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RESEARCH ARTICLE

Toxic Effect of Bleached Kraft Pulp and Paper Mill Effluents on the River Water and It's Organism

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ABSTRACT- The present study deals with the physicochemical characteristics of river water Aami. A pulp and paper mill namely Rayana Paper Board Industries Ltd Khalilabad, Uttar Pradesh, were studied for sample collection and analysis of various pollution parameters. Three sampling sites have been selected for this study. The water of this river has toxic effects on fish *Channa punctatus*. The 96 h LC_{50} values of these sites are 8.99% dilution for site 1, 22.96 % dilution for site 2 and 41.15% dilution for site 3. Fishes were treated with different sub-lethal doses of water samples, it is shown significant alterations in different biochemical and haematological parameters of fish.

Key-words- Biochemistry, characteristics, effluents, Haematological, Pulp paper mill, Physicochemical, River Aami

INTRODUCTION

The nature and distribution of the flora and fauna in the aquatic system are mainly controlled by the fluctuations in the physical and chemical characteristics of the water body [1-4]. Paper and Pulp waste is serving threat to aquatic life because its effluent characteristically contains very high COD, BOD and very strong colour [5]. The presence of lignin in the waste which is derived from raw cellulosic materials and is not easily biodegradable makes the COD/BOD ratio of the waste very high.

The process water of pulp and paper mills contains approximately 700 organic and inorganic compounds [6,7]. Excessive loads of organic matter and inorganic nutrients also cause eutrophication within the receiving watercourses. The effluents contain a high concentration of suspended particulate material [8], dissolved and particulate organic matter; partly consist of cellulosic fibres and carbohydrates, which impair the oxygen balance of the water body, and sediments.

A multitude of the organochlorine compounds has been chemically characterized as lipophilic [9], meaning they are easily able to penetrate cell membranes and frequently possess high bioaccumulation factors. Another significant share of the toxic compounds in pulp and paper industry effluents is fatty acids and resin acids.

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In particular resin acids are considered the major factors in the toxicity of these effluents [10,11].

Due to the high chemical diversity of the organic pollutants in paper and pulp mill process water, a high variety of toxic effects on aquatic communities in recipient watercourses have been observed [12]. The pollutants concerned also kill fish or affect their reproductive physiology [13], or may induce male-biased sex ratios among fish embryos [14] on the exposure of fish to sub-lethal concentration of pulp and paper mill effluent. Baer *et al.* [15] were studied the effects of pulp and paper mill effluent on physiological and hematological endpoints in fingerling Largemouth bass (*Micropterus salmoides*). Basu *et al.* [16] said that Components of pulp and paper mill effluents contain neuroactive substances that may impair fish reproduction. Thus the objective of the current study was to evaluate the effect of pulp and paper mill effluent on the physicochemical characteristic of receiving water course and their effect on freshwater fish *C. punctatus*.

MATERIALS AND METHODS

Description of paper mill plant- A pulp and paper mill namely Rayana Paper Board Industries Ltd situated at Khalilabad, an industrial area of Gorakhpur region, India discharges its effluent into river Aami. This pulp and paper mill manufacture writing and craft wrapping paper, produced about 64 tones of the paper/day.

Water sample collection and Analysis- Water samples were collected from three sampling sites

Site- I: From the entry point of the effluents into river Aami

Site- II: 200 meters downstream from the site I
Site- III: 200 meters upstream from the site I

Measurement of physico-chemical parameters like biological oxygen demand (BOD), chemical oxygen demand (COD), total solids (TS) and suspended solid (SS) were done by the methods of APHA [17]; Trivedi & Goel [18]. The pH was recorded at the sampling site with the help of Water Analysis Kit.

Collection of experimental animal- Fish *C. punctatus* (wt. 29.21±1.83 gm; length 14.5±1.20 cm) were collected from the Ramgarh Lake of Gorakhpur district, India. The collected fishes were maintained in glass aquaria containing 100 l de-chlorinated tap water for acclimatization to laboratory conditions for 1 week. The water in aquaria was aerated continuously. The dead animals were removed from the aquaria to avoid any contamination.

