

IMPACT OF VARIOUS INDUSTRIAL EFFLUENTS ON WATER QUALITY OF RIVER AAMI AND ITS EFFECT ON BIOCHEMISTRY OF FRESH WATER INHABITING FISHES

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ABSTRACT

River Aami is known as Lifeline of Magahar, originates from Sikhaira Tal, Siddharth nagar. The river passes through, Siddharth Nagar, Basti, Sant Kabir Nagar and Gorakhpur. It traverses approximately 102 Kms. and ultimately confluences in the River Rapti near village Sohagaura of tehsil Bansgaon of district Gorakhpur. During its journey, it receives industrial effluents at several locations. Some of them are Rayana Paper Board industries limited, Khalilabad and GIDA in Sahjanva which is chain of several industries namely textile, steel plants, india glycol limited (IGL) etc. The aim of present study was to evaluate the extent of toxicity posed on the river water due to various industries situated on the river bank and its influence on biochemistry of fishes and for this study *Cyprinus carpio* was selected as this fish is widely

present and inhabit Aami river and its availability is adequate. Several important sites of river Aami were selected that mark the boundaries of the flow root of this river and exoeriment was set eventually. The physico-chemical properties and parameters were tested and taken in consideration to define the quality of water. It was found that, the temperature, pH, colour odour, TS (total solids), TSS (total suspended solids), DO(dissolved oxygen), BOD(biochemical oxygen demand), COD(chemical oxygen demand),was found much different and were not in accordance with recommended tolerance limit by WHO. In biochemical profiling, an alteration in blood and serum parameters was clear indication of the physiological stress imposed on fishes by pollution i.e. decrease in level of protein, AchE (acetylcholine esterase) and increase in level of glucose and Transaminase enzymes (sGOT and sGPT).

INTRODUCTION

Water is such a important entity required for our survival on earth but despite of it, water is poorly managed resource of the world.^[1] Besides drinking, water is also used for pisciculture and aquaculture, irrigation, hydroelectricity generation etc. But now-a-days water is becoming more and more unfit for consumption of mankind as well as other organisms thriving their life on water due to mismanagement.^[2]

The increasingly obvious effects of pollution of the biosphere in general and aquatic ecosystems in particular are known to everyone and are the subject of daily accounts in the popular press as well as textbooks^[3,4] and scientific monographs.^[5,6] The main source of aquatic pollution include industrial waste, mining activities, sewage and waste water, marine dumping, burning of fossil fuels, accidental oil leakage, global warming, atmospheric deposition, urban development etc. Many of the pollutants especially, industrial waste water and effluent discharges are lipophilic which is insoluble in water but is able to cross lipid bilayer of cell membrane of aquatic organisms inhabiting the water and adversely affecting the aquatic biodiversity. The industrial effluent has caused great impact on water quality and also these toxins can cause a host of health issues, from cancer to hormone disruption to altered brain function. Children and pregnant women are particularly at risk.

Pollutants generally produce relatively rapid changes in characteristics of fish population.^[7,8] This was made clear by the by the findings from the experiments and researches done on fishes which were found to inhabit water which received industrial wastes from industries situated in Mugpoo, Darjeeling. Findings showed a significant alterations in morphology as well as internal structure and function of these organisms.^[8] Fishes serve as important biological indicators of water quality and can highlight the potential dangers of new chemicals introduced in the aquatic environment, because, fishes respond to the toxicant in a manner similar to higher vertebrates. They play different roles in the trophic web, undergoing bioaccumulation of pollutants, biotransformation of xenobiotics through cytochrome P450 – dependent oxidative metabolism and respond to mutagens at low concentration.

