

Physico- Chemical Characteristic of Pulp and Paper Mill Effluent and its impact on Morphology and Behaviour of Fresh Water Catfish, *Clarias batrachus* (Linn.)



Shivani Srivastava

Research Scholar
Deptt. of Zoology,
Kamala Nehru Institute of
Physical and Social Sciences,
Sultanpur, U.P., India

Indu Singh

Associate Professor & HOD
Deptt. of Zoology,
Kamala Nehru Institute of
Physical and Social Sciences,
Sultanpur, U.P., India

Sadguru Prakash

Assistant Professor
Deptt. of Zoology,
M.L.K.(P.G.) College,
Balrampur, U.P., India

Abstract

The aim of the present study to evaluate the physico-chemical characteristic of pulp and paper mill effluent and their toxic impact on morphology and behavior of fresh water cat fish, *Clarias batrachus*. The pH, electrical conductivity and phenol were found within the desirable limits and colour, order, BOD, COD and chloride were higher the desirable limits of Central Pollution Control Board of India. During the study period, the colour of the experimental fish become progressively darker with increasing concentrations and was directly proportional to the concentrations of the test solution unlike those in the control. The opercular beat (number of beat / min) and surface activity of fish increase with increasing the concentrations of effluent and duration of exposure. Thus from the present study it may be concluded that air breathing catfish, *Clarias batrachus* is sensitive to pulp and paper mill effluent and can be used as indicators of effluent related stress in the water as it is evident from behavioural responses and morphological changes.

Keywords: Pulp and Paper Mill Effluent, *Clarias Batrachus*, Morphology, Behaviour

Introduction

Rapid urbanization and industrialization in India has resulted in the substantial increase in the liquid waste which is traditionally discharged in open land or into nearby natural water, causing a number of environmental problems including threat to plants and animal lives present in that environment. Many of the toxic substances released from these industries are lipophilic and weren't adversely affected by water. These substances accumulate in fish fatty tissues or become protein bound, so it is of importance to know the critical concentration above which human being are affected and the commercial fish species become unsuitable food. Industrial establishments are often located on the banks of rivers or other waterways where waste treatment facilities are lacking. They discharge their solid and liquid wastes directly or indirectly into the water bodies and finally find their way into the ocean. Paper and pulp mills use large volumes of water during scouring, bleaching, digesting, rinsing, cortication, lime treatment, and other finishing processes. In India, around 905.8 million m³ of water is consumed and around 695.7 million m³ of wastewater is discharged annually by this sector. Thus pulp and paper mills are normally located at the banks of rivers or lakes to ensure adequate supply of water and a means of disposing effluents, which are generally characterized by high concentrations of chemical oxygen demand, biological oxygen demand, suspended solids, extreme pH, and elevated temperatures.

Review of Literature

The effluents contain a complex mixture of bleaching agents, dyes, fluorescent whitening agents, different acids, coagulating agents, China clay, and many other organic and inorganic chemicals, including some metals and naturally occurring wood extractives, especially resin acids (Khan and Talukder, 1993). In India 34 large scale paper mills account for 51% of total capacity and 271 small paper mills account for the remaining 49%. Even with the most modern and efficient operational techniques, about 60 m³ of wastewater is generated during the production of a ton of paper (Thompson *et al.* 2001). In India, around 905.8 million m³

of water is consumed and around 695.7 million m³ of wastewater is discharged annually by this sector. The Indian pulp and paper industry is highly water intensive, consuming 100-250 m³ of freshwater per tones of paper, and also generates the corresponding wastewater, i.e. 75- 225 m³ per tone of paper (Pandey, *et al.*, 2015)

Ghaghra is one of the significant rivers of the northern India. It originates near the Manasarowar Lake in Tibet. The total length of Ghaghra River up to its confluence with the Ganges at Doriganj in Bihar is 1,080 kilometres. It is the largest tributary of the Ganges by volume and the second longest tributary of the Ganges by length after Yamuna. Its total catchment area is 127,950 sq km of which 45% is in India. Ghaghra River passes through Nepal where it is known as Kauriala. Rapti, Little Gandak, Sarda and Sarju are the major tributaries of this river. Fish are the important members of aquatic ecosystem and important sources of animal protein food for human. Fish lived in polluted ponds, lakes and rivers cannot avoid exposure to these chemicals that suspended or dissolved in water, being less than land animals to move to favourable regions to avoid unfavourable condition. The effluents discharged from paper mill makes its way into the river, thus polluting the river and making it unhealthy for the survival of aquatic organisms (Nath, 2016). These industrial pollutants alters the natural condition of aquatic medium that causes behavioural changes as well as morphological imbalance of aquatic organisms (Misra *et al.*, 2016).