Toxicity Experiment- Toxicity experiments were performed by the method of Singh and Agarwal [19].Ten experimental animals *C. punctatus* were kept in glass aquaria containing 6 litters dechlorinated tap water. Fishes were exposed for 24 hrs, 48 hrs, 72 hrs and 96 hrs at 4 different concentrations of bleached effluent collected from river Aami, control animals were kept in similar condition without any treatment. Each set of experiments, were replicated six times. Mortality was recorded after every 24h during the observation period of 96 h. Dead animals were removed to prevent the decomposition of body in experimental aquarium.
Toxicity data obtained from this study was computed through POLO computer program of Robertson *et al*. [20].

Biochemical Experiment- The biochemical experiments were performed by the method of Tripathi and Singh [21]. The bio-chemical experiment was conducted in freshwater ponds, 29.28 m^3 in area and 9.19 m^3 in water volume. The pond was stocked with 60 fishes with a size difference not greater than 1.5 times and then treated with different sub- lethal doses of water samples (9.5%, 10.5%, 15.5% respectively of site I, II and III). Fishes of the control group were kept in similar conditions without any treatment. After completion of 96 hrs the muscle and liver tissue in fish *C. punctatus* of treated as well as control group were quickly dissected out and used for bio-chemical estimations.

Protein- Protein levels were estimated according to the method of Lowry *et al.* [22] using bovine serum albumin as standard. Homogenates (10 mg/ml, w/v) were prepared in 10% TCA.

Total free amino acids- Estimation of total free amino acid was made according to the method of Spices [23]. Homogenates (10 mg/ml, w/v) were prepared in 95% ethanol, centrifuged at 6000 xg and used for amino acid estimation.

Nucleic acids- Estimation of nucleic acids (DNA and RNA) was performed, by methods of Schneider [24] using diphenylamine and orcinol reagents, respectively. Homogenates (1 mg/ml, w/v) were prepared in 5% TCA at 900C, centrifuged at 5000 xg for 20 min and the supernatant was prepared and used for estimation. Both DNA and RNA have been expressed as µg/mg tissue.

Glycogen- Glycogen was estimated by the Anthrone method of Van Der Vies [25]. In the present experiment 50 mg of tissue was homogenates with 5 ml of cold 5% TCA. The homogenate was filtered and 1.0 ml of filtrate was used for the assay.

Protease- Protease activity was estimated by the method of Moore and Stein [26]. Homogenate (50 mg/ml, w/v) was prepared in cold distilled water. Optical density was measured at 570 nm. The enzyme activity was expressed in µmol of tyrosine equivalent/mg protein/h.

Transaminases- Glutamic oxalic transaminase (GOT) and Glutamic pyruvic transaminase (GPT) activities were measured according to Reitman and Frankel [27]. Homogenates (100 mg/ml, w/v) were prepared in phosphate buffer for 5 minutes and centrifuged at 1000 g for 15 minutes and the supernatant was kept for estimation of enzyme activity.

Haematological Experiment- Hematological experiment was conducted in freshwater ponds, 29.28 m^3 in area and 9.19 m^3 in water volume. Each pond was stocked with 20 adult *C. punctatus* and then treated with different sub-lethal doses (5.5%, 5.5% and 8.5% of site I, II and III). No artificial feeding was carried out. All fish were examined after three weeks exposure. This parameter was measured according to Lehmann *et al.* [28].
Approximately 10µl of blood from each fish were used to make the blood smears. The smears were stained with May-Grunwald and Giemsa solutions. The major and the minor axes of 500 erythrocytes on each smear were measured by means by means of a light microscope (1000×magnification). The ratio of the major/minor axes plus the surface area of the erythrocytes were calculated. Two hundred leucocytes on each smear were determined.

RESULTS

Physico-chemical observation- The physicochemical parameters are considered as the most important principles in the identification of the nature, quality and type of the water for any aquatic ecosystem.
The pH serves as valuable guide for showing the acid alkali balance of water. Maximum level of pH was found for site I (9.3) followed by site II (8.6) and site III (7.5).
Suspended Solids consist of the particles of different types ranging from coarse cellulose fibers to fine colloidal particles of various organic complexes. Maximum value of suspended solid was found for site I (670 mg/l) followed by site II (530 mg/l) and site III (330 mg/l).

