



Bioremediation of Industrial Effluents Using Fresh Water Cyanobacterial Species

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Abstract

Today cyanobacteria are widely used in bioremediation of industrial effluents. Hence the present study was attempted to analyse the effect of individual and combination effect of Cyanobacterial species isolated from Mukkumbu, the River Cauvery on bioremediation of pollutants. Results indicate that all the species showed the ability to degrade pollutants. However, the growth and degradable abilities were found to differ in different species, pollutant concentration and exposure; while the Cyanobacterial combination performed better at low concentrations, individual organisms recorded higher degradability at higher concentration. Thus, the results clearly demonstrate the ability of the organisms to degrade the pollutants. However, the growth and degradable abilities were found to differ among the species, pollutant concentration as well as the exposure time.

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Introduction

Cyanobacteria are known to occupy a vast variety of habitats even though they are susceptible to sudden physical and chemical changes in light, salinity, temperature and nutrient availability (Boominathan, 2005; Semyalo, 2009).

Today, they are widely used in waste water and industrial effluent treatment, bioremediation of aquatic and terrestrial habitats, chemical industries, biofertilizers, food, feed, *etc.* (Cairns and Dickson, 1971) besides being a source of fine chemicals and as a source of renewable fuel (Lem and Glck, 1985).

With increasing pollution, there has been a series of studies to combat it and one of the thrust areas has been the use of bioremediation measures using cyanobacteria. However, the beneficial application of cyanobacteria in

remediation of contaminated waters either in natural aquatic environments or industrial effluents is still not optimally manipulated (Dubey *et al.*, 2011). Hence the present study was done to assess the biodegradable abilities of some naturally occurring cyanobacteria.

Materials and Methods

Microorganisms

Cyanobacterial species were isolated from the Cauvery River, Mukkumbu of Tiruchirappalli District, Tamil Nadu, India. Micro flora dominates the Cauvery River and small ponds. All species involved in the study were isolated and identified in the Laboratory of Zoology, Arignar Anna Government Arts College, Musiri-621211, Tamil Nadu, India. These species included *Anabaena circinalis*, *Oscillatoria salina*, *Synechococcus elongatus*, *Spirulina major* and *Lyngbya trunicicola*.

Sampling of industrial effluent

Samples were collected from the paper industry and textile industry effluents, Karur District, Tamil Nadu, India. Grab samples representing all wastes entering the plant during 24 hr were collected from both industries to avoid the fluctuation in the flow and the strength of the influent. Physico-chemical analysis were done according to APHA (1998).

Media and culture conditions

BG-11 modified medium (SP) was used. This consisted of solutions A and B, containing (in grams): A; NaHCO₃ 11.61 g, Na₂CO₃ 3.53 g, K₂HPO₄ 0.5 g dissolved in 500 ml distilled water and B; NaNO₃ 5 g, K₂SO₄ 1 g, NaCl 1 g, MgSO₄.7H₂O 0.2 g, CaCl₂ 0.04 g, and 1 ml EDTA (0.5 M). Micronutrient solution (CHU No.10) consisted of the following trace metals (in milligrams) dissolved in one litre distilled water: Na₂-EDTA 50 g, H₃BO₃ 618 g, CuSO₄.5H₂O 19.6 g, ZnSO₄.7H₂O 44 g, CaCl₂.6H₂O 20 g, MnCl₂ 12 g and Na₂MoO₄.2H₂O 12.6 g. Solutions-A and B were sterilized by autoclaving separately at 121°C for 20 min. Micronutrient solution was sterilized by filtration through 0.22mm polycarbonate membrane to avoid interaction and precipitation of heavy metals. After sterilization, solutions-A and B were combined and 1 ml of the micronutrient solution and 1 ml of vitamin B₁₂ (stock solution 15.0 × 10⁻⁶ g) were added. Two modifications of BG₁₁ were developed in order to define the optimal conditions required for the enrichment and propagation of cyanobacterial isolates and to enhance the natural biodegradation activity of the purified isolates. The first modification was doubling vitamin B₁₂ concentration, which enhanced growth. The second was increasing nitrogen content, based on the fact that the enzyme responsible for dechlorination activities (nitrate

reductase) is induced at high nitrogen levels (Kuritzet *al.*, 1997). Optimization of cyanobacterial growth included adjusting the light/dark cycle, with 16/8 hr of white light 8 ft 40 W with light intensity (3200 lux), temperature at 25 to 30°C, and shaking (120 rpm) of the cultures, all of which led to enhancement of mass production. Prior to biodegradation bioassays, all cultures were tested for the presence of heterotrophic bacteria microscopically and by plating on bacterial nutrient medium (nutrient agar, Difco, UK) and incubating at 30°C for 1 week. Only axenic cultures, either uni- or multi-algal species, were used in the assays (Dubey *et al.*, 2011).