Some other forms of pollutants and contaminants in the form of spills, waste disposal sites, and abandoned facilities, their primary contaminants are known as below:

- ❖ Fuel hydrocarbons
- ❖ Chlorinated ethenes

- ❖ PCBs and polychlorinated dibenzo-p-dioxines (PCDDs) from wastes of pesticide manufacturing
- ❖ Methylmercury from contaminated soils and wastewater
- ❖ Radionuclides from former nuclear weapons test sites and
- ❖ Radioactive waste repositories and nitroaromatic explosives from ammunition plants to name just a few. Discarded materials are, however, often not well characterized and heterogeneous.^[9]

Rivers of India do not have proper arrangements of industrial waste and effluent disposal, as a result of which rivers as well as other freshwater sources are at the verge of devastation in the form of aquatic pollution and have caused great loss to biodiversity and fresh water habitants, even they have imposed great threat to human health.^[10] This is true for the river aami which is known as Lifeline of Magahar, which originates from Sikhaira Tal, Siddharthnagar. The river passes through, Siddharthnagar, Basti, Sant Kabir Nagar and Gorakhpur. It is presently affected severely by industrial pollution ever since the establishment of paper mill at Khalilabad, distillery and sugar unit at Rudhauri at GIDA near Sahjanwa in 1989. As the river has been the life line of the population located in the nearby villagers across its serpentine length of 126 km from Sohnara to Sogaura, it is necessary to look into the qualitative aspect of river water in time and space.

So, the aim of present work is to trace the physical and chemical parameters of water present in the river and hematological as well as biological assessment of the fishes flourishing in that habitat and altogether the effect of this heavy pollution on the health status of the fishes. For this, fish *Cyprinus carpio* was selected, as this fish is widely and easily available in this river as well as shows great adaptation and resilience to the course of time with the uniform increment of pollution day by day.

MATERIAL AND METHOD

Study area and selected sites for study: Gorakhpur is a city in the eastern part of the state of Uttar Pradesh in India, near the border with Nepal. River Aami is very important river of this region and traverses approximately 102 Kms. and ultimately confluences in the River Rapti near village Sohgaora of tehsil Bansaon of district Gorakhpur. It is tributary of Rapti river on its right bank receives industrial effluents at several locations in a major stretch starting from Rudhauri to its confluence point at Sohgaora into Rapti river and serves as a lifeline for

the people of Siddarth Nagar, Sant Kabir Nagar, Basti and Gorakhpur district in Eastern Uttar Pradesh.

The river water, is presently affected severely by industrial pollution ever since the establishment of paper mill at Khalilabad, distillery and sugar unit at Rudhauli at Gorakhpur industrial development area (GIDA), Sahjanva, which is an industrial stretch in Gorakhpur which is important not only for its contribution in Indian economic growth but this rapid growing industrial area has got an important environmental impact which is none other than polluted river Aami, which is backbone of agricultural as well as household needs for people inhabiting near this area. GIDA has many factories which are ranked on state as well as national level and some of them are IGL (India Glycol Limited), Parle, ARP (Azam Rubber Products) footwear as well as powerlooms, plywood and the only jute mill in Uttar Pradesh. The stretch of the river from Rudhauli, Basti to Sohgauna, Gorakhpur has been identified as a polluted stretch.

As per records, approximately 12 million litres per day (MLD) sewage is contributed in river Aami. According to Uttar Pradesh Pollution Control Board (UPPCB), Industrial discharge in the River is taking place through 03 drains. Out of 3 drains 1 drain is mixed drains which carry treated industrial effluent as well as untreated sewage.

3 sites or the areas of river were chosen for the study which come under the sources of industrial discharge in the river water from Sahjanva to the final confluence point of Aami to Rapti river at Sohgauna and further work was carried out. The areas chosen for the study are as follow and Water samples were collected from the sites chosen in plastic containers in undisturbed stage. The sites selected for study are below:

Site 1: part of river present near India glycol limited, GIDA, Sahjanva, Gorakhpur.

Site 2: river flowing through Adilapur village, where all the different types of industrial effluents are dumped together in the river such as paper wastes, alcoholic chemical, jute and textile mill effluents, metal processing waste materials, hospital and pathological effluents etc.

Site 3: confluence point of 2 rivers i.e. aami and rapti near Sohgauna, where aami meets rapti river and thus, water of river thus obtained having physico- chemical properties derived from both the rivers.

