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A STUDY ON MICROBIAL DIVERSITY AND PHYSICO-CHEMICAL CHARACTERISATION OF TEXTILE INDUSTRY EFFLUENT

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Abstract
Pollution has arisen as a serious environmental concern to the present world after industrialization of human societies. It has severely affected our air, soil and water sources. Looking to its global, national, regional and local dimensions, it is now imperative to check it at each and every level. In the present study, effluent on the microbial diversity viz. bacteria, fungi and cyanobacteria. Results of one year ecological study revealed that altogether 10 species of bacteria, 11 species of fungi and 20 species of cyanobacteria were isolated from the effluent stream. The species isolated, *Pseudomonas* with two species and others with single each were recorded. Among the fungi recorded the *Aspergillus* was found to be dominant with four species viz., *A. flavus, A. fumigatus, A. luchensis* and *A. niger*. Altogether 20 species of cyanobacteria belonging to ten genera were collected from the effluent stream. Among the genera, *Oscillatoria* dominated the effluent with seven species followed by *Phormidium, Lyngbya, Nostoc* and *Anabaena* with 2 each species and *Synechococcus elongates, Gleocapsa pleurocapsoides, Plectonema radiosum, Chroococcus minutes* and *Fischerella ambigua* with single species each. Higher amounts of phosphates and nitrates with sufficient amount of oxidizable organic matter, limited dissolved oxygen content and slightly alkaline pH were probably the factors favoring the growth of microbes especially cyanobacteria. The utilization of dominant species of cyanobacteria to monitor pollution in detergent effluent has been discussed.

KEYWORDS: Textile industry effluent, bacteria, fungi, cyanobacteria.

INTRODUCTION
Water is essential to all forms of life and makes up to 50-97% of the weight of all plants and animals and about 70% of human body. Water is also a vital resource for agriculture, manufacturing, transportation and many other human activities. Despite its importance, water is the most poorly managed resource in the world. The availability and quality of water always have played an important role in determining the quality of life. Water quality is closely linked to water use and to the state of economic development (Ramamurthy *et al*., 2014). Ground and surface waters can be contaminated by several sources. In urban areas, the careless disposal of industrial effluents and other wastes may contribute greatly to the poor quality of water (Ramamurthy *et al.*, 2015). Most of the water bodies in the areas of the developing world are the end points of effluents discharged from industries.

Non-hazardous industrial wastes are those that do not meet the EPA's definition of hazardous waste - and are not municipal waste. The EPA estimated in 1980 that more than 70,000 different chemicals were being manufactured in the U.S., with some 1,000 new chemicals being added each year. Industrial waste has been a problem since the industrial revolution. Industrial waste may be toxic, ignitable, corrosive or reactive. If improperly managed, this waste can pose dangerous health and environmental consequences. In United States, the amount of hazardous waste generated by manufacturing industries in the country has increased from an estimated 4.5 million tons annually after World War II to some 57 million tons by 1975. In 1990, this total had shot up to approximately 265 million tons. This waste is generated at every stage in the production process, use and disposal of manufactured products. Thus, the introduction of many new products for the home and office - computers, drugs, textiles, paints and dyes, plastics also introduced hazardous waste, including toxic chemicals, into the environment. These, too, must be managed with extreme care to avoid adverse environmental or human health impacts.

Textile industries represent a very diverse sector in terms of raw materials, processes, products and equipments and have very complicated industrial chain (Savin and Butnaru, 2008). A number of dyes, chemicals and other materials are used to impart desired grade and quality to fabrics. These industries generate substantial quantity of effluents, which contaminates the natural water bodies altering their physical, chemical and biological nature. Textile effluents can seep into aquifer, thus, polluting the underground water. The impact of textile industry on environment, both in terms of the discharge of pollutants and of the consumption of water and energy has long been recognized (Lacasse and Baumann, 2006).

Textile processing employs a variety of chemicals, depending on the nature of the raw material and product (Aslam et al., 2004). Major pollutants in textile wastewaters are high suspended solids, chemical oxygen demand, heat, colour, acidity, and other soluble substances. Colour is imparted to textile effluents because of various dyes and pigments used. In addition to dyes, various salts and chemicals are the major sources of heavy metals in wastewater.