Results and Discussion

The various physico-chemical variables that were analysed during the period of study are presented in Table-1. The degradation efficiency of the individual organisms to various concentrations for a period of nine days are presented in Table-2. At 5 ppm concentration and an exposure period of three days, among the various species analysed, *Spirulina major* recorded maximum degradation (98.2%), while after six days, *Lyngbya trunicide* (98.6%) and *Anabaena circinalis* (98.4%) recorded maximum degradation and after 9 days, *A. circinalis* recorded maximum degradation (96.4%); with regard to a residual concentration of 10 ppm, results clearly suggest that *L. trunicide* recorded maximum degrading ability at all the days examined when compared to others. Nevertheless, a closer perusal reveals that the degrading ability of all the organisms showed an increasing trend when compared to a residual concentration of 5 ppm. Thus, the results clearly demonstrate the ability of the organisms to degrade the pollutants. However, the growth and degradable abilities were found to differ among the species, pollutant concentration as well as the exposure time.

Table.1 Biodegradation of industrial effluents by the selected cyanobacterial species isolated from Mukkomby, the River Cauvery

S. No.	Cyanobacterial species	Exposure time (days)	5 ppm		10 ppm	
			Residual concentration*	Removal efficiency (%)	Residual concentration*	Removal efficiency (%)
1.	<i>Anabaena circinalis</i> + <i>Spirulina major</i>	3	0.03	99.5	0.40	96.0
		6	0.00	100.0	0.20	99.8
		9	0.00	100.0	0.28	97.1
2.	<i>Oscillatoria salina</i> + <i>Spirulina major</i>	3	0.16	97.2	0.39	97.0
		6	0.03	99.4	0.43	95.6
		9	0.31	92.0	0.86	90.5
3.	<i>Synechococcus elongatus</i> + <i>Spirulina major</i>	3	0.08	99.7	0.67	99.4
		6	0.03	99.8	0.00	100.0
		9	0.05	99.0	0.84	92.0

* Recovery 90%, Mean standard error

Table.2 Selected species of Fresh water Cyanobacteria and its removal efficiency

S. No.	Cyanobacterial species	Exposure time (days)	5 ppm		10 ppm	
			Residual concentration*	Removal efficiency (%)	Residual concentration*	Removal efficiency (%)
1.	<i>Anabaena circinalis</i>	3	0.28	94.2	0.12	99.0
		6	0.81	98.4	0.18	98.2
		9	0.12	96.4	1.20	90.2
2.	<i>Oscillatoria salina</i>	3	0.12	96.0	0.50	95.4
		6	0.18	97.0	0.19	98.4
		9	0.13	80.1	0.18	98.2
3.	<i>Synechococcus elongatus</i>	3	0.72	88.2	0.42	96.7
		6	0.08	97.6	0.12	99.0
		9	0.26	94.5	0.29	98.0
4.	<i>Spirulina major</i>	3	0.28	98.2	0.73	93.6
		6	0.09	97.4	0.46	96.0
		9	2.90	78.6	1.67	84.0
5.	<i>Lyngbya trunicicola</i>	3	0.46	92.0	0.04	99.6
		6	0.02	99.6	0.07	99.3
		9	1.56	70.2	0.18	98.8

* Recovery 90%, Mean standard error

Table.3 Physico-chemical analysis of industrial effluents collected from Karur

Industrial effluent	Unit	Textile Dye Industry	Paper Industry
Colour		Pale Blue	Colourless
Temperature	°C	32	33
pH		6.8	7.1
BOD	mg/l	260	289
COD	mg/l	640	686
DO	mg/l	2.4	2.6
Ammonia	mg/l	186	67
Nitrite	mg/l	76	75
Nitrate	mg/l	180	154
Organic Phosphate	mg/l	22	20
Inorganic Phosphate	mg/l	21	22
Calcium	mg/l	57	81
Chloride	mg/l	1268	1440
Magnesium	mg/l	46	70

A perusal of literature reveals that Dubey *et al.*, (2011), while assessing the use of cyanobacteria in bioremediation of industrial effluents also recorded a similar finding. In fact, they also noticed an enhanced degrading ability with an increase in residual concentration.

However, they recorded highest removal efficiency percentages during the fourth day. In the present study, it was uniformly noticed on the sixth day after which it decreased. Dubey *et al.*, (2011) attributed this to accumulation of the species as well as appearance of mutants.

Results of the cyanobacterial combination studies on the effect of degradation at different concentrations are presented in Table-3. As evident from the table, at 5 ppm residual concentration set-III containing a combination of *Synecoccus* and *Spirulina* recorded maximum degradation (99.7%) on the third day, while Set-I having a combination of *Anabaena* and *Spirulina* recorded 100% degradation on the sixth and ninth day. At 10 ppm residual concentration, Set-III recorded highest degradation on third and sixth days, while Set-I recorded maximum degradation on the ninth day. A comparison between the individual and combination effect reveals that at 5 ppm concentration, the cyanobacterial combination appeared to work more effectively than the individual combination.

However, a closer perusal of 10 ppm concentration reveals that the individual species recorded higher degradation than the cyanobacterial combination. Hence, further studies need to be done to understand this phenomenon.

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