Sampling was done from the depth ranging from 30 to 45 cm at the study sites in summer, rainy and winter seasons at the regular intervals. Proper care was taken to avoid any disturbance due to the loose sediments and rocks present in the river water. It was kept in mind, in order to avoid entry of unnecessary air bubbles the lid was applied in the river itself after sample bottle was filled up to the brim. The collected samples were brought immediately to the laboratory and physico-chemical properties of water samples were estimated with the help of methods provided in APHA manual of water and waste water management, 2005.^[11]

Samples collected were evaluated by the Total Solids (TS), Total Suspended Solids (TSS), Dissolved Oxygen (DO), Carbon Dioxide (CO₂), Biochemical Oxygen Demand (BOD), Chemical Oxygen Demand (COD).

Other parameters such as pH, temperature, colour and odour was noted down on the spot itself before the collection of samples.

Biochemical study

- 1. Collection of fishes:** The adult fishes of same weight and same length approximately and of same species were captured. It was kept in mind that the species captured were native to that area. Capturing of fish was done with the help of fisherwoman accompanied and afterwards, they were brought in department laboratory as soon as possible.
- Diseased as well as injured fishes were removed as soon as possible and experiment was conducted on their blood and tissue samples to assess the alterations due to biochemical parameters included in the study.
- 10 healthy fishes of nearly approximately same weight and same length of *Cyprinus carpio* was captured with the help of cast net and brought as soon as possible to natural product laboratory, DDU Gorakhpur University for assessment of the various biochemical parameters included in the present study. This fish belongs to one of the largest members among the cyprinidae family of common carp. Its peculiar feature which can be seen to identify this fish are having two pairs of barbells on each side of upper jaw and the colour is brassy yellow with serrated dorsal and anal fin. This fish was selected because they are the major inhabitants of the river and also show greater level of adaptation and resilience towards the present scenario and status of the river water i.e. heavy intoxication and contamination due to industrial effluents of the factories and mega industries.

- 2. Experimental plan:** to show the potential effects inserted on the biochemical prospects of fish due to industrial effluents and toxicants analysis and was shown by the set of biochemical experiments the blood as well as tissues (liver and muscle) collected from the fishes and parameters were random glucose, transaminase enzymes such as SGOT and SGPT, acetylcholine esterase and free amino acids in the fishes captured from all the sites chosen among routes of river Aami. Afterwards, the obtained results were compared with the fishes from comparatively less polluted water body named control group.

Biochemical assessment

Fishes were on the spot captured from the sites with the help of fisherwoman assisted from village side for capturing the fishes. All the fishes were sacrificed and blood was collected first in microcentrifuge tubes by puncturing the caudal vein. The blood was kept in anticoagulant free microcentrifuge tubes for serum collection. Samples were centrifuged at 8000 rpm for 15 minutes and stored in freezer at - 20°C for further biochemical analysis.

Later on, Liver and muscle tissues were dissected out and further biochemical experiments were carried out. First of all, homogenates were made according to the protocol given.

- Total protein estimation was evaluated by the method described by Lowry et al., (1951)^[12] in the serum samples of fishes captured from all the three sites mentioned above for the study
- Total free amino acids by Ninhydrin method described by Spies, (1957)^[13] in the serum obtained from the blood of fishes.
- Glycogen level estimation by anthrone method given by van der Vies, (1954).^[14] 40 mg of each liver and muscle tissue was homogenized by adding 5% TCA by the help of electric homogenizer and filtered. 1 ml of filtrate was taken and 4ml of 30% KOH was added and mixture was boiled for nearly 20 minutes till clear solution was obtained. Excess alkali was neutralized by adding 5ml of GAA (glacial acetic acid) and distilled water was added for maintaining total volume of upto 10 ml. The tissue homogenate thus obtained was subjected for analysis.
- Determination of transaminase enzymes GOT (glutamate oxaloacetate transaminase) and GPT (glutamate pyruvate transaminase) i.e. Liver Function Test, method devised and described by Reitman and Frankel, (1957).^[15] Was done in blood serum obtained from the fishes.