Total solids are the amount of solid present in dissolved and suspended form. Maximum level of total solid was found for site I (1180 mg/l) followed by site II (1070 mg/l) and site III (780 mg/l).

Biological Oxygen Demand **(**BOD) is the amount of oxygen used by the microorganism to decompose the organic material. Maximum level of BOD was found for site I (340 mg/l) followed by site II (255 mg/l) and site III (63 mg/l).

Chemical Oxygen Demand (COD)- represents the amount of oxygen required to oxidize all of the organic matter both biodegradable and non-biodegradable by a strong chemical oxidant like Potassium dichromate. Maximum value of COD was observed for site I (738 mg/l) followed by site II (572 mg/l) and site III (298 mg/l) (Table 1).

Pollutant levels of effluents were much greater than the limits as prescribed by the Central Pollution Control Board, Government of India (Suspended solid 50 mg/l–300 mg/l, Total solid 20 mg/l–1000 mg/l, BOD 30 mg/l, COD 250 mg/l, pH 6.5–8.4) for Site I and II. The pollutant level for site III was much lower than other sites (Table 1).

Table 1: Physico-chemical parameters of effluent and water samples collected from three different sites

Parameter	Site I	Site II	Site III
pH	9.3	8.6	7.5
Suspended Solid (mg/l)	670	530	330
Total Solid (mg/l)	1180	1070	780
BOD (mg/l)	340	255	63
COD (mg/l)	738	572	298

BOD= Biological Oxygen Demand
COD= Chemical Oxygen Demand

Toxicological Observation- The toxicity of effluents was time and dose dependent. There was a significant negative correlation between LC values and exposure periods. Thus, with increase in exposure periods the LC_{50} values decreased from 24.29% dilution (24 h) to 8.99 % dilution (96 h), from 31.77% (24 h) to 22.96% dilution (96 h) and from 49.22% dilution (24 h) to 41.15% dilution (96 h) for site I, II and III respectively (Table 2). The LC_{50} values of site III were much higher than site I and site II, so it was cleared that the water sample collected from site III was less toxic than site II and site III.

Table 2: Toxicity (LC values) of bleached effluent collected from site I, II and III against *C. punctatus* at different time intervals

	Site 1		**Site 2**		**Site 3**	
Exposure Periods	**LC_{50} values**	**Limits LCL-UCL**	**LC_{50} value**	**Limits LCL-UCL**	**LC_{50} value**	**Limits LCL-UCL**
24h	24.29	18.83-41.36	31.77	29.68-35.10	49.22	47.46-53.56
48h	20.51	15.65-37.21	28.70	26.86-31.03	43.55	42.19-48.03
72h	13.55	10.60-19.27	25.81	23.82-27.68	37.29	35.20-41.03
96h	8.99	6.60-11.20	22.96	20.86-24.55	41.15	29.55-38.68

LCL - Lower confidence limit
UCL - Upper confidence limit
LC_{50} - Lethal concentration for 50 percent of the exposed fish

Biochemical Observation- Sub-lethal doses of water samples caused significant alterations in the level of total protein, total free amino acids, glycogen and nucleic acids and activity of enzyme protease and transaminases in the liver and muscle tissue of fish. Total protein level was reduced to 56% and 53% of control, Nucleic acid level such as DNA level was reduced to 52% and 40% of control, similarly, RNA level was reduced to 48% and 40% of control, Glycogen level was reduced to 70% and 62% of control, while The total free amino acid level was induced to 255% and 200% of control, respectively in liver and muscle tissues. The activity of transaminases Glutamic oxalic transaminase (GOT) was increased up to 114% and 108% of control, whereas Glutamic pyruvic transaminase (GPT) level was increased up to 186% and 108% of control, Protease activity was significantly increased 141% and 157% of control respectively in liver and muscle tissues of fish after 96 h exposure to sub lethal dose of effluent collected from Site I (Table 3). Similar trend of result were observed for site II and site III (Table 3).

