

Physicochemical and fungal diversity analysis of two different sources of polluted water of Cachar District Assam

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ABSTRACT

Analysis of physicochemical and fungal species from two different sources of water pollution, as Paper Mill Effluents (PME) of Hindustan Paper Corporation and City Domestic Sewage (DS) of Cachar district, Assam was carried out during pre and post monsoon season (February, 2016 and May, 2016). Water samples were collected and analyzed for standard physicochemical parameters where pH, turbidity, electrical conductivity, total hardness were found within the limits prescribed by WHO and ISI where as total alkalinity, Total Dissolved Solids (TDS), nitrate and Phosphate content were found to be exceeding the permissible limits for both PME and DS. *Aspergillus niger*, *A. flavus*. and *Penicillium citrinum* were among the most commonly encountered species of fungi from both the sites. Highly significant microbial load was observed in domestic sewage (cfu/ml =22.0x10³, P<0.001) during the post monsoon season (May, 2016).

ABBREVIATIONS: PME: PAPER MILL EFFLUENT; DS: DOMESTIC SEWAGE

KEY WORDS: PAPER MILL EFFLUENTS, DOMESTIC SEWAGE, FUNGAL SPECIES, PHYSICO CHEMICAL PARAMETERS

INTRODUCTION

Water is the basic need of life, a precious gift of nature to man and all the living creatures of this universe. So the quality of water is of vital concern as it is directly linked with human welfare. Only 3% of the water in the universe is fresh and among the fresh waters, only around

5% or 0.15% of the total water are available for use. The total water resource available in India is 1850 km³, which is approximately 4% of the worlds fresh water resources (EPA-PWD, 2001) . But very fast it is becoming a scare commodity in many parts of the universe.

The water quality monitoring results obtained during 1995 to 2006 indicate that the organic and bacterial

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contamination are continued to be critical in water bodies. This is mainly due to discharge of domestic wastewater mostly in untreated form from the urban centers of the country and at the same time the receiving water bodies also do not have adequate water for dilution. Therefore, the oxygen demand and bacterial pollution is increasing day by day which is mainly responsible for water borne diseases.

The total surface water resource of Assam State is estimated at about 600 billion cubic meters, where the annual replenishable groundwater resource of the State has been estimated as 27.23 billion cubic meters. But prolonged discharge of industrial effluents, domestic sewage and solid waste dump causes pollution in the water resources which leads to enormous health problems. The rapid growth of industrial resources has further affected water quality due to overexploitation and improper waste disposal practices. Hence there is always a need for concern over the protection and management of water resources. Considering the above aspects of contamination, the present study was undertaken to investigate the level of water pollution and at the same time to measure the microbial contamination due to industrial and domestic exposure in the country rivers.

MATERIALS AND METHODS

SAMPLING SITE AND PREPARATION OF WATER SAMPLES

The present study was carried out during the pre and post monsoon season (February, 2016 and May, 2016). Raw paper mill effluent sample was collected from the outlet pipes in the local river Barak. For comparative analytical study domestic sewage was collected from Silchar, Municipal drainage system at Tarapur area, where all the debris and discharges of the whole locality have been found to be discharged. Both the effluent and sewage samples were collected in plastic gallons, pH was measured and stored at -20°C to prevent further microbial growth.

PHYSICO-CHEMICAL PARAMETERS AND MICROBIAL DIVERSITY STUDY

The paper mill effluent and domestic sewage were analyzed for a number of standard physico-chemical properties, including Color, temperature, pH, turbidity, Dissolved Oxygen (DO), Electrical Conductivity (EC), Total alkalinity (TA), Total Dissolved Solids (TDS), Total Hardness (TH), FCO_2 , Nitrate and Phosphate content were analyzed followed by the standard methods (USEPA, 1996; APHA, 1998).

For the isolation of fungi from both the effluent and sewage samples, serial dilution plate technique was

used. 0.1 ml of final dilution of sample was inoculated on Czapek Dox Agar medium. In sterile Petri dishes Martin's Rose Bengal was added as a bacteriostatic agent according to Smith and Dawson (1944). The tubes of colony forming units (cfu) was determined after incubating the inoculated plates in 3 replicates at 25°C ($\pm 2^\circ\text{C}$) for 4-5 days, thereafter fungal species were identified using the keys as described by Gilman (1957); Raper and Fennel (1965).

STATISTICAL ANALYSIS

Results were presented as Mean \pm SE followed by microbial diversity studies where mean value was calculated from three individual readings of a particular set. ANOVA was performed to determine the level of significance of microbial diversity studies. ANOVA was done using graph pad PRISM (Graph pad Inc., San Diego, CA, USA). Percentage occurrence used in this study include,

Occurrence of Species A

Percentage occurrence of species = $\frac{A}{I} \times 100$

Occurrence of all species

RESULTS AND DISCUSSION

The result of physicochemical parameters performed from both paper mill effluent and domestic sewage are presented in Table 1 and 2.

