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Treatment of Sugar Mill Effluents with Cyanobacteria

Jayanthi Abraham[1] and Sonil Nanda[2]

[1]Division of Environmental Biotechnology,
School of Bio Sciences and Technology, VIT University, Vellore - 632 014, India.
[2]Orissa University of Agriculture and Technology, Bhubaneswar - 751 003, India.

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The disposal of sugar mill effluents into the environment creates serious adverse effects by altering the normal physiochemical properties of soil and water. The wastewater from sugar factories readily provides a source of soluble food which is an ideal substrate for the bacterial growth. Being rich in carbohydrates, they disturb the normal microbial growth thereby causing oxygen depletion. The rich organic matter favours the growth of various microorganisms and depletes oxygen by rapid respiration and oxidation which is recognized to degrade the pollutants. In this study, the effectiveness of cyanobacterial treatment system for bioremediation of sugar mill effluents was investigated. In the treatment system, *Oscillatoria* was employed for the bioremediation of sugar mill effluents. The effluents were analysed for their physiochemical and elemental parameters. The major interests were evaluating the percent removal of colour, biological oxygen demand (BOD), chemical oxygen demand (COD) and total dissolved solids (TDS) of the effluents. The results revealed a considerable decrease of 39.2% in colouration, 25.69% in BOD, 37.91% in COD and 48.51% in TDS of sugar mill effluents after 4 weeks of treatment with *Oscillatoria*.

Key words: Sugar mill effluents, *Oscillatoria*, bioremediation

Industrialization counts one of the basic reasons for a nation's development. During the production process, industries generate a bulk amount of waste products that are generally released into the natural environment causing water, soil and air pollution.

The effluents from various industries like chemical, fertilizer, food, sugar mill, textile, paper, dairy, tannery etc. are the major sources of these pollutions.[1] All types of effluents and their byproducts possess a serious threat to aquatic and terrestrial ecosystems.

Sugar mill are one of such sources of effluents whose disposal into the environment creates serious adverse effects by altering the normal physiochemical properties of soil and water. In fact, sugar and organic matter present in the effluents favour the growth of various microorganisms as ideal substrates and deplete oxygen by rapid respiration and oxidation. As a result, a major oxygen deficiency is encountered by the aquatic ecosystem which is deleterious for the native flora and fauna.

Sugar mill industries are found to be economically important in trading and fetching foreign exchange. Producing over 18.2 million tons of sugar annually, India bags its position as

* To whom all correspondence should be addressed.
Mob.: + 91-9843580709;
E-mail: jayanthi.abraham@gmail.com,
isonil@yahoo.co.in

one of the largest producers of sugar in the world.[2] At all stages of sugar production, starting from mill house, process house, boiler house to cooling pond and distillery are water-intensive, discharging waste water containing significant proportion of oil, suspended solids, organic matter and chemicals.[3] The amount of wastewater released from a sugar factory is usually greater than the volume of raw water input because of the high moisture content of sugarcane i.e. about 70 to 75%. Despite soil and water pollution, sugar mills also contribute to noise pollution. The polluted wastewater from sugar mills poses a substantial danger to health and environmental quality. Pollution control measures for such a large-scale industrial operation must be undertaken and perfectly monitored to minimize risks to health and environment.

The major intension of the study was to analyze the biodegradability of sugar mill effluents by cyanaobacteria, particularly *Oscillatoria* sp. The motive behind such type of research is to promote the biotreatment as the same as in natural conditions and in a laboratory design. However, this rationale is to attract attention on natural resources that would aid in cleaning up the environment. Since, cyanobacteria generally do not require any specific nutrients, our interest was to grow the species in aqueous solution of sugar mill effluents and study its degradability by checking the degree of treatment required to bring the effluent quality to permissible levels, thus, ensuring that the final product is either safe for disposal or acceptable for specific re-use or recycling.

MATERIALS AND METHODS

Sample collection

The wastewater samples were collected from Arignar Anna Sugar Mill, Kurungulam, Thanjavur, India, in sterilized bottles and transported to the laboratory for further study. The samples were collected once in two weeks.

Physiochemical analysis

Physiochemical analyses of the sugar mill effluents were performed following the standard methods by APHA.[4] The parameters analysed were colour, conductivity, pH, total dissolved solids (TDS),[5] biological oxygen demand (BOD), chemical oxygen demand (COD) and total nitrogen. Methods described by APHA[6] were followed for determination of bicarbonate, carbonate, sulphite and sulphate. Gravimetric estimation for chloride was performed[7] and phosphate was estimated by procedures described by Murphy and Riley.[8] The pH and electrical conductivity of the samples were measured using a glass electrode pH meter and conductivity meter, respectively. Total nitrogen was estimated using Kjeldhal N-analyzer.

