Cyanobacterial remediation of industrial effluents I. Tannery effluents

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Abstract: Tannery effluents are of large-scale environmental concern because they colour and diminish the quality of water bodies into which they are released. Their disposal into the environment creates adverse effects by altering the normal physiochemical properties of soil and water. In this study, cyanobacteria, particularly *Nostoc* was employed for bioremediation of tannery effluents. The percent removal of biological oxygen demand (BOD), chemical oxygen demand (COD), total dissolved solids (TDS) and colour of the effluents were studied. Other analyses involved the physiochemical and elemental properties of the effluents. The results revealed a 57.5%, 37.8%, 48.6% and 66.1% decrease in BOD, COD, TDS and colour of the tannery effluents after 4 weeks of treatment with *Nostoc*. [New York Science Journal 2010;3(12):32-36]. (ISSN: 1554-0200).

Keywords: Tannery effluents, *Nostoc*, bioremediation

1. Introduction

Tanning is a process of making leather from skin and hides. The process involves a complex combination of mechanical and chemical practices as a result of which organic and inorganic materials become chemically bound to the protein component of the hides and preserve it from deterioration. A significant number of operations within a tannery industry are wet operations consuming large amounts of water, chemicals and energy leading to large amounts of polluted water.

The industrial effluents contain several types of chemicals such as dispersants, leveling agents, acids, alkalis, carriers and various dyes, phenols, carbonates, alcohols, cyanide, heavy metals etc (Cooper, 1995). Leather industries and tanneries generate massive by-products, solid wastes, high amounts of wastewater rich in organic wastes with different loads of pollutants and emissions into the air. Majority of tanneries worldwide use chromium salts (Cr III and Cr IV) which are highly toxic and possess a serious threat to the environment upon improper disposal of their wastewaters. Even in low concentrations, these salts have a toxic effect upon daphnia, thus disrupting the food chain for fish life and possibly inhibiting photosynthesis of aquatic plants (Bosnic *et al*., 2000). Effluents from raw hide processing tanneries contain compounds of Cr III and sulphides in most cases with a major proportion of dyes. Release of these effluents into aquatic ecosystems alters the pH, increases the BOD and COD and gives the water intense colourations (Ajayi and Osibanjo, 1980). However, the toxicity of effluents depends on operations employed in each tannery.

In general, treatment of effluents includes physiochemical methods such as filtration, specific coagulation, use of activated carbon and chemical flocculation (Olukanni *et al*., 2006). Conventional treatment of tannery effluents for the purpose of detoxification requires application of physical and chemical methods which involves the chrome precipitation and sulphide treatment. Due to associated problems in these treatments such as cost and intense experimental set-up (Do *et al*., 2002; Maier *et al*., 2004), biological treatment methods using various bacteria and fungi have been widely studies. However, it is now becoming apparent that cyanobacteria also play a major role in degrading organic materials from the ecosystem. In this current research, *Nostoc* was employed in bioremediation of tannery effluents.

2. Material and Methods

2.1. Sample collection

Tannery effluents were collected from a few tannery industries situated in eastern India. As per the request, the details of the industries and proprietors have been kept confidential and are not discussed anywhere in this article. The effluents were collected in sterile glass bottles, transported in cold condition to the laboratory for physiochemical and microbiological analysis. For microbiological analysis, the effluent samples were sterilized by autoclaving at 121^{o}C for 15 mins.

2.2. Physiochemical analysis

Physiochemical analyses of the tannery effluents were performed following the standard methods by APHA (1992). The parameters analysed were colour,

conductivity, pH, TDS (Valentine, 1996), BOD, COD and total nitrogen. Methods described by APHA (1980) were followed for determination of bicarbonate, carbonate, sulphite and sulphate. Gravimetric estimation for chloride was performed (Strickland and Parson, 1972) and phosphate was estimated by procedures described by Murphy and Riley (1962). Total nitrogen was estimated using Kjeldhal N-analyzer.

2.3. Elemental analysis

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) (Hossner, 1996).

2.4. Source of organism and culture

Nostoc was cultured in BG11 medium in Erlenmeyer flasks at 30^{o}C and 190 rpm (Yoon *et al*., 2002) for about 21 d. The culture environment was illuminated properly to facilitate the cyanobacterial growth. The organism was obtained in mats and maintained for further analysis on the effluent samples.

