0.091 (100)	9.32 ± 0.150 (73)	7.62 ± 0.041 (60)	6.46 ± 0.094 (51)	4.58 ± 0.102 (36)	3.04 ± 0.134 (24)
Shoot Length (cm)	15.12 ± 0.155 (100)	12.62 ± 0.102 (83)	9.11 ± 0.106 (60)	7.53 ± 0.107 (50)	5.44 ± 0.102 (36)	4.36 ± 0.097 (29)
Leaf Area (cm ²)	11.43 ± 0.109 (100)	8.93 ± 0.088 (78)	6.96 ± 0.110 (61)	4.73 ± 0.039 (41)	3.78 ± 0.030 (33)	3.35 ± 0.015 (29)
Fresh Weight (gm)	1.12 ± 0.014 (100)	0.92 ± 0.050 (83)	0.77 ± 0.059 (70)	0.64 ± 0.037 (58)	0.51 ± 0.013 (46)	0.37 ± 0.091 (33)
Dry Weight (gm)	0.97 ± 0.040 (100)	0.79 ± 0.013 (82)	0.49 ± 0.015 (51)	0.28 ± 0.015 (29)	0.15 ± 0.043 (16)	0.10 ± 0.040 (10)

Values in parenthesis indicate percent activity; value represents mean of 10 samples with their standard error (±)

Table 3: Impact of various concentration of Plate Making Effluent on the photosynthetic pigments of *Cyamopsis tetragonoloba* Taub

Pigments	Control	20%	40%	60%	80%	100%
Chlorophyll (mg/gLFW)	.a 1.59 ± 0.080 (100)	1.16 ± 0.048 (73)	0.90 ± 0.060 (57)	0.72 ± 0.016 (45)	0.53 ± 0.180 (33)	0.45 ± 0.026 (28)
Chlorophyll (mg/gLFW)	.b 0.87 ± 0.016 (100)	0.65 ± 0.021 (75)	0.51 ± 0.080 (59)	0.40 ± 0.90 (46)	0.30 ± 0.60 (35)	0.25 ± 0.20 (29)
Total.Chlorophyll (mg/gLFW)	2.45 ± 0.021 (100)	1.807 ± 0.051 (74)	1.41 ± 0.014 (57)	1.12 ± 0.016 (46)	0.83 ± 0.022 (34)	0.70 ± 0.046 (29)
Carotenoids (mg/gLFW)	0.483 ± 0.011 (100)	0.391 ± 0.050 (81)	0.315 ± 0.016 (65)	0.247 ± 0.060 (51)	0.184 ± 0.109 (38)	0.162 ± 0.010 (33)
Anthocyanin (mg/gLFW)	1.43 ± 0.077 (100)	1.63 ± 0.015 (114)	1.75 ± 0.014 (122)	2.49 ± 0.025 (174)	3.17 ± 0.088 (221)	3.75 ± 0.133 (262)

Values in parenthesis indicate percent activity; value represents mean of 5 samples with their standard error (±)

The reduction in sugar content maybe attributed to reduction in chlorophyll content of the leaf and also a decline in protein. This change might have already affected the photosynthetic activity of the plant and hence reduction in contents (Swaminathan *et al.*, 1998; Dowton, 1997).

Accumulation of proline has been frequently used as biochemical marker for water stress in plants (Alia and Saradhi, 1991; Schat *et al.*, 1997). An increase in the aminoacid and proline content after Plate

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making, sugar industrial effluent treated has already been reported (Ramasubramanian *et al.*, 1993; Jeyarathi and Ramasubramanian, 2002). The leaf nitrate content was found to be more in effluent treated plants paralleling with the reduction in *in vivo* nitrate reductase activity.

Table 4: Impact of various concentration of Plate Making Effluent on the Biochemical characteristics of *Cyamopsis tetragonoloba* Taub

Parameters	Control	20%	40%	60%	80%	100%
Total soluble Sugar (mg/g LFW)	45.87 ± 0.88 (100)	34.03 ± 1.245 (74)	27.54 ± 0.754 (60)	24.05 ± 0.250 (52)	19.89 ± 0.145 (43)	16.69 ± 0.686 (36)
Total soluble Protein (mg/g LFW)	10.81 ± 0.157 (100)	9.20 ± 0.096 (85)	8.210 ± 0.168 (76)	7.25 ± 0.174 (67)	5.96 ± 0.044 (55)	4.213 ± 0.125 (39)
Amino acid (µM/g LFW)	1.61 ± 0.040 (100)	1.93 ± 0.027 (121)	2.31 ± 0.014 (144)	2.94 ± 0.038 (184)	4.50 ± 0.074 (281)	4.68 ± 0.212 (292)
Proline (µM/g LFW)	25.58 ± 0.060 (140)	36.32 ± 0.087 (142)	38.47 ± 0.678 (150)	42.47 ± 0.566 (166)	46.85 ± 0.635 (183)	48.94 ± 0.338 (191)
Leaf nitrate (mg/g LFW)	1.74 ± 0.011 (100)	2.24 ± 0.015 (129)	2.53 ± 0.070 (146)	2.92 ± 0.0475 (168)	3.52 ± 0.081 (203)	4.20 ± 0.157 (242)