- Glucose concentration was measured by the method given by Seist et al., (1981)^[16] in the blood serum.
- Acetylcholine esterase activity was measured in blood serum by the method provided by ellman et al.^[17] the enzyme activity was expressed in $\mu\text{g SH hydrolysed/min/mg protein}$.
- **Statistical analysis:** Each experiment was repeated atleast six times and the values are expressed as mean \pm SE (standard error) of six replicates. For visualizing significant changes student' t test and anova (analysis of variance) was applied.

RESULTS AND DISCUSSION

The results of physico - chemical analysis of the river aami is tabulated given below of the three seasons summer, rainy and winter of year 2019-20 in table -1 of three different sites chosen and the result of biochemical assessment of fishes collected from the sites are presented in table 2.

Table 1: Physical and chemical properties of water sample collected from sites included in present study of river aami of year 2019-20, summer season (march 2019-june 2019), rainy (july 2019-august 2019) and winter season (december 2019 –february 2020).

Properties	season	Chosen sites			Standard value set by CPCB (central pollution control board)	Reference value set by WHO (world health organisation)
		Site 1	Site2	Site 3		
Colour	summer rainy winter	Reddish brown Light brown Reddish brown	Blackish brown Light brown Blackish brown	Earthy Earthy Earthy	Not applicable	Not applicable
Odour	summer rainy winter	Alcoholic Alcoholic Alcoholic	Pungent Pungent pungent	Muddy Muddy muddy	Not applicable	Not applicable
Temperature	summer rainy winter	29.43 \pm .04°C 28°C 20.04 \pm .17°C	29.8 \pm .16°C 28.04 \pm .26°C 20.8 \pm .14 °C	28.9 \pm .03°C 27.8 \pm .30°C 21.2 \pm .32°C	Should not exceed 5°C than receiving water temperature.	
pH	summer rainy winter	8.2 \pm .04 8.07 \pm .04 8.74 \pm .04	8.3 \pm .08 8.3 \pm .08 8.8 \pm .12	7.8 \pm .05 7.4 \pm .04 7.8 \pm .14	5.5-8.5	6.5-8.5
Total solids(TS) (mg/L)	summer rainy winter	1869 \pm 4.4 1807 \pm 5.2 1863 \pm 11.8	1750.33 \pm 3.2 1713.33 \pm 3.3 1773.67 \pm 5.8	1617.67 \pm 4.9 1588.67 \pm 7.7 1418.67 \pm 9.5	20- 1000 mg/L	500- 1500 mg/L
Total	summer	507 \pm 2.1	512.67 \pm 5.7	372.34 \pm 11.02	100 mg/	100mg/L

Suspended Solids (TSS) (mg/L)	rainy winter	470 ± 14.7 496 ± 3.1	492.34± 4.02 561 ± 8.6	369.67± 4.6 367.67± 5.3	L	
DO (Dissolved Oxygen) (mg/L)	summer rainy winter	2.07± .12 2.3± .13 2.17± .05	1.7 ± .08 2 ± .08 1.7 ± .17	2.7 ± .05 4.3 ± .08 4.1 ± .08	4 mg/L	-
COD (Chemical Oxygen Demand) (mg/L)	summer rainy winter	690.43 ± .5 649.5 ± .9 671.5 ± 1.8	710.5 ± 1.5 698.27± 1.8 708.67 ± 2.05	394.03 ± 3.4 280.2 ± 4.5 365.87 ± 1.65	250 mg/L	250 mg/L
BOD (Biochemical Oxygen Demand) (mg/L)	summer rainy winter	149.6 ± 5.8 118.4± 4.09 162.4± 3.4	179.96 ± .7 160.73±1.06 175.83±2.65	98.04 ± 1.7 95.57 ± .7 96.6 ± 2.97	30 mg/L	30 mg/L

Table 2: Biochemical profile in blood and tissues of the fishes captured from river aami from the sites chosen during November 2019 to January 2020.