Sediments, suspended and dissolved solids are important repositories for toxic heavy metals and dyes causing rapid depletion of dissolved oxygen leading to oxygen sag in the receiving water (Alihameed and Ahmed, 2008). The metals and contaminants like dyes tend to persist indefinitely, circulating and eventually accumulating throughout the food chain. The dyes and metals have direct and indirect toxic effects in the form of cancers and allergies besides, inhibiting growth at different trophic levels (Kant Rita, 2012). Taking the above facts into consideration, a survey was undertaken in detergent effluent to explore the nature of microbial flora such as bacteria, fungi and cyanobacteria are screen in order to biodiversity index.

MATERIALS AND METHODS

Effluent was collected from textile industry waste, Tirupur, Tamil Nadu, India. Samples were collected in large sterilized container and brought to the laboratory. Physico-chemical characteristics were done on the same day when the samples were brought to the laboratory. The effluent samples were filtered through cotton to remove suspended coarse particles before use. Population of microbes was isolated from the effluent samples by serial dilution technique. Bacteria were identified based on colony characteristics, Gram staining methods and by various biochemical studies as given by Bergey's (1984) Manual of Determinative Bacteriology. Fungi were identified by using standard manuals, such as Mannual of Soil Fungi (Gillman, 1957), Dematiaceous Hyphomycetes (Ellis, 1971). Effluent samples were collected in duplicate from station in pre-sterilized bottles. Samples were collected from the places along with effluents in polythene bags. Standard microbiological methods were followed for the isolation and identification of cyanobacteria (Desikachary, 1959). Physico-chemical characteristics of effluent were done according to the Standard Methods (APHA, 1995). Temperature and pH of the effluent were measured at the station itself.

RESULTS AND DISCUSSION

The results of physico-chemical characterization of textile effluent are analyzed. The effluent was slightly alkaline and contained high amounts of nitrate, nitrite and ammonia, total phosphate, inorganic and organic phosphate and calcium in all the seasons examined. Very low level of dissolved oxygen and high levels of BOD and COD were recorded during the study period. High amount of total suspended solids were recorded in summer and pre-monsoon seasons (Table 1). Total dissolved solids were high in summer followed by post monsoon and pre monsoon seasons. Bicarbonate was observed only during monsoon seasons. Nutrients such as nitrate, nitrite and inorganic phosphates were high in monsoon, whereas total and organic phosphates were maximum during monsoon season. BOD and COD were very high during summer on the other hand high level of DO was recorded during monsoon. Dissolved oxygen level was very low during summer and high in monsoon. Most of the parameters tested were slightly higher in summer than monsoon, post monsoon and pre monsoon seasons.

The physicochemical analysis of the effluent revealed that it is slightly alkaline in nature and also the presence of high quantity of both organic as well as inorganic nutrients in all the seasons examined (Table 1). Values of DO were very low indicating highly obnoxious conditions. Though BOD and COD levels in the present study were high as per WHO standards, their levels were not so much high as compared to other types of effluents such as paper (Somashekar and Ramasamy, 1983; Manoharan and Subramanian, 1992b), distillery (Jain *et al.*, 2001, Veerasamy *et al.*, 2011) and dye effluent (Sulaiman *et al.*, 2002). Most of the parameters tested were slightly higher in summer than in other seasons. Somashekar and Ramaswamy (1983) reported similar results in paper mill effluent in different seasons. They recorded objectionable amounts of BOD and COD, oil and grease, total dissolved solids, and algal nutrients such as ammonia nitrogen, nitrate nitrogen, silicates, phosphates and calcium. Such a trend was observed in the detergent effluent also. Sahai *et al.* (1985) analyzed pollution load of four different effluents such as fertilizer, sugar, distillery and domestic sewage. Among these, highly objectionable amounts of various pollutants including BOD and COD were recorded in distillery followed by sugar, fertilizer and domestic sewage.

Bacteria isolated from the textile effluent were identified based on colony morphology, Gram staining and various biochemical characteristics. The characteristics of isolated bacteria are given in the table 2. Totally ten different bacteria were isolated from the effluent sample. The species isolated were *Escherichia coli, Enterobacter aerogens, Klebsiella pneumonia, Proteus vulgaris, Pseudomonas* sp, *Pseudomonas aerogenosa*, *Shigella sonnei, Streptococcus faecalis, Bacillus subtilis* and *Staphylococcus faecium*. All the species were recorded in all the seasons. All the species were recorded in all the seasons. There was not been much work regarding the isolation and identification of bacteria from textile industry and other related effluent samples. Jain *et al.* (2001) isolated three different bacterial strains from the distillery sludge to treat predigested distillery wastewater. In the present study totally eleven different species of fungi belonging to eight genera were isolated from the effluent (Table 3). Among the fungi recorded the *Aspergillus* was found to be dominant with four species viz., *A. flavus, A. fumigatus, A. luchensis* and *A. niger*. The rest of the genera such as *Candida* sp, *Penicillium javanicum, Saprolespgia* sp, *Trichoderma viride, Alternaria* sp, *Fusarium oxysporum* and *Rhodosporium* were recorded with single species each. Similarly *Neurospora crussa* was not observed during rainy season. Kousar *et al.* (2000) isolated 23 species of fungi from dye effluent polluted habitat with *Aspergillus* as the dominant genus.