Values in parenthesis indicate percent activity; value represents mean of 5 samples with their standard error (±)

Table 5: Impact of various concentration of Plate Making Effluent on the Enzymes activity of *Cyamopsis tetragonoloba* Taub

Parameters	Control	20%	40%	60%	80%	100%
Nitrate Reductase activity (µMole/g LFW)	0.54 ± 0.014 (100)	0.41 ± 0.060 (76)	0.35 ± 0.051 (64)	0.29 ± 0.048 (54)	0.24 ± 0.015 (43)	0.17 ± 0.085 (32)
Catalase activity (µMole/g LFW)	0.248 ± 0.068 (100)	0.307 ± 0.039 (124)	0.413 ± 0.023 (167)	0.517 ± 0.094 (208)	0.560 ± 0.070 (226)	0.667 ± 0.067 (269)
Peroxidase activity (µMole/g LFW)	4.24 ± 0.017 (100)	6.94 ± 0.030 (164)	8.55 ± 0.048 (202)	9.15 ± 0.020 (216)	10.55 ± 0.022 (249)	11.37 ± 0.023 (268)

Values in parenthesis indicate percent activity; value represents mean of 5 samples with their standard error (±)

In the present study an enhanced peroxidase activity was observed with the increase in the concentration of Plate making industrial effluent. Balasimha (1982) reported that peroxidase plays a vital role in IAA and chlorophyll degradation. Thus observed increase in peroxidase activity can be correlated with the observed reduction in chlorophyll content, fresh weight and biomass. Paper industrial effluent also caused similar increase in peroxidase activity of *Eleusine corocana* (Selvarathi *et al.*, 2006).

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Catalase is antioxidant and scavenging enzyme, it was found to be increased with the increasing concentration of Plate making industry effluent. Catalase is special type of peroxidase enzyme which catalyses the degradation of H_2O_2 , which is natural metabolism and also toxic to plants (Balasimha, 1982).

Table 6: Effect of Plate Making Effluent and *Ulva lactuca* on the Morphometric characteristics of *Cyamopsis tetragonoloba* Taub

Growth	Control (Water)	Control (Plate Making Effluent 60%)	Plate Making Effluent +		
			2 gm /L SWP	4 gm /L SWP	6 gm /L SWP
Root Length (cm)	12.75 \pm 0.091 (100)	6.46 \pm 0.094 (51)	6.85 \pm 0.055 (54)	9.25 \pm 0.132 (73)	10.96 \pm 0.140 (86)
Shoot Length (cm)	15.12 \pm 0.155 (100)	7.53 \pm 0.107 (50)	8.06 \pm 0.145 (53)	11.11 \pm 0.317 (73)	13.49 \pm 0.314 (89)
Leaf Area (cm ²)	11.43 \pm 0.109 (100)	4.73 \pm 0.039 (41)	5.55 \pm 0.112 (49)	8.31 \pm 0.143 (73)	10.40 \pm 0.63 (91)
Fresh Weight (gm)	1.12 \pm 0.014 (100)	0.64 \pm 0.037 (58)	0.743 \pm 0.017 (67)	0.89 \pm 0.180 (80)	1.067 \pm 0.072 (96)
Dry Weight (gm)	0.97 \pm 0.040 (100)	0.28 \pm 0.015 (29)	0.41 \pm 0.016 (42)	0.74 \pm 0.04 (76)	0.82 \pm 0.079 (84)

Values in parenthesis indicate percent activity; value represents mean of 10 samples with their standard error (\pm)

Table 7: Effect of Plate Making Effluent and *Ulva lactuca* on the photosynthetic pigments of *Cyamopsis tetragonoloba* Taub

Pigments		Control (Water)	Control (Plate Making Effluent 60%)	Plate Making Effluent +		
				2 gm /L SWP	4 gm /L SWP	6 gm /L SWP
Chlorophyll (mg/gLFW)	.a	1.59 \pm 0.080 (100)	0.72 \pm 0.016 (45)	0.85 \pm 0.021 (54)	1.04 \pm 0.044 (65)	1.45 \pm 0.112 (91)
Chlorophyll (mg/gLFW)	.b	0.87 \pm 0.016 (100)	0.40 \pm 0.90 (46)	0.57 \pm 0.016 (66)	0.73 \pm 0.020 (84)	0.85 \pm 0.028 (98)
Total. Chlorophyll (mg/gLFW)		2.45 \pm 0.021 (100)	1.12 \pm 0.016 (46)	1.42 \pm 0.033 (58)	1.77 \pm 0.041 (72)	2.30 \pm 0.084 (94)
Carotenoids (mg/gLFW)		0.483 \pm 0.011 (100)	0.247 \pm 0.060 (51)	0.302 \pm 0.023 (63)	0.404 \pm 0.040 (84)	0.461 \pm 0.010 (95)
Anthocyanin (mg/gLFW)		1.43 \pm 0.077 (100)	2.49 \pm 0.025 (174)	2.18 \pm 0.084 (152)	1.85 \pm 0.048 (129)	1.48 \pm 0.038 (104)