Objective of The Study

The main objective and aim of the present study is to evaluate the physio-chemical Characteristics of pulp and paper mill effluent and to study its acute toxic effect on morphology and behaviour of fresh water cat fish, *Clarias batrachus* in the context of opercular beats, Surfacing, balancing and swimming activities.

Materials and Methods

The present study area Darshan Nagar, Ayodhya is situated in the east of Uttar Pradesh, India. It lies between 26°9' and 26°50' N latitudes, and 81°85' E longitude span of river Ghaghra. This river receives bleached and unbleached effluents discharged from Yes paper mill Ltd. Darshan Nagar, Ayodhya. Effluent samples were collected from pulp and paper mill in glass stoppered bottles at two sites i.e. point of origin of effluent (Po) and point of discharge (Pd) into river Ghaghra. The collected samples of effluent were transported immediately to the laboratory for analyzed the physical and chemical characteristics. The pH effluent was measured on sampling site using digital portable pH meter. The electrical conductivity (EC) of the effluent was measured on the sampling site using a digital portable conductivity meter at 25°C. The BOD, COD, Chloride and Phenols were measured according to APHA (1998).

Healthy specimens of Indian fresh water catfish, *Clarias batrachus* (45±5 g & 12±5 cm) were collected from local fish farm at Sultanpur, U.P. and were transported in containers to the laboratory. In the

laboratory, the fishes were carefully examined for any injury and then kept in 1% solution of KMnO₄ for few hours to get rid off dermal infection, finally they were kept in large plastic jar containing 50L of clean tap water and acclimatized for 15 days to the laboratory conditions, during which time they were fed on boiled egg yolk and commercial fish food. The acclimatized fishes were divided into six different groups each containing 10 fishes for the experiments. Group I served as control (0%). While group II, III, IV, V and VI exposed with 20%, 30%, 40%, 50% and 60%, respectively for the period of 96 hours to study the morphological and behavioural changes.

Results and Discussion

The results of physico-chemical analysis of effluent for different seasons are given in the Table 1. The values for various physico-chemical characteristics obtained in the present study are in good agreement with the characteristics of pulp and paper mill effluents as reported by other workers. The dark brown colour of pulp and paper mill effluent might be due to the presence of low and high molecular weight chlorinated organic compounds generated from lignin degradation products during wood cooking, conventional bleaching and alkali extraction of the pulp. The characteristic pungent odour of pulp and paper mill effluent was due to the presence of a number of dissolved chemicals (specifically mercaptans and hydrogen sulphide) and the raw materials which were used during the manufacture of paper. Similar colour and order of pulp and paper mill effluent were observed by Afroz and Singh (2014). Similarly, most workers have reported the colour of the discharged effluent was dark brown to light brown (Mahajan, 1985; Singh *et al.*, 1996). The pH of the effluent is an important factor because fluctuation in water pH affects the species composition and life processes of aquatic animal. It is a measure of the acidity or alkalinity of water and is one of the stable measurements. In the present study the pH of the effluent was ranged between 6.1-7.2 in all the seasons. The desirable limit of pH recommended by Central pollution control board of India for effluent are 5.5-9.0. Singh *et al.* (1996) reported pH range of 6.9 to 8.6 in the combined effluents of Shreyans paper mills Ltd., Ahmedgarh, Punjab. The mean electrical conductivity values in the present study from both sides ranged from 1.47-2.07 mD/cm in all seasons. This values of EC was within the maximum desirable limits (2.5mD/cm) of Central pollution control board of India. The values of electrical conductivity increased along with increasing concentration of effluents (Gagnetten *et al.*, 2007). Biological Oxygen Demand (BOD) represents the amount of oxygen used by the microorganism to decompose the organic material. Mean BOD values obtained in the present study from different sites ranged from 128.0 to 561.75mg/L. This values of BOD was higher the maximum desirable limit (30mg/L) of Central pollution control board of India. Chemical Oxygen Demand (COD), represents the amount of oxygen required for oxidizing the all organic matters. The maximum permissible limit of COD recommended

by Central Pollution Control Board of India is 250mg/L while maximum values of COD were found much greater than standard values at both sites which varied between 273.02-1549.03 mg/L. These values are higher than the range of 75 to 145 mg/L and 595 to 800 mg/L for BOD and COD, respectively as reported by Singh *et al.*, (1996). These differences may be due to variations in manufacturing processes, production capacity and efficiency of in-mill treatment plants as well as sites of effluent collection. Chloride concentration in water indicates the presence of organic waste, primarily of animal origin. The maximum permissible limit of chloride recommended by Central Pollution Control Board of India is 1000.0 mg/L while maximum values of chloride were found less than standard values at both sites which varied between 145-246 mg/L. The maximum permissible limit of phenolic compound recommended by Central Pollution Control Board of India is 1.0 mg/L while maximum values of phenol were found within the standard values at both sites which varied between 0.12-0.65mg/L.