Altogether 20 species of cyanobacteria belonging to ten genera were collected from the effluent stream (Table 4). Among the genera, *Oscillatoria* dominated the effluent with seven species followed by *Phormidium, Lyngbya, Nostoc* and *Anabaena* with 2 each species, *Synechococcus elongates, Gleocapsa pleurocapsoides, Plectonema radiosum, Chroococcus minutes* and *Fischerella ambigua* with single species each. Many publications emphasize the importance of light, temperature, pH, carbon dioxide, organic matter, alkalinity, nitrates and phosphates as factors important in determining the distribution of cyanobacteria (Singh, 1960; Philipose, 1960; Venkateswaralu, 1969, Munawar, 1970). In the present study, as a whole, conditions in the effluent appeared to be favourable for the cyanophycean members. The effluent had high oxidizable organic matter, nutrients such as nitrates and phosphates with high calcium content. They noted that oxygen deficiency favoured cyanobacterial growth. Dominant and persistent occurrence of most of the species of *Oscillatoria* and *Phormidium* indicate their capacity to thrive in the type of man-made habitat. Moreover these findings show that there are certain species of cyanobacteria which are tolerant to organic pollution and resist environmental stress caused by the pollutant.

Rich blooms of some cyanobacteria such as *O. animalis, O. salina, O. brevis, O. subbrevis, P. tenue* and *Anabena variabilis* have been observed throughout the year. This abundance is attributed to favourable conditions of oxidizable organic matter, less dissolved oxygen and high calcium content an observation which supports Rao (1955) and Venkateswarlu (1969). Observations of Munawar (1970a, b) suggest that Cyanophyceae grow luxuriously with great variety and abundance in ponds rich in calcium. Venkateswarlu (1969) observed that high orthophosphate levels favoured the development of cyanobacterial bloom. Sarojini (1996) observed positive correlation between phosphate and cyanobacteria. The luxuriant growth of cyanobacteria at low concentration of oxygen and in the presence of high concentration of nitrogen and phosphate has also been reported by Rai and Kumar (1977). Similar observations were also made in the present study with reference to various nutrients. Rai and Kumar (1976a) observed non heterocystous forms in the polluted waters rich in nitrogen. Present investigation also showed dominance of non heterocystous forms and not single species of heterocystous cyanobacteria.

Table 1: Characteristics of effluent observed in four seasons.

S.No.	Parameters	Summer	Premonsoon	Monsoon	Post monsoon
1.	Temperature^{0}C	22.75 ± 1.05	20.17 ± 0.65	18.85 ± 0.17	20.45 ± 0.16
2.	pH	8.95 ± 0.26	8.76 ± 0.28	8.52 ± 0.37	8.80 ± 0.22
3.	Total suspended solids	1990 ± 15.5	1976 ± 28.7	1951 ± 58.2	1968 ± 22.5
4.	Total dissolved solids	1430 ± 15.2	1385 ± 14.5	1374 ± 18.3	1393 ± 11.7
5.	Free carbon-di-oxide	25.3 ± 1.06	24.9 ± 1.07	17.8 ± 1.12	21.7 ± 1.16
6.	Carbonate	2.38 ± 0.15	2.16 ± 0.18	3.68 ± 0.14	2.58 ± 0.16
7.	Bicarbonate	56.5 ± 1.81	57.3 ± 2.13	59.8 ± 1.19	58.9 ± 1.26
8.	BOD	291 ± 5.27	248 ± 6.51	236 ± 9.15	259 ± 7.26
9.	COD	512 ± 8.12	438 ± 4.84	385 ± 6.71	440 ± 5.18
10.	Dissolved oxygen	3.21 ± 0.12	4.18 ± 0.17	4.93 ± 0.24	4.09 ± 0.17
11.	Nitrate	92.7 ± 2.44	93.9 ± 2.81	96.9 ± 2.29	94.1 ± 2.52
12.	Nitrite	66.1 ± 2.27	69.9 ± 2.17	78.2 ± 2.12	71.5 ± 2.16
13.	Ammonia	41.3 ± 3.24	44.6 ± 3.08	46.2 ± 3.17	45.4 ± 3.42
14.	Total phosphate	70.02 ± 4.18	72.05 ± 4.13	80.25 ± 4.19	71.45 ± 4.71
15.	Inorganic phosphate	36.52 ± 2.19	38.93 ± 2.17	39.25 ± 2.18	38.25 ± 2.32
16.	Organic phosphate	35.50 ± 3.15	38.06 ± 3.18	40.50 ± 3.42	39.24 ± 3.72
17.	Calcium	191 ± 2.10	178 ± 2.42	170 ± 2.17	181 ± 2.18
18.	Magnesium	150 ± 2.17	143 ± 2.54	132 ± 2.52	141 ± 2.17
19.	Chloride	69.2 ± 4.62	63.1 ± 4.57	62.3 ± 4.43	65.4 ± 4.35