PH

pH is used to express the intensity of acid or alkaline condition of a water sample. In our case pH value ranged from 4.78-6.74, found within the permissible limits. Highly acidic pH was recorded in DS in both the seasons specially during the month of May. Water samples with low pH (<6.0) attributes to the discharge of acid contents into these effluents by agricultural or domestic activities. A decrease in pH may be caused by the increase in the amount of organic carbon, total carbonates available in the sewage resources. Though pH has no direct effect on human health still the different kinds of biochemical reactions taking place within human body are sensitive to variation of pH.

TURBIDITY

Turbidity is due to colloidal and fine dispersions in water resources. The turbidity value varied between 2.5-3.2 and found within the permissible limit of WHO and ISI.

DISSOLVED OXYGEN (DO)

Dissolved Oxygen is an important parameter for water quality assessment which reflects the biological pro-

Table 1. Physico chemical characteristics of PME and DS in the month of February, 2016.

Parameters	Paper Mill Effluent (PME)	Domestic Sewage (DS)	WHO Standards	ISI Standards
Color	Pale brown	Dark brown	-	-
Temperature	28	24	-	-
pH	6.16	4.78	6.5-8.5	6.5-8.5
Turbidity	2.5	2.8	5.0	10
Dissolved Oxygen (DO)	23.24	4.44	-	5.0
Electrical conductivity (EC)	0.9	1.9	1400	
Total Alkalinity	460	355	120	200
Total Hardness	214.67	138.67	500	300
FCO ₂	7.34	14	-	-
Total Dissolved Solids (TDS)	665	3695	1000	500
Nitrate	7.942	8.414	5	45
Phosphate	2.192	3.778	0.1	-

Table 2 . Physico chemical characteristics of PME and DS in the month of May, 2016.

Parameters	Paper Mill Effluent (PME)	Domestic Sewage (DS)	WHO Standards	ISI Standards
Color	Pale brown	Dark brown	-	-
Temperature	26	23	-	-
pH	6.74	5.12	6.5-8.5	6.5-8.5
Turbidity	2.7	3.2	5.0	10
Dissolved Oxygen (DO)	17.96	3	-	5.0
Electrical conductivity (EC)	1.2	2.1	1400	
Total Alkalinity	417	323	120	200
Total Hardness	298.67	202.66	500	300
FCO ₂	14.34	24.67	-	-
Total Dissolved Solids (TDS)	617	3432	1000	500
Nitrate	8.123	9.235	5	45
Phosphate	2.431	4.537	0.1	-

All parameters are in mg/L except Temperature, pH, Turbidity, Electrical conductivity. Temperature in °C, Turbidity in NTU, Electrical conductivity in micromho /cm

Table 3. Result of microbial (fungal) diversity isolated from PME and DS (February, 2016 and May, 2016).

Treatment Groups	cfu /ml (x 103)	
	February, 2016	May, 2016
Paper Mill Effluent (PME)(%)	4.67±0.17	5.67±0.45
10	9.34a±0.45	10.34b±1.2
25	10.67b±0.45	19.34a,b±0.75
50		
Domestic Sewage (DS) (%)	7.34±0.34	8.0±0.78
10	13.67c,b±0.45	12.67b±1.047
25	15.67c,b,a±0.75	22.0b,c,a±1.95
50		

Where, P < 0.05 = a, P < 0.01 = b, P < 0.001 = c

cesses taking place in water bodies. The DO value indicates the degree of pollution in water .In our study the DO content was found minimum during the month of May, which may be because the increased algal productivity due to increased rate of photosynthesis (Rajkumar *et al*, 2004).

ELECTRICAL CONDUCTIVITY (EC)

Electrical conductivity is a measure of water capacity to convey electrical current. It signifies the amount of total dissolved salts. EC value was found within the range of 0.9-2.1 micromho /cm where DS showed higher EC value in both the seasons indicating the presence of high amount of dissolved inorganic substances in the ionized

Table 4. Occurrence of fungi isolated from PME and DS (February, 2016 and May, 2016)