The Exposure of fish to different concentrations of effluent showed changes in behavioral responses of test fish (Table2). The opercular activity (number of beat / min) of fish increases with increasing the concentrations of effluent and duration of exposure. Increased opercular beat may be due to the physiological adaption of fish in hypoxic conditions. In fishes, during hypoxic conditions, breathing rate (opercular activity) increases in order to compensate the decreased PO₂ level of blood. Similar observations have been reported by Singh *et al.*, (2011). In *Clarias batrachus*, surface activity increased with increase in concentration within 24 hours but decreases thereafter. Increase in surfacing and gulping of surface waters appears as an attempt to avoid breathing in the poisoned water. As a defense

mechanism to neutralize the toxic effect of effluent, copious amount of mucus is secreted by the fish gradually covers the whole body, gill and buccal cavity. Chocking of gills by mucus may also be one of the reasons for frequent surfacing of fishes. Among other factors the PO₂, PCO₂ and swimming space are also responsible to elicit surfacing in fish (Singh *et al.*, 2011). The restlessness and hyperactivity in fish may occur due to the inactivation of acetylcholinesterase, leading to accumulation of acetylcholine at synaptic junctions (Fulton and Key, 2001). In the last fish loses their balance and hang horizontally, finally they fell on the bottom, leading to death. Thus whole behavioural activity is affected due to the enzymatic system and loss of organ of balance in fishes.

The morphological changes in fish, *Clarias batrachus* exposed to different concentrations of effluent for 96 hours are shown in Table 3. The effluent exposed fishes show the excessive secretion of the mucus. The blood clotting was observed on the gill and body surface of fish. Many other changes occurs in the body of fishes due to exposure of effluent as changes in gill colour from dark red to dull red and skin become erupted. Thus from the present study it may be concluded that air breathing catfish, *Clarias batrachus* is sensitive to pulp and paper mill effluent and can be used as indicators of effluent related stress in the water as it is evident from behavioural responses and morphological changes. Although no symptoms of a particular disease may not appear in the present study in the fishes but repeated and chronic exposure of fishes to effluent may interfere with the physiological mechanisms leading to the development of stress. It may be dangerous in the future to the extent that threat may appear to the existence of the species. Thus timely scientific attention should be paid on the bio-monitoring of this fauna.

Table1

Physico-chemical characteristic of pulp and paper mill effluent collected from Pulp and Paper mill in Summer (Mar.-Jun.), Rainy (Jul.-Oct.) and Winter (Nov.-Feb.) seasons.

Characteristics	Seasons	Magnitude		Standard of CPCB of India for Effluents
		Site-Po	Site-Pd	
Colour	S R W	Dark Brown	Brown	Disagreeable
Odour	S R W	Pungent	Pungent	Disagreeable
pH	S R W	6.1±0.12 7.1±0.14 6.6±0.11	6.5±0.01 7.2±0.01 7.2±0.02	5.5-9.0
EC(mS/cm)	S R W	2.58±0.02 2.07±0.04 2.32±0.06	1.51±0.07 1.49±0.01 1.65±0.02	2.25
BOD(mg/L)	S R W	512.00±4.12 561.75±5.24 561.50±4.16	166.00±4.57 128.00±3.21 146.25±5.12	30.0
COD(mg/L)	S R W	1309.33±54.13 1327.76±82.13 1549.03±72.18	273.02±18.21 331.25±16.32 341.22±17.11	250.0

Chloride(mg/L)	S R W	246.00±11.10 224.50±12.14 229.00±13.12	174.00±5.54 145.25±6.41 154.00±7.12	1000.0
Phenol(mg/L)	S R W	0.53±0.02 0.65±0.01 0.64±0.04	0.12±0.02 0.15±0.06 0.15±0.05	1.0

Table2.

Changes In Behavioural Parameters In *Clarias Batrachus* Exposed To Up To 96hours At 20% Pulp Paper Mill Effluent

Exposure Period (Hours)	Opercular Beats/min	Surfacing	Loss of Balance	Swimming	Mucus secretions
0	+	++++	-	++++	-
24	+	+++	-	+++	+
48	++	++	-	++	++
72	+++	++	+	+	+++
96	++++	+	++	-	+++

+ = Less, ++ = Slightly higher, +++ = More higher, ++++ = Prominent, - =no change

Table 3

Effect of pulp and paper mill effluent on morphology of *Clarias batrachus* exposed for 96 hours.

Parameter (Colour)	Concentration of pulp and paper mill effluent					
	Control	20%	30%	40%	50%	60%
Gill	Dark Red	Red	Red	Dull Red	Blood Clotted	Blood Clotted
Skin	Normal grey	Grey	Dark grey	Dull Black	Few Black & white patches appears	More Black & white patches appear
Fin	Grey	Light Slimy grey	Moderate slimy grey	High Slimy grey	Grey and wounded	Grey and wounded

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