(Each value represents mean ± SD of three observation)

Monsoon (Oct-Dec); Postmonsoon (Jan-Mar); Summer (Apr-June); Premonsoon (July-Sep)
* Except pH and temperature all values expressed in mg^{-1}.

Table 2: Bacterial flora observed from textile effluent of four different seasons.

S.No.	Name of bacteria	Summer	Premonsoon	Monsoon	Post monsoon
1.	*Escherichia coli*	+	+	+	+
2.	*Enterobacter aerogens*	+	+	+	+
3.	*Klebsiella pneumoniae*	+	+	+	+
4.	*Proteus vulgaris*	+	+	+	+
5.	*Pseudomonas* sp.	+	+	+	+
6.	*P. aerogenosa*	+	+	+	+
7.	*Shigella sonnei*	+	+	+	+
8.	*Streptococcus faecalis*	+	+	+	+
9.	*Bacillus subtilis*	+	+	+	+
10	*Staphylococcus faecium*	+	+	+	+

+: Observed in all the months.

Table 3: Fungal flora observed from textile effluent of four different seasons.

S.No.	Name of fungi	Summer	Premonsoon	Monsoon	Post monsoon
1.	*Aspergillus flavus*	++++	+++	+++	+++
2.	*A. fumigatus*	++	+	+	+
3.	*A. luchensis*	++	++	++	+
4.	*A. niger*	+++	++++	+++	++++
5.	*Candida* sp.	+	+	-	+
6.	*Penicillium javanicum*	+++	++	++	++
7.	*Saprolespgia* sp.	++	+	+	+
8.	*Trichoderma viride*	+++	+++	++	+++
9.	*Alternaria* sp.	+	-	-	-
10.	*Fusarium oxysporum*	++++	+++	+++	+++
11.	*Rhodosporium* sp.	+++	+++	++	+++

++++ : Observed in all the months
+++ : Observed in above five months
++ : Observed in two to four months only
+ : Observed in one month only
- : Not observed

Table 4: Cyanobacterial flora observed from detergent effluent of four different seasons.

S.No.	Name of cyanobacteria	Summer	Premonsoon	Monsoon	Post monsoon
1.	*Oscillatoria salina*	+++	+++	+++	+++
2.	*O. chlorine*	+++	+++	-	+++
3.	*O. tenuis*	+++	++++	+++	+++
4.	*O. brevis*	+++	+++	+	+++
5.	*O. guttulata*	+	++	++	++
6.	*O. subbrevis*	+++	+	-	+
7.	*O. clarisentrosa*	+++	-	++	-
8.	*Synechococcus elongatus*	+++	++	++	++
9.	*Phormidium tenue*	++	++++	++	+++
10.	*P. ceylanicum*	++++	++	++	++
11.	*Lyngbya majuscula*	+++	+++	+++	+++
12.	*L. spiralis*	+	-	+++	-
13.	*Gleocapsa pleurocapsoides*	+++	+++	+++	+++
14.	*Plectonema radiosum*	+++	+++	+++	++
15.	*Nostoc communae*	+++	+++	++	+++
16.	*N. muscorum*	+++	+++	+++	+++
17.	*Chroococcus minutus*	+++	+++	+++	+++
18.	*Anabaena variabilis*	+++	+++	+++	+++
19.	*A. flosaquae*	++	+	+	+
20.	*Fischerella ambigua*	-	++	++	+

++++ : Observed in all the months
+++ : Observed in above five months
++ : Observed in two to four months only
+ : Observed in one month only
- : Not observed

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