Elemental analysis

Toxic metals in the effluents were determined by atomic absorption spectrophotometer following wet oxidation of the effluent sample by di-acid digestion method with a mixture of concentrated HNO_3:$HClO_4$ (3:1 v/v).[9]

Source of organism and culture

The cyanobacteria *Oscillatoria* was isolated from the effluent samples and cultured in BG11 medium in Erlenmeyer flasks at 30°C and 190 rpm for about 20 days.[10] The culture environment was illuminated properly to facilitate the cyanobacterial growth. The organism was obtained in pure culture as mats and maintained for further analysis on the effluent samples.

Determination of biodegradability

Effluent samples were inoculated with *Oscillatoria* in Erlenmeyer flasks and kept under illumination at 30°C for 28 days. For first 48 hours of incubation, the flasks were kept in an incubator shaker at 100 rpm for the purpose of uniform mixing of cyanobacteria and effluents. Periodic weekly monitoring was done for investigating the physiochemical characteristics and biodegradability of the effluents. Potential decolourization with BOD, COD and TDS removal of the effluent by *Oscillatoria* was investigated on weekly basis for 4 weeks. Decolorization of the effluents was determined by measuring the absorbance of the simulated effluents at 485 nm wavelength.

RESULTS AND DISCUSSION

Sugar mill effluent samples were analyzed for their physiochemical and elemental characteristics before and after treatment with *Oscillatoria*. Table 1 makes a comparison between

the effluent parameters before and after treatment with *Oscillatoria*. A change in colour of the effluents was an initial indication of biodegradation. The initial effluent colour at the time of collection was dark olive and after about a month of treatment with cyanobacteria it turned lighter. The colour is a contribution of dissolved solids and minerals of vegetable origin, tannins, synthetic dyes etc. The dyes in the effluent colour the water bodies and hampers light penetration which is a very critical factor for aquatic life forms.[11] However, after a due course of discharge of the effluents into the water bodies, there is a marked loss in colouration between 10-15%.[12]

The acceptable limits for discharge of wastewaters for both surface waters and sewers vary, ranging between from pH 5.5 to 10.[13] The pH of the effluent was 8.3 ± 0.3. This alkalinity may be due to the rate of aerobic decomposition. Formation of NH_3 from NH_4^+ is favoured by an alkaline pH which might result in NH_3 volatilization.[14] This interaction can be related to the total N content of the effluents. The total N content of the effluents after 28 days of incubation with *Oscillatoria* was 926.4 ± 12.5 mg/l which is a considerable decrease from 1445.0 ± 25.5 mg/l as before treatment. The effluents were initially characterized by a high electrical conductivity of 21.2 ± 0.5 dSm/l and after treatment a satisfactory decrease to 10.5 ± 0.5 dSm/l was found. The high conductivity however appeared not to have affected cyanobacterial activity during

Table 1. Physiochemical and elemental parameters of sugar mill effluent before and after treatment with *Oscillatoria*

Parameter	Concentration		
	Before treatment	After treatment	
		Day 14	Day 28
Physiochemical analysis			
Colour	Dark olive	Light olive	Light olive
Electrical conductivity (dSm/l)	21.2 ± 0.5	16.9 ± 0.5	10.5 ± 0.5
pH	8.3 ± 0.3	8.1 ± 0.2	7.6 ± 0.2
Total dissolved solids (mg/l)	3048.0 ± 36.3	2714.0 ± 30.0	1569.5 ± 28.0
Biological oxygen demand (mg/l)	2419.5 ± 23.5	1972.0 ± 20.2	1798.0 ± 16.5
Chemical oxygen demand (mg/l)	1820.0 ± 26.2	1571.0 ± 16.5	1130.1 ± 10.0
Total nitrogen (mg/l)	1445.0 ± 25.5	1131.2 ± 15.5	926.4 ± 12.5
Bicarbonate (%)	210.27 ± 10.3	116.0 ± 4.1	92.3 ± 3.0
Carbonate (%)	107.17 ± 4.5	95.08 ± 2.2	63.92 ± 2.2
Chloride (mg/l)	85 ± 5.0	68 ± 1.0	49 ± 1.0
Phosphate (mg/l)	28.81 ± 6.1	16.27 ± 2.1	14.70 ± 1.4
Sulphite (mg/l)	97.0 ± 0.5	74.9 ± 0.6	58.10 ± 0.5
Sulphate (mg/l)	105.1 ± 4.5	97.0 ± 2.0	66.1 ± 1.0
Elemental analysis			
Arsenic (mg/l)	2.8 ± 0.1	1.5 ± 0.02	0.7 ± 0.02
Calcium (mg/l)	98.0 ± 2.5	76.2 ± 1.5	43.1 ± 0.5
Cadmium (mg/l)	5.1 ± 0.01	1.7 ± 0.01	0.4 ± 0.02
Chromium (mg/l)	0.5 ± 0.02	BDL	BDL
Cobalt(mg/l)	0.4 ± 0.1	0.2 ± 0.01	0.1 ± 0.01
Copper (mg/l)	4.1 ± 0.1	1.0 ± 0.01	0.3 ± 0.01
Iron (mg/l)	6 ± 0.5	3 ± 1.0	1.7 ± 0.03
Lead (mg/l)	2.1 ± 0.2	1.0 ± 0.5	0.8 ± 0.02
Magnesium (mg/l)	9.5 ± 1.5	8.1 ± 1.0	2.2 ± 0.01
Sodium (mg/l)	93.3 ± 2.5	71 ± 1.5	54 ± 0.5
Zinc (mg/l)	15.0 ± 0.1	9.3 ± 0.2	4.7 ± 0.2