Biochemical parameters		CONTROL	Experimental sites		
			Site -1	Site- 2	Site -3
Length (cm)		15 ± .5	12 ± .5	11 ± .62	13 ± .67
Weight (g)		85 ± 2.5	80 ± 2.5	80 ± 2.6	81 ± 2.5
Random blood glucose (mg/dl)		60.33 ± .39	79.16 ± 2.09	104.2 ± 4.68	61.28 ± .7
sGOT (serum Glutamate Oxaloacetate Transaminase enzyme) (µmoles pyruvate /mg protein/ h)		40.9 ± .57	67.266 ± 1.32	73.2 ± 1.93	52.44± .5
sGPT (serum Glutamate Pyruvate Transaminase enzyme) (µmoles pyruvate /mg protein/ h)		45.47 ± .57	62.26 ± .93	72.32 ± .70	49.66 ± .207
AChE (µm SH hydrolysed /min/mg of protein)		.055	0.0278 ± .003	0.0186	0.0428
Protein (µg/mg)		125.23 ± .45	89.24 ± 1.64	90.3± .76	121.9 ± 2.01
Glycogen (mg/g of tissue)	Liver	61.42 ± .47	48.5 ± 1.26	44.94± 2.9	61.88± .725
	Muscle	75.26 ± .58	64.52 ± 4.57	66.32 ± 2.6	72.08 ± .42
Amino acid (µg/mg)		39.75 ± .27	44.36 ± 3	56.86 ± .76	34.52 ± .7

Values are expressed as mean ± SE (standard error) of the replicates.

The results of physico-chemical analysis of the river Aami is been tabulated in table 1 above of three different sites and then, biochemical assessment was carried over in blood and tissue of fishes collected from the sites which is also presented in table 2. The study has been done in the time interval between march 2019 to feb 2020. The assessment of the results obtained is been done on the basis of the three seasons of the year 2019-20 i.e. summer (march 2019 – june 2019), rainy (july 2019- august 2019) and winter (November 2019- February 2010).

It is clear from the table 1 that the colour of water purely depends on the type of effluent discharged by the industry present on bank of river. The colour change is the indication of heavy pollution imposed in the water body. Light penetration in the depth of the water body is affected and in turn growth of flora and fauna is altered because light is needed for the growth of plants and photosynthesis and as a result of this fishes thriving on phytoplanktons find difficulty to survive in such a hostile situation. Site 1 and site 2 have very dark colour of effluent discharge suggesting a heavy pollution whereas in site 3, river water is in natural muddy or earthy colour. In accordance with river water norms it is suggested that, there should be no noticeable colour in river water. Thus, it can be suggested that river Aami is heavily polluted at all the sites selected for the study.

On the basis of the odour, it can be drawn that river aami is facing adverse condition, which can be seen in the light of smell observed in all the sites. The relevant norms for class a water suggest that there should be no offensive colour present in the water body. Hence, it also indicates the heavy pollution in site 1 which is IGL (india glycol limited, sahjanva), having alcoholic smell and site 2 water having pungent smell.

Temperature of the river water varied for all the sites and for different seasons. Temperature recorded was high in summer for site 2 then, in site 1 and at last in site 3. Whereas, lowest was recorded in winter season for site 1. The water temperature is one of the important physical property of water ecosystem and depends on the quality, types of nutrients and habitat surrounding the water body. It affects the growth and survival of the flora and fauna. Increase in temperature water causes growth of the microbial activity. also, any increase in water temperature causes hormonal fluctuations in fishes in turn, altering the reproduction and spawning in the fish species. The increase in temperature of water is caused by the type industrial effluents discharge in the form of waste by products by ongoing chemical reactions by heavy machines, heavy metals and toxic chemicals.

pH data noted in all the different sites showed variations for different sites with changes in season with highest or alkaline pH in site 1 and site 2 and lowest pH for site 3 which is near neutral. the pH is an important factor in deciding the quality of water. The water which has pH value above neutral pH 7, then, it means that water is contaminated by strong base NaOH or any other base and also pH data provides the information about the type of contaminants discharged in the water body are either acidic or basic.