2.5. Determination of biodegradability

50 ml of sterile tannery effluent samples were supplemented to 250 ml of BG11 media inoculated with *Nostoc* in Erlenmeyer flasks and kept under illumination at 30^{o}C for 28 d under aerobic condition. For control, 50 ml of sterile effluents were added to 250 ml BG11 media without inoculation of *Nostoc*. For first 48 h of incubation, the flasks were kept in an incubator shaker at 100 rpm for the purpose of uniform mixing of the media and effluents. Periodic weekly monitoring of the samples was done for investigating the physiochemical characteristics and biodegradability of the effluents. For determining decolouration of the effluents, the media was centrifuged at 5000 rpm for 15 mins to get cell free filtrate. The clear filtrate was analysed in a spectrophotometer for measuring its absorbance at 485 nm wavelength. Percent removal of colour, BOD, COD and TDS of the effluents by *Nostoc* was evaluated.

3. Results and Discussion

Effluent samples were analyzed for their physiochemical and elemental characteristics before and after treatment with *Nostoc*. Table 1 makes a comparison between the control and treated effluents on a timescale basis. A change in colour of the effluent was an initial indication of biodegradation. The initial effluent colour at the time of collection was dark brown and finally after cyanobacterial treatment for 4 wks it turned tan. The colour is a contribution of dissolved solids and minerals of vegetable origin, tannins, synthetic dyes etc. The dyes colour the water bodies and hampers light penetration which is a very critical factor for aquatic life forms (Goncalves *et al*., 2000). However, after a due course of discharge of the effluents in the water bodies there is a marked loss in colouration between 10 to 15% (Vaidya and Datye, 1982). As the chief ingredients of BG11 medium are salts, hence the supplementation of tannery effluents into the minimal medium acted as the carbon source for the cyanobacteria to metabolize it and reduce its concentration from the medium.

The acceptable limits for discharge of wastewaters to both surface waters and sewers vary, ranging between from pH 5.5 to 10 (Bosnic *et al*., 2000). The initial pH of the effluent was 9.2±0.2. This alkalinity is 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 (Contreras-Ramos *et al*., 2004). This interaction can be related to the total N of the effluents. The total N of the effluents on day 28 was 171.4±5.5mg/l which is a considerable decrease from 552.0±10.5mg/l as of before treatment. Several components in tannery effluent contain nitrogen. The most common sources are the deliming materials that are rich in ammonia and the nitrogen contained in proteinaceous materials contributed through liming and unhairing operations (Bosnic *et al*., 2000).

The tannery effluents were initially characterized with a high electrical conductivity of 31.2±0.5dSm/l and after treatment a satisfactory decrease to 16.1±0.5dSm/l was found. The high conductivity however appeared not to have affected cyanobacterial activity during bioremediation. Santamaria-Romero and Ferrera-Cerrato (2001) reported that salt concentration above 8.0dSm/1 negatively affected the microbial populations as well as biotransformation of organic matter. TDS were almost reduced to half its original concentration after the cyanobacterial treatment i.e. from 2200±20.0mg/l to 1130.5±13.0mg/l as on the final day of treatment.

Figure 1, 2, 3 and 4 illustrate the percent removal of BOD, COD, TDS and colouration of the effluent after cyanobacterial treatment for 28 d. On day 28 of treatment, the percent degradation for BOD, COD, TDS and colour were 57.5%, 37.8%, 48.6% and 66.1%, respectively. The high BOD often creates septic conditions, generating foul-smelling hydrogen sulphide, which in turn precipitates iron and any dissolved salts, turning the water black and highly toxic for aquatic life (Akbar and Khwaja, 2006). Moreover, wastewaters from leather industries are characterized with a high BOD because of the

presence of organic matter contributed from the skins and hides of animals.

The elemental analysis implies that the effluent is rich in calcium, sodium and chromium. Chromium was initiated by the tanning and retanning processes and is displaced from leathers during dyeing process. The high sulphide and sodium content in tannery effluent results from the use of sodium sulphide and sodium hydrosulphide, and the breakdown of hair in the unhairing process (Bosnic *et al*., 2000). Sulphates emanated from the use of sulphuric acid and products with high sodium sulphate content.

With increasing heavy metal pollution, cyanobacteria are found indispensable tools for their bioremediation (Nriagu and Pacyna, 1988). Chloride was introduced into tannery effluents from large quantities of sodium chloride used in hide and skin preservation or the pickling process. Certain bacteria and fungi are often sensitive to higher levels of these salts which cause their cellular breakdown. On the other hand, negligible effect of salts is found over cyanobacteria as it prefers to grow in salt mediums.