Values in parenthesis indicate percent activity; value represents mean of 5 samples with their standard error (\pm)

Bioadsorption studies showed that the morphometric features increased (Table 6) by the application of seaweed powder. The chlorophyll content increased (Table 7) with increasing amount of seaweed powder. This result coincides with the results of Ramasubramanian *et al.*, (2006). The total soluble sugar, protein content (Table 8) and nitrate reductase activity (Table 9) also increased after the application of seaweed powder. This may be due to the bioadsorption by plant biomass (*Ulva lactuca* powder) to remove / adsorb toxic elements which are present in effluent (Selvarathi *et al.*, 2010). In contrary, leaf

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nitrate, free amino acid and proline (Table 8) and the activity of enzymes such as catalase and peroxidase (Table 9) were found to be reduced after the application of seaweed in our studies. Our results are in accordance with findings of (Ramasubramanian *et al.*, (2006).

Table 8: Effect of Plate Making Effluent and *Ulva lactuca* on the Biochemical characteristics of *Cyamopsis tetragonoloba* Taub

Parameters	Control (Water)	Control Making 60%)	(Plate Effluent	Plate Making Effluent +		
				2 gm /L SWP	4 gm /L SWP	6 gm /L SWP
Total soluble Sugar (mg/g LFW)	45.87 ± 0.88 (100)	24.05 ± 0.250 (52)		28.81 ± 0.182 (63)	34.76 ± 0.672 (76)	41.08 ± 0.843 (90)
Total soluble Protein (mg/g LFW)	10.81 ± 0.157 (100)	7.25 ± 0.174 (67)		8.13 ± 0.049 (75)	9.77 ± 0.052 (90)	10.21 ± 0.168 (94)
Amino acid (µMole/g LFW)	1.61 ± 0.040 (100)	2.943 ± 0.038 (184)		2.29 ± 0.181 (143)	1.82 ± 0.086 (113)	1.7 ± 0.136 (106)
Proline (µMole/g LFW)	25.58 ± 0.060 (142)	42.47 ± 0.566 (166)		39.39 ± 0.399 (154)	30.08 ± 0.228 (118)	27.31 ± 0.731 (107)
Leaf nitrate (mg/g LFW)	1.74 ± 0.011 (100)	2.92 ± 0.0475 (168)		2.63 ± 0.059 (151)	2.20 ± 0.105 (127)	1.70 ± 0.081 (98)

Values in parenthesis indicate percent activity; value represents mean of 5 samples with their standard error (±)

Table 9: Effect of Plate Making Effluent and *Ulva lactuca* on the Enzymes activity of *Cyamopsis tetragonoloba* Taub

Parameters	Control (Water)	Control Making 60%)	(Plate Effluent	Plate Making Effluent +		
				2 gm /L SWP	4 gm /L SWP	6 gm /L SWP
Nitrate Reductase activity (µMole/g LFW)	0.54 ± 0.014 (100)	0.29 ± 0.048 (54)		0.35 ± 0.021 (65)	0.47 ± 0.017 (86)	0.56 ± 0.019 (103)
Catalase activity (µMole/g LFW)	0.25 ± 0.068 (100)	1.25 ± 0.094 (499)		0.86 ± 0.051 (348)	0.69 ± 0.036 (279)	0.37 ± 0.092 (150)
Peroxidase activity (µMole/g LFW)	4.24 ± 0.017 (100)	9.15 ± 0.020 (216)		8.54 ± 0.244 (201)	6.47 ± 0.387 (153)	4.61 ± 0.191 (109)

Values in parenthesis indicate percent activity; value represents mean of 5 samples with their standard error (±)

Abbreviation (g – Gram, LFW – Leaf Fresh weight, L – litre, mg – Milligram, SWP – Seaweed powder).

Conventional methods of removal of toxins present in the Plate making industrial effluent are expensive and hence the use of low cost environment friendly bioadsorbents has been tested. The dried algal biomass used in the present study is available in large quantities in the East Coast area of Indian sub continent. This can be utilized for removal of heavy metal and also found to be a potential, economic and effective safe alternative (Jeyakumar and Ramasubramanian, 2009).

Result of the present study clearly shows that the green algae used *i.e.* *Ulva lactuca* can efficiently remove the toxicity from effluent. Hence we strongly suggest that *Ulva lactuca* can be used as a bioadsorbent to remove the toxicity of effluent polluted environment for sustainable agriculture.

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