Amount of total solids are also different in all the three places in different seasons. Their level is above the reference value assigned by CPCB (centre of pollution control board) and WHO (world health organisation) suggesting water is highly contaminated by industrial discharge by chain of industries of GIDA (Gorakhpur Industrial Development Area). The total solids and total suspended solids level hampers growth of aquatic organisms and plants. They are solid byproducts of waste materials discharged by industries. Maximum amount of total solids were present in site 1 in summer season and total suspended solids are also present in site 2 in winter season.

Dissolved oxygen plays a very important role in sustaining the aquatic life. In any case, to support aquatic life, it should not be less than 3.5 mg/l. In the light of this fact, it was least in site 2 in summer as well as in winter season. In site 1 and 2, site 3 water sample has higher concentration of DO. Although, roughly we can say that in rainy season DO is highest as compared to summer and winter season.

BOD is the oxygen requirement to decompose the organic matter. The permitted level of BOD assigned by CPCB and WHO is 30 mg/l. Above that, the water quality deteriorates rendering it unfit for human consumption and hampers the survival of organisms. From the data in table 1. It is clear that BOD values are higher in water sample of site 2, then, in site 1, and, among that, it is higher in summer and winter season, site 3 possesses a lower BOD.

COD (chemical oxygen demand) is a measurement of oxygen required to oxidize soluble and particulate organic matter present in water. The reference range of COD according to CPCB and WHO is 250 mg/l, while in the analysis of river Aami water, the values found were much higher. In all the three sites. Also, the values were found to be high in summer and winter season, as compared to rainy season and site 1, site 2 possesses a very high amount of COD in its water in comparison to site 3.

So, in totality, on the basis of all the physico-chemical tests performed in the water sample from river Aami, it can be clearly suggested that site 1 IGL (India Glycol Limited), Sahjanva and site 2 Chataye bridge appear highly polluted in comparison of site 3 Sohagaura, the confluence point of the 2 river Aami and Rapti.

The biochemical parameters in fishes caught directly from all these sites of river aami were analysed and it suggest that there is stress imposed by fishes due to heavy industrial effluent which can be clearly depicted from the data presented in table 2.

The random blood glucose level was found to be elevated in the fishes captured from the sites. It was significantly higher in *Cyprinus carpio* which was selected for the study and blood glucose was found to be 79.16 ± 2.09 and 104.2 ± 4.68 mg/dl in site 1 and 2 fishes respectively and it was low in site 3 fishes and lowest in control group of fishes .According to previous study on pollution and stress physiology, it is suggested that under the stress condition body of the fishes emits immediate response which is perceived by CNS (central nervous system) as a result, stress hormones like cortisol, catecholamines (adrenaline and epinephrine) are relesed in blood stream by endocrine system. Finally, leading to increase in blood glucose to fuel the entire tissue of organism leading to a condition known as hyperglycemia. The rise in blood glucose is mediated by cortisol mediated gluconeogenesis.^[18] The increase in level of glucose for higher needs of osmoregulation and an important source of energy for maintaining fish homeostatsis during chronic stress.^[19]

Also, activity of specific enzymes for are also found decreased such as LDH (lactatye dehydrogenase), PFK (phospho fructokinase), citrate kinase, which are enzymes of carbohydrate metabolism i.e. glycolysis, as per the studies done by Bedi and Kenan (2005), Choudhary etal., (2004) and Almeida etal., (2001).^[20,21,22]

The activity of transaminase enzymes sGOT/AST (Serum Glutamate Oxaloacetate Transaminase/aspartate aminotransferase) and sGPT /ALT (Serum Glutamate Pyruvate Transaminase/ alanine aminotransferase) indicate the impact of pollutants on fish health. generally, these enzymes are distributed in the cells of vital organs such as liver, kidney, heart and gills. But an increase in their level in blood serum is an indication of tissue injury. Hence, it act as bio-marker to test the tissue injury and liver dysfunction. Its level was found more in fishes caught from site 1 and site 2 than site 3, suggesting higher level of pollution in these 2 sites in comparison with control.