The value of all biochemical parameters of site III was nearly same to control value, so it was cleared that the water sample collected from site I and site II have more profound effect on biochemical parameters of fish *C. punctatus.*

Table 3: Changes in Protein, Amino acids, Nucleic acid, Glycogen, Protease and Transaminases in liver and muscle tissue of *Channa punctatus* after sub-lethal doses of bleached effluent collected from site I, II and III after 96 h

Parameter	Tissue	Control	Site I	Site II	Site III
Protein (µg/ mg)	Liver	136±0.01 (100)	76.5±0.01* (56)	83.8±0.01* (62)	130±0.02* (95)
	Muscle	154±0.01 (100)	82±0.005* (53)	90±0.003* (58)	143±0.01* (92)
Amino acid (µg/ mg)	Liver	5.5±0.02 (100)	14.0±0.01* (255)	11.5±0.02* (209)	5.8±0.01* (105)
	Muscle	7.5±0.005 (100)	15.0±0.01* (200)	13.5±0.01* (180)	8.1±0.002* (108)
DNA (µg/ mg)	Liver	27±0.002 (100)	14±0.004* (52)	15.3 ±0.02* (57)	26±0.02* (96)
	Muscle	23.5±0.01 (100)	9.4±0.005* (40)	12±0.01* (51)	22±0.001* (94)
RNA (µg/ mg)	Liver	30.5±0.01 (100)	14.8±0.02* (48)	16.3 ±0.006* (53)	28.9±0.003* (95)
	Muscle	27.2±0.005 (100)	10.8±0.01* (40)	13.2±0.005* (49)	25.5±0.002* (93)
Glycogen (mg glycogen/g of tissue)	Liver	4.41±0.01 (100)	3.1±0.01* (70)	3.5±0.01* (79)	4.3±0.005* (97)
	Muscle	3.4±0.01 (100)	2.1±0.003* (62)	2.30±0.01* (68)	3.4±0.001* (94)
Protease (µmoles pyruvate/g tissue)	Liver	0.968±0.001 (100)	1.37±0.003* (141)	1.13±0.005* (116)	1.03±0.002* (106)
	Muscle	0.812±0.002 (100)	1.28±0.01* (157)	1.16±0.01* (142)	104±0.002* (128)
GOT (µmoles pyruvate/mg protein/hr)	Liver	5.82±0.004 (100)	6.66±0.02* (114)	6.46±0.01* (110)	6.0±0.002* (103)
	Muscle	5.22±0.01 (100)	5.68±0.01* (108)	5.45±0.004* (104)	5.35±0.01* (102)
GPT (µmoles pyruvate/mg protein/hr)	Liver	6.24±0.003 (100)	11.6±0.01* (186)	11.3±0.01* (181)	6.5±0.003* (106)
	Muscle	5.78±0.01 (100)	6.26±0.003* (108)	6.08±0.01* (105)	5.82±0.002* (101)

Values are mean ± SE of six replicates
Values in parentheses are % change with control taken as 100%
Data were analysed through student's 't' test
***Significant (P<0.05), when treated groups were compared with controls**

Haematological Observation

Hematological study is important for toxicological research, environmental monitoring of fish and their health conditions during culture because the fish generally are so intimately associated with the aquatic environment. On the exposure of bleached effluent after three weeks, Lymphocyte value was reduced, monocyte value was nearly same and value of granulocyte total was higher than control group. The mean size of the erythrocytes was different, the major axis of erythrocytes was significantly longer, while the minor axis was significantly shorter than control groups, and this means that their cells were more elongate. As a consequence, the ratio of the major and the minor axis were also different; ratio was higher than the control group. However the surface area of the cells did not differ (Table 4). The value of haematological parameters of site III was lower than site I and site II.