Table 1. Physiochemical and elemental characteristics of tannery effluents before and after treatment with *Nostoc*

Parameter	**Concentration**				
	Day 1	**Day 14**		**Day 28**	
		Control	**Treatment**	**Control**	**Treatment**
Physiochemical analysis					
Colour	Dark brown	Dark brown	Light brown	Light brown	Tan
Odour	Rotten egg smell	Rotten egg smell	Rotten egg smell	Rotten egg smell	Foul smell
pH	9.2 ± 0.2	9.0 ± 0.2	8.3 ± 0.2	8.8 ± 0.2	7.1 ± 0.2
Electrical conductivity (dSm/l)	31.2 ± 0.5	30.7 ± 0.5	22.9 ± 0.5	28.8 ± 0.5	16.1 ± 0.5
Total dissolved solids (mg/l)	2200.0 ± 20.0	2187.0 ± 18.2	1480.0 ± 15.0	2091.0 ± 17.0	1130.5 ± 13.0
Biological oxygen demand (mg/l)	740.1 ± 36.2	733.4 ± 25.5	471.9 ± 31.2	721.0 ± 21.2	314.1 ± 27.0
Chemical oxygen demand (mg/l)	892.0 ± 44.5	889.7 ± 42.0	710.4 ± 31.2	862.2 ± 35.0	554.7 ± 26.5
Total nitrogen (mg/l)	552.0 ± 10.5	511.6 ± 8.5	302.2 ± 5.5	472.1 ± 8.1	171.4 ± 5.5
Bicarbonate (%)	8.17 ± 0.3	8.0 ± 0.2	7.91 ± 0.1	7.79 ± 0.3	5.32 ± 0.1
Carbonate (%)	8.70 ± 0.5	8.22 ± 0.5	6.19 ± 0.2	7.81 ± 0.1	4.71 ± 0.2
Chloride (mg/l)	89.1 ± 5.0	83.9 ± 4.1	68.7 ± 4.0	81.3 ± 4.0	32.3 ± 0.5
Phosphate (mg/l)	33.8 ± 2.5	31.1 ± 2.0	21.2 ± 1.0	30.8 ± 2.1	12.4 ± 1.0
Sulphide (mg/l)	78.0 ± 4.0	72.9 ± 4.5	66.9 ± 3.5	72.0 ± 2.4	45.8 ± 1.0
Sulphate (mg/l)	65.1 ± 2.5	61.7 ± 1.7	47.0 ± 2.0	59.5 ± 1.2	25.1 ± 2.0
Elemental analysis					
Arsenic (mg/l)	2.8 ± 0.2	2.1 ± 0.2	1.5 ± 0.2	1.9 ± 0.1	1.2 ± 0.2
Calcium (mg/l)	318.0 ± 15.5	311.0 ± 15.2	227.2 ± 15.5	306.0 ± 14.9	185.6 ± 8.5
Cadmium (mg/l)	5.0 ± 1.0	4.4 ± 1.2	3.3 ± 0.5	4.2 ± 1.0	1.4 ± 0.1
Cobalt(mg/l)	1.4 ± 0.1	1.4 ± 0.07	0.3 ± 0.01	1.1 ± 0.03	0.2 ± 0.01
Chromium (mg/l)	269.1 ± 2.5	263.1 ± 2.1	217.0 ± 2.5	254.1 ± 1.7	128.1 ± 1.5
Copper (mg/l)	4.1 ± 0.1	4.0 ± 0.1	3.2 ± 0.01	3.7 ± 0.2	1.1 ± 0.01
Iron (mg/l)	18.0 ± 5.0	16.8 ± 5.0	13.0 ± 2.0	15.1 ± 3.2	6.0 ± 2.0
Lead (mg/l)	4.0 ± 1.2	3.8 ± 1.0	3.2 ± 1.0	3.1 ± 1.2	1.0 ± 0.05
Magnesium (mg/l)	9.0 ± 2.5	9.0 ± 1.5	6.1 ± 1.0	8.3 ± 0.4	5.2 ± 0.05
Sodium (mg/l)	292.3 ± 12.5	285.3 ± 11.5	245.0 ± 10.0	276.3 ± 10.1	161.0 ± 6.5
Zinc (mg/l)	18.6 ± 0.1	18.3 ± 0.2	8.8 ± 0.2	17.6 ± 0.06	5.7 ± 0.2

Note: Mean ± standard deviation (n = 5)