Note: Mean ± standard deviation (n = 5); BDL: Below detectable levels

bioremediation. Santamaria-Romero and Ferrera-Cerrato[15] reported that salt concentration above 8.0 dSm/1 negatively affected the microbial populations as well as biotransformation of organic matter. Levels of TDS were almost halved after the cyanobacterial treatment i.e. from 3048.0 ± 36.3 mg/l to 1569.5 ± 28.0 mg/l as on the final day of treatment.

Fig. 1 gives a percent removal of BOD, COD, TDS with decolouration of the effluent after *Oscillatoria* treatment for 28 days. There was a considerable decrease of 39.2% in colouration, 25.69% in BOD, 37.91% in COD and 48.51% in TDS of sugar mill effluents after 4 weeks of treatment with *Oscillatoria*. The high BOD often creates septic conditions, generating foul-smelling hydrogen sulfide, which in turn precipitates iron and any dissolved salts, turning the water black and highly toxic for aquatic life.[3] Sugar processing wastewaters are characterized by a high BOD because of the presence of sugars and other organic material from sugarcanes. Presence of pesticides, agro-residues and pathogens in the effluents results from the improper washing and processing of crude raw materials. In addition, discharge of effluents with a high TDS level has adverse impact on aquatic life, renders the receiving water unfit for drinking, reduces crop yields if used for irrigation and exacerbates corrosion in water systems and pipes.[16]

The elemental analysis implies that the effluents were rich in calcium and sodium

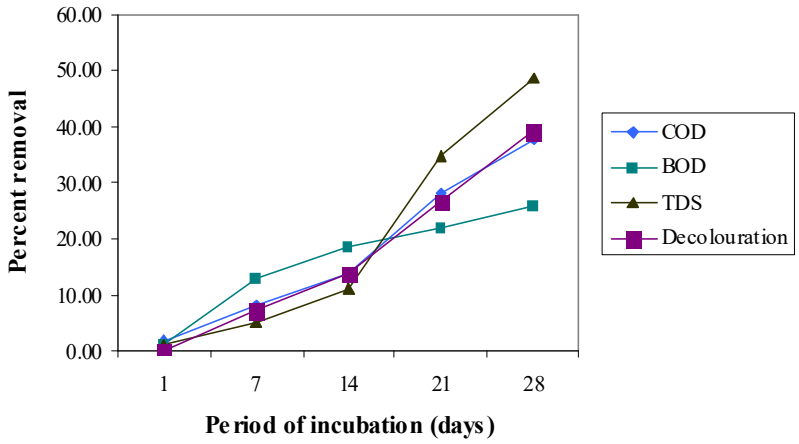

Fig. 1. Percent removal of BOD, COD, TDS and colouration of sugar mill effluents under aerobic condition

followed by zinc. With increasing heavy metal pollution, cyanobacteria are found indispensable tools for their bioremediation.[17-19] The pH enhanced the heavy metal bioremediation by *Oscillatoria*. The pH of the effluents varied between 7.5 and 8.5 during cell growth, which is similar to the natural variations in seawater, thus indicating no significant precipitation of heavy metals by alkalinization.[17] The concentration of chromium was 0.5 ± 0.02 initially and on day 14 of treatment it was below detectable levels. An acceptable decrease in the concentrations of heavy metals was evident on day 28 of cyanobacterial treatment.

CONCLUSIONS

This study indicates that cyanobacterial treatment method is a feasible technique for bioremediation of sugar mill effluents. There was a considerable decrease of 39.2% in colouration, 25.69% in BOD, 37.91% in COD and 48.51% in TDS of sugar mill effluents after 4 weeks of treatment with *Oscillatoria*. The mass culture of the cyanobacteria in form of mats enhanced the bioremediation process in laboratory set-up as in natural conditions.

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