Table 4: Effect on differential leucocyte counts and the size of erythrocytes of *C. punctatus* after sub-lethal doses, 5.5% dilution of site I and site II and 8.5% dilution of site III, of bleached effluent after three weeks

Parameters	Control	Site I	Site II	Site III
	Leucocytes			
Lymphocyte,%	70.53±0.006	62.83±0.007	65.16±0.005	67.22±0.006
Monocyte,%	10.15±0.005	10.31±0.006	10.02±0.005	10.08±0.002
Granulocyte total	16.72±0.007	20.26±0.008	18.96±0.007	17.22±0.005
	Erythrocytes			
Major axis µm (ranges)	10.93±0.006 (9.00-17.00)	13.23±0.007 (12.00-17.00)	12.12±0.006 (10.0-16.0)	11.55±0.002 (9.0-15)
Minor axis µm (ranges)	7.43±0.005 (6.00-10.00)	7.12±0.006 (6.00-9.00)	7.28±0.006 (6.0-9.0)	7.18±0.005 (6.0-9.0)
Major axis/Minor axis (ranges)	1.47±0.002 (1.25-2.55)	1.85±0.001 (1.55-2.75)	1.66±0.002 (1.35-2.55)	1.53±0.006 (1.25-2.45)
Surface area µm (ranges)	80.21±0.008 (60.0-110.0)	81.11±0.007 (65.0-108.0)	79.06±0.007 (63.0-105.0)	80.38±0.001 (64.0-109.0)

Results are expressed as mean ±SE of six replicates

DISCUSSION

Physicochemical Observation- The hydro biological study is a pre-requisite in any aquatic system because the nature and distribution of the flora and fauna in the aquatic system are mainly controlled by the fluctuations in the physical and chemical characteristics of the water body [3;29-31]. The physico-chemical parameters are considered as the most important principals in the identification of the nature, quality and type of the water (fresh, brackish, saline) for any aquatic ecosystem [1].

pH serve as a valuable guide for showing the acid alkali balance of water. Aquatic organisms are affected by pH because most of their metabolic activities are pH dependent [32,33]. Optimal pH range of sustainable aquatic life is pH 6.5–8.2 [34]. On receiving the bleached effluent pH of river water is alkaline because during the bleaching process many chlorinated compounds released, which makes the effluent alkaline.

Solids (Suspended solid and total solid) are very useful parameters for describing the chemical constituents of the water and can be considered as a general of edaphic relations that contribute to productivity within the water body [35]. The toxicity of an effluent is related to its potential to cause oxygen depletion, which is due to the presence of high amount of toxic organic matters.

COD represents chemically oxidisable load of organic matter in water. BOD determination is a most useful technique to assess the level of organic pollution in the river system [36]. High BOD was due to the presence of lignin and much organic material, which was discharged by pulp and paper mill.

The present study showed wide variations between three different sites. This variation was mostly according to the chemical constituents. Site III has the lowest values of physicochemical parameters and was considered the weakest toxicant in comparison to other sites.

Toxicological Observation- The data obtained from present study indicates that the pulp and paper mill effluents bring out behavioral changes, in fish *C. punctatus*. By changing a large number of behavioral responses, fishes try to resist the change in aquatic environment and reduce the harmful effect of stem bark extracts. These included changes in skin color and start scratching their nostril at the bottom of the aquarium and frequently came at the water surface for gasping air. The nature and rapidity of onset of fish behavioral responses indicate that the effluent become active at the neuromuscular system of fish *C. punctatus*. Animal behavior is a neurotropically regulated phenomenon, which is mediated by neurotransmitter substances [37]. The stressful breathing behavior exhibited by fish may be as a result of respiratory impairment due to effect of toxicant on the gills.

Mortality caused by the pulp and paper mill effluent showed a significant positive correlation between dose and mortality. The positive correlation between dose and mortality was noted because the increased concentration of toxic chemicals, present in pulp and paper mill effluent in aquarium and pond water resulted in more intake or entry of toxic chemicals in the body of animals. This trend is also dependent upon several factors such as, rate of penetration, nature of slope, variability and maximal effects of active chemicals [38].

Biochemical and haematological Observation- Data of biochemical observation, indicates that after exposure to 96 h of fish with sub-lethal doses of effluent caused significant ($P<0.05$) decreased in the level of total protein, glycogen, nucleic acids and enhancement in total free amino acid level, protease as well as significant ($P<0.05$) enhancement in the activities of transaminases (GOT & GPT) in liver and muscle tissue of fish (Table 2).