Fungal Species Identified	Water Samples											
	Paper Mill Effluent (PME)						Domestic Sewage (DS)					
	10%		25%		50%		10%		25%		50%	
	Feb	May	Feb	May	Feb	May	Feb	May	Feb	May	Feb	May
<i>A. flavus</i>	42.85	47.05	50	38.7	50	39.65	27.27	0	31.7	0	31.91	0
<i>A. niger</i>	0	41.17	0	48.38	0	44.82	45.45	44.45	39.02	50	40.42	56.06
<i>Penicillium citrinum</i>	28.57	11.76	21.42	12.9	25	15.51	9.09	0	9.75	0	10.63	0
<i>A. fumigatus</i>	0	0	0	0	0	0	18.18	51.85	19.51	42.1	17.02	31.81
<i>Mucor sp.</i>	14.28	0	17.85	0	15.625	0	0	3.7	0	7.89	0	6.06
<i>Rhizopus sp.</i>	14.28	0	14.28	0	9.375	0	0	0	0	0	0	0

form in domestic sewage as the conductance of water increases with salts. Higher the concentration of electrolyte in water the more is its electrical conductance.

TOTAL ALKALINITY

The alkalinity of water is caused mainly due to OH, CO₃, HCO₃ ions, borates, phosphates and organic acids. It is an estimation of the ability of water to resist change in pH upon addition of acid. In the present study, alkalinity was found beyond the permissible limits of both WHO and ISI standards. Maximum alkalinity may be due to low temperature bringing down the rate of decomposition of salts to a minimum thus by increasing alkalinity.

TOTAL HARDNESS

Water hardness is a measure of capacity of water to precipitate soap. It is defined as the sum of Calcium and Magnesium concentration, both expressed as Calcium carbonate. In the present investigation, the Total Hardness value (138.67-298.67 mg/L) ranged within the permissible limits with maximum hardness during summer (May), which might be due to reduced inflow of water and rate of evaporation. The FCO₂ value was recorded minimum in PME specially in February, 2010 which may be due to less suspended microbial load on suspended particles and algal masses (Michael, 1984).

TOTAL DISSOLVED SOLIDS (TDS)

TDS content was found within the range of 617-3695 mg/L where DS showed higher TDS content specially in pre monsoon season which exceeded the maximum permissible limit of ISI Standards indicating the enormous storage of different types of salts such as carbonates, bicarbonates, chlorides, phosphates, nitrates, magnesium calcium, sodium, potassium, manganese and organic matters.

Nitrate and Phosphate content were found at higher concentrations in domestic sewage specially in the summer season (May, 2010) where Nitrate and Phosphate level for both the effluents exceeded the permissible

limit of WHO Standards. Higher Phosphate concentration may be because of the discharge of domestic wastes, soaps, detergents, fertilizers and human activities.

All together six species of fungi belonging to four genera were isolated and identified from the water samples. The majority of fungi were encountered rarely. Of the genus *Aspergillus* the best represented species were *Aspergillus niger*, *A. fumigatus*, *A. flavus*, and *Penicillium citrinum* which were isolated from PME as well as from DS. The fungi isolated from different samples of Barak River were likely to be originated from soil or entered the water with plant remains. The isolated fungi from river Barak belong mainly to the category of transient accidental microorganisms, according to ecological classification of aquatic heterotrophic microorganisms (Park, 1972b). Transient and accidental microorganisms can develop sporadic activity and soil fungi may participate in microbiological processes in water bodies (Park, 1972b). Among the fungal population *Aspergillus* was represented by highest number of species. This is in accordance with the statement of Barron (1968) that *Aspergillus* is biologically one of the most successful of all fungi and is expected to occur on all sorts of organic debris. Carlie and Watkinson (1997) in their study observed that *Aspergillus* and *Penicillium* sp have cellulolytic enzymes and wood degrading capability. Cellulolytic filamentous fungi have the ability to penetrate cellulose substrate through hyphal extensions. cfu rate was found to be highly significant in DS (22.0x10³) especially in summer followed by ANOVA. Low cfu rate in February may be due to cold climatic condition inhibiting fungal growth. Maximum fungal diversity was recorded in domestic sewage in comparison to PME. Higher occurrence of *A. flavus* and *Penicillium citrinum*, was noted in PME where as *A. niger* and *A. fumigatus* were among the mostly encountered species in DS in both the seasons.

CONCLUSION

As the river water is being purified by the municipal corporations before public consumption, so human being

have least possibilities to get effected as the effluents and sewages get diluted in the river and after purification possibilities to develop water borne disease becomes negligible. But aquatic flora and fauna are directly getting exposed to the water of river Barak , might be tremendously effected by the exposure of this polluted water specially in the form of microbial contamination as no precautions have been undertaken yet for the safety of aquatic ecosystem including the aquatic plants and organisms specially fishes. So, proper measures have to be undertaken for the maintenance of water quality in Barak river, so that the water quality and aquatic life including the aquatic flora and fauna can be protected from microbial contamination and water pollution as well.

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