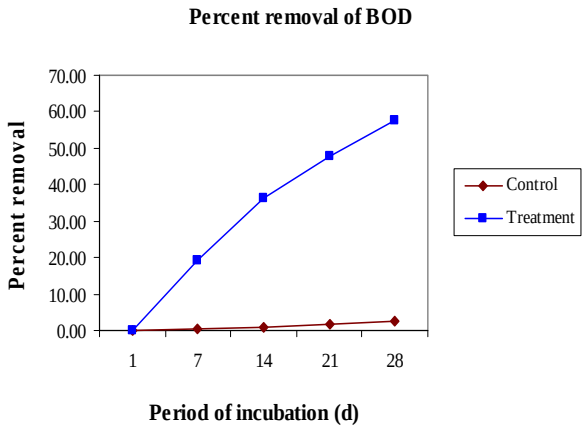

Figure 1. Percent removal of BOD of tannery effluents under aerobic condition

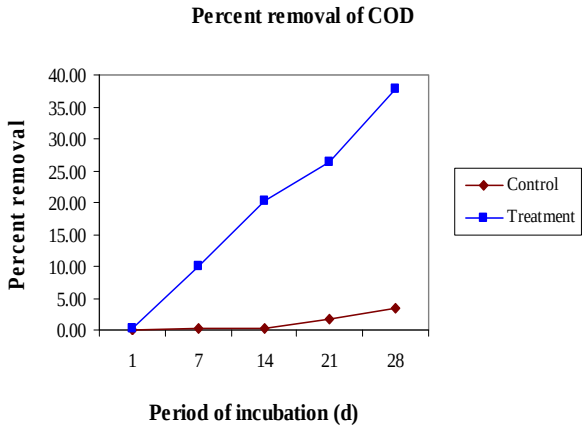

Figure 2. Percent removal of COD of tannery effluents under aerobic condition

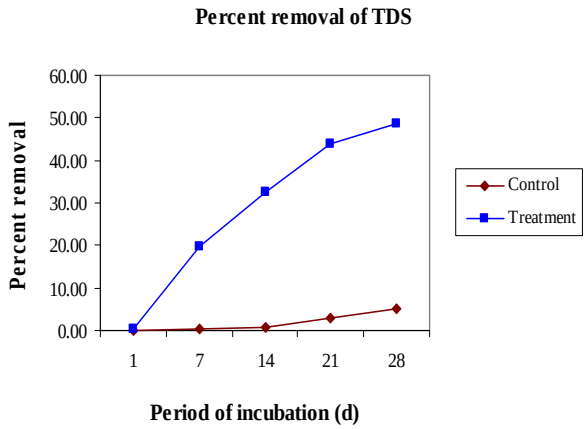

Figure 3. Percent removal of TDS of tannery effluents under aerobic condition

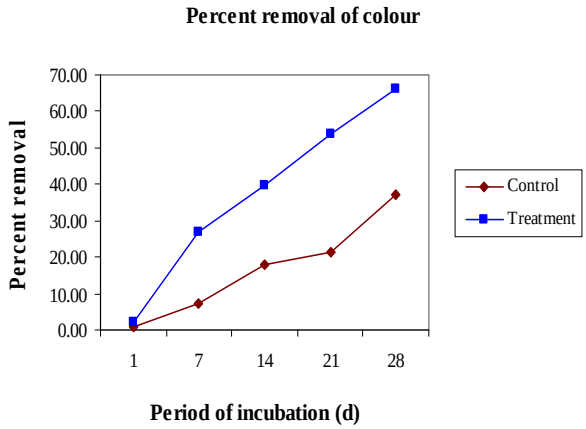

Figure 4. Percent removal of colour of tannery effluents under aerobic condition

Removal of heavy metals, especially cadmium by cyanobacteria has been extensively studied (Matsunaga *et al.*, 1999; Les and Walker, 1984). The pH enhanced the heavy metal bioremediation by *Nostoc*. The pH of the effluents varied between 7.1 and 9.2 during cell growth, which is similar to the natural variations in seawater, thus indicating no significant precipitation of heavy metals by alkalinization (Matsunaga *et al.*, 1999). During alkaline pH, the solution was rich in sulphides. Bosnic *et al.* (2000) found that with a fall in pH of the effluent below 9.5 there was liberation of hydrogen sulphide from the effluent which was characterised by a smell of rotten eggs. Another factor for odour may be the ammonia and urea released from the hides. The odour of the effluent did not vary much through out the treatment process. An acceptable decrease in the heavy metals concentration was evident on day 28 of cyanobacterial treatment.

However, slight reduction of the heavy metals in the control flasks was due to precipitation of their salts in aqueous solution. As an attempt for exploiting cyanobacteria in bioremediation, we discovered promising potency of *Nostoc* in treating tannery effluents.

4. Conclusions

The results indicated that cyanobacterial treatment method is a feasible technique for bioremediation of tannery effluents. The cyanobacterial interaction with tannery effluents effectively decreased their colour intensity, BOD, COD, TDS and metal concentrations. The results revealed a 57.5%, 37.8%, 48.6% and 66.1% decrease in BOD, COD, TDS and colouration of the tannery effluents after 4 weeks of treatment with *Nostoc*. The main economic advantage of this system is the lack of a serious sludge disposal problem and consequently much less operating cost.

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