The depletion of protein fraction may have been due to their degradation and possible utilization of degraded products for metabolic purposes. Depletion in tissue proteins of fishes due to the low rate of protein synthesis under metallic stress was also reported by several workers. The enzyme protease functions in hydrolyzing proteins to free amino acids and small peptides. Increased protease activity in the body tissue corroborates the enhancement in the free amino acid level, the formation of which might be the result of protein hydrolysis in the tissues suggesting stimulation during toxic stress. Similar trend of results of protease activity was also reported by several workers in different animals as *Tilapia mossambica*, *Pila globosa* and various mammals. Increase in free amino acids level was the result of the breakdown of protein for energy requirement and impaired incorporation of amino acids in protein synthesis. It also attributed to lesser use of amino acids and their involvement in the maintenance of an acid-base balance. The decrease in the glycogen content in tissues indicates its rapid utilization by the perspective tissues as a consequence of toxic stress felt by the animals during the experiment. Inhibition of DNA synthesis might affect both protein as well as amino acid levels by decreasing the level of RNA in protein synthesis machinery. Similar results were also observed by Yadav *et al*. [39]. Transaminases are released during cellular damage or lysis. Stress conditions induce elevation in the transamination pathway. Many other workers were also observed the increased transaminases activity in freshwater fishes exposed to various toxicants. The increase may also due to damage of the organs resulting in increase protein and CHO metabolism as suggested by Nemcso'k *et al*. [40].

Hematological studies help in understanding the relationship of blood characteristics to the habitat and adaptability of the species to the environment. Many factors such as environmental and physiological are known to influence fish hematology; these include stress due to capturing, transportation, sampling, age and sex. The decline in leucocyte counts reported by Tana and Nikunen [41] is probably attributed to reduction in the number of circulating small lymphocyte, which may be resulted in the reduced resistance of stress fish to diseases. Sizes of erythrocytes increas than the control because the animal was in stress so consume large amount of O_2 (Table 3). Red blood cells are normally more elongated when they get older. The red blood cell count of *Clarius gariepinus* was reported to have increased significantly by Annue *et al*. [42] when the fish was subjected to zinc treatment. They attributed the red blood cell elevation to blood cell reserve combined with cell shrinkage as a result of osmotic alteration of blood by the action of metal. In fish blood, oxygen is carried in combination with haemoglobin and this was very important for survival of the fish. These results are also supported by many workers [43-45].

CONCLUSIONS

On the basis of the present investigations, it may be concluded that Rayana Paper mill effluent has several toxic compounds and responsible for water pollution and the high mortality rate and deleterious consequences on the health of fish subjected to exposure of pulp and paper mill effluent. The effluent of pulp and paper mill industries also has a profound effect on the biochemical aspects like haematological and muscle profile, with particular reference to energy metabolism in freshwater fish *C. punctatus*.

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REFERENCES

[1] Mohamed HA. Physico-Chemical characteristics of ABU ZA'BAAL ponds Egypt. Egyptian J. Aqua. Res., 2005; 31(2): 1-15.

[2] Parra O, Karrasch B, Cid H, Mehrens M, Paheco P, et al. Effects of pulp and paper mill effluents on the microplankton and microbial self-purification capabilities of the Biobio River, Chile. Science of the Total Environment, 2006; 359: 194-208.

[3] Sundaramanickam A, Sivakumar T, Kumaran R, Ammaiappan V, Velappan R. A comparative study of physico-chemical investigation along parangipettai and cuddalore coast. J. Environ. Sci. Tech., 2008; 1(1): 1-10.

[4] Lynda S, Rouabhi R, Berrebbah H, Djebar M. Survey of the physicochemical quality of the waste waters of Biskara city rejected in Chobat Roba, Messdour and Wadi Z'ommer (Algeria). African J. Environ. Sci. Tech., 2008; 2(8): 231-38.

[5] Yadav SK. Human health implications due to water toxicity by pulp and paper mill. J. Hum. Ecol., 2006; 20(2): 91-96.

[6] Mall ID. Waste management in pulp and paper industry: A system perspective. IPPTA, 2000; 12: 4.
[7] Swamy YV, Mohanty JN, Murty BVR, Tripathy HK, et al. Trearment of black Liquor and Utilisation of Waste Material Generated in Small Paper Plant. IPPTA, 2000; 12 (2): 53-60.
[8] Karels AE, Niemi A. Fish community responses to pulp and paper mill effluents at the southern Lake Saimaa, Finland. Environ Pollut., 2002; 116: 309-17.
[9] Fahraeus-Van Ree GE, Payne JF. Enzyme cytochemical responses of mussels (*Mytilus edulis)* to resin acid constituents of pulp mill effluents. Bull Environ Contam Toxicol., 1999; 63: 430-37.
[10] Mohn WW, Stewart GR. Bacterial metabolisms of chlorinated dehydroabietic acids occurring in pulp and paper mill effluents. Appl Environ Microbiol., 1997; 63: 3014-20.
[11] Yu Z, Stewart GR, Mohn WW. Apparent contradiction: Psychrotolerant bacteria from hydrocarbon-contaminated arctic tundra soil that degrade diterpenoids synthesized by trees. Appl. Environ. Microbiol., 2000; 66: 5148-54.
[12] Zahrim AY, Melissa L, Gilbert J, Jidaon J. Treatment of pulp mill effluent using photo Fenton's process. Journal of Appl. Sci., 2007; 7(15): 2164-67.
[13] Van den, Heuvel MR, Ellis RJ. Timing to exposure to a pulp and paper effluent influences the manifestation of reproductive effects in rainbow trout. Environ Toxicol. Chem., 2002; 21: 2338-47.
[14] Larrson DGJ Forlin L. Male-biased sex ratios of fish embryos near a pulp mill: temporary recovery after a short-term shutdown. Environ Health Perspect, 2002; 1170: 739-42.
[15] Baer Kevin N, Bankston Cristy R, Mosadeghi Sasan and Schlenk Daniel. The effects of pulp and paper mill effluent on physiological and hematological endpoints in fingerling Largemouth bass (*Micropterus salmoides*). Drug Chem. Toxicol., 2009; 32(1): 59-67.
[16] Basu Niladri, Chieu Anh Ta, Waye, Andrew, Mao, Jinquin, Hewit, Mark, Arnason John T and Trudeav Vance L. Pulp and paper mill effluents contain neuroactive substances that potentially disrupt neuroendocrine control of fish reproduction. Environmental Sci. Technol., 2009; 43(5): 1635-41.
[17] APHA. Standard methods for the examination of water and waste water. 20th ed. New York, USA; American public health association, 1998.
[18] Trivedi RK, Goel PK. Chemical and biological methods for the water pollution studies. Env. Publ, 1984.
[19] Singh A, agarwal RA. Possibility of using latex of Euphorbiales for snail control. Sci. Total Environ., 1988; 77: 231-36.
[20] Robertson JL, Russel RM, Preisler HK, Saven ME. Bioassay with Arthopods: Polo: A new computer programme. CRC. Fransis and Taylor, 2007; 1-224.
[21] Tripathi PK, Singh A. Toxic effect of alphamethrin (synthetic pyrethroid) on oxidative metabolism of freshwater snail *Lymnaea accuminata* In: (Gargh, S. L.Ed.) Proc. Inter. Cong. Chem. Environ, 2001; 238-43.
[22] Lowry OH, Rosebrough NJ, Farr AL, Randall RJ. Protein measurement with folin phenol reagent. J. Biol. Chem., 1951; 193: 265-75.
[23] Spies JR. Calorimetric products for amino acids. In: Methods in Enzymology. Calowick, S.P. and Kaplon, N.O. Eds., Academic Press, 1957; 468.
[24] Schneider WC. Determination of nucleic acids in tissue by pentose analysis. In: Enzymology (Calowick, S.P. and Kaplon, N.O. Eds.). Academic Press, 1957; 680.
[25] Van der, Vies J. Two methods for the determination of glycogen in liver. Biochem. J., 1954; 57: 410-46.
[26] Moore S, Stein WH. A modified ninhydrin reagent for the photometric determination of amino acids and related compounds. J. Biological. Chemi., 1954; 211: 907-13.
[27] Reitman S, Frankel S. A calorimetric method for the determination of glutamic-oxaloacetic and glutamic pyruvic transaminase. Am. J. Clin. Pathol., 1957; 28: 56-63.
[28] Lehmann J, Sturnberg FJ, Hesse F. Hematological serologic Substratuntersuchungen an der Regenbogenforelle (Salmo gairdneri, Rich.) III. Normwerter des Hematogramms unter Beruksichtingung alters-und jahreszeitlich bedingter Schwankungen. Gewasser Abwasser, 1976; 59: 1-32.
[29] Rajasegar V. Physico-chemical characteristic of the Vellar estuary in relation to shrimp farming. J. Environ. Boil, 2003; 24: 95-101.
[30] Singh SK, Singh D. Water quality of River Ganga at Ghazipur district. Bioved, 2004; 15(1,2): 99-105.
[31] Kamal K, Joshi B, Deepali D. Physico-chemical characteristics of pond water at Khanpur village in Bareilly District (U.P). Him. J. Adv. Zool., 2005; 19(1): 89-92.
[32] Wang W, Wang A, Chen L, Liu Y, Sun R. Effects of pH on survival, phosphorus concentration, Adenylate energy. Charge and Na^{+}-K^{+} ATPase activities of *Perracus chinemis* Osbelk juveniles. Aqua. Toxico, 2002; 60: 75-83.
[33] Fagbenro OA. Tilapia fish for thought Inaugural Lecture Series 32. Federal University of Technology, Akure, 2002; 77.
[34] Murdoch T, Cheo M O'Laughbin K. Stream keepers field Guide watershed inventory and stream monitoring methods, Adopt-A-Stream foundation. Everett, WA, 2001; 297.
[35] Goher MEM. Chemical studies on the presipitation and dissolution of some chemical elements in Lake Qarun. Ph.D. thesis. Fac. Sci. AI-Azhar Univ. Egypt., 2002.
[36] Sabata BC, Nayar MP. River pollution in India. A case study of Ganga River. Pub. Corp. New Delhi, 1995.
[37] Bullock TH, Kand R, Grinnella A. Introduction to nervous system. W.H. Freeman and Company, San Francisco, 1977.
[38] Goodmann LS, Gillman AG, Rall TW, Murad F. The pharmacological basis of Therapeutic. Macmillan Publishing Company, New York, 1985.
[39] Yadav RP, Tiwari S, Singh A. Toxic effect of taraxerol extracted from Codiaeum variegatum stem-bark on target vector snail *Lymnaea acuminata* and non target fish. Iberus, 2005; 23(1): 1-13.
[40] Nemcso kJ, Benedeczky I, Boross L, Asztalos B, Orban L. Subcellular localisaton of transaminase enzymes in fishes and their significance in the detection of water pollution. Acta Biol. Szeg., 1981; 27: 9-15.
[41] Tana J, Nikunen E. Impact of pulp mill effluent on egg hatchability of pike (*Esox lucius* L). Bull Environ Contam Toxicol., 1986; 36: 738-43.
[42] Annune PA, Ebele SO, Olademeji AA. Acute toxicity of cadmium to juveniles of *Clarias gariepinus* (Teugels) and *Oreochromics niloticus* (Trewawas). J. Environ. Sci. Health, 1994; 29: 1357-65.
[43] Jeney Z, Valtonen S, Tellervo E, Galina J, et al. Effects of bleached kraft pulp and paper mill effluent on physiological parameters of roach (*Rutilus rutilus*) infected

by the digenean *Rhipidocotyle fennica*. Folia Parasitologica., 2002; 49: 103-08.

[44] Ovie KS, Jeg A, Avworo UM. Sublethal effects of some selected hematological parameters of Heteroclarias (A hybrid of *Heterobranchus bidorsalis* and *Clarias genepinus*). Int. J. Zool. Res., 2006; 2: 77-83.

[45] Kori-Siakpere Ovie, Oghoghene Ubogu Ewoma. Sub lethal hematological effects of Zinc on the freshwater fish, Heteroclarias sp. (Osteeichthyes: Clariidae). African J. Biotech., 2008; **7**(12): 2068-73.

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