Previous studies done on T. zilli and M. capito in Qaran lake in year 2019 suggest that under influence of different heavy metals and pollutants causing physiological and endocrinological stress, ultimately, leading to damage of tissue with concomitant liberation of transaminase enzymes into circulation.^[23]

There was an observable difference in level of acetylcholine esterase in fishes obtained from different sites. Site 1 and site 2 which are heavily polluted have acetyl choline esterase 0.0278 ± 0.003 and 0.0186 respectively which is lower than that of site 3 which is 0.0428 , suggesting that acetylcholine serve as very important biomarker to assess the pollution status. Acetylcholine esterase enzyme is vital for regulation of neurotransmitter acetylcholine in sensory and neuromuscular system of fish and other organisms.^[24] Its main function is to break acetylcholine and acetic acid and then, to facilitate nerve impulse transmission from one cholinergic neuron to next one.^[18] Due to its inhibition, there is an spurt in the level of acetylcholine in circulation causing prolonged neural stimulation finally leading to tetany and death of organism.

Proteins play an important role in physiologically and biochemically. Enzymes which are engaged in metabolic pathways are in the form of protein and these proteins are made up of several blocks carbon compounds called amino acids which has amino group and carboxylic group. In the table 2, it is clear that site 1 and site 2 fishes show lower amount of protein content i.e. 89.24 ± 1.64 and 90.3 ± 0.76 $\mu\text{g}/\text{mg}$ respectively, than site 3 fishes which have 121.9 ± 2.01 $\mu\text{g}/\text{mg}$ of protein content, suggesting that pollution causes decline in protein content. This decline in protein content is due to catabolism of protein into amino acids to cope up with the hostile environment due to effluent stress.

It can be clearly lined out from the data of amino acid concentration of fishes caught from sites showing just opposite the previous discussion of protein content. The amino acid concentration is more in site 1 and 2 i.e. 44.36 ± 3 $\mu\text{g}/\text{mg}$ and 56.86 ± 0.76 $\mu\text{g}/\text{mg}$ respectively than site 3 fishes having 34.52 ± 0.7 $\mu\text{g}/\text{mg}$. The reduction in protein content indicates that tissue protein undergoes proteolysis which results in production of amino acids and are used in TCA cycle for energy production during stress condition.^[20]

In the studies done previously on river aami fishes have shown that protein content decrease with the increase in pollution stress.

Liver act as reservoir of glycogen content and it is also present in muscle. Under stress condition or emergency need of energy, the body begins to break this stored glycogen to derive energy. Glycogen is a readily accessible storage form of glucose. Muscle glycogen is not used because it lacks enzyme glucose 6- phosphatase which is required to pass glucose into blood stream.^[25] On the basis of experimental analysis it was found that in all cases, liver

has low amount of glycogen than muscle and out of that site 1 and site 2 have low glycogen content than site 3 fishes. As compared to site 3 fishes, site 1 and site 2 fishes have more energy requirement to thrive in such a hostile environment.

CONCLUSION

River Aami is the tributary of river rapti flowing in eastern part of Uttar Pradesh majorly from city Gorakhpur. It is backbone of the rural people dependent on. It is clear from the above set of experiments and biochemical analysis that condition of the river is not good and it is deteriorating day by day causing people and terrestrial and aquatic animals to face a kind of stress affecting their survival. People who depend on the river for fishing and agricultural activities have to face various problems due to deteriorated quality of the river water. However, seriousness and depth of this problem have realised in previous years and many work as well as media attention is drawn towards this direction for its conservation from various industrial and domestic pollutants. Hence, the present work is also an attempt made in this direction for conservation of this river by analyzing the river water qualities by physical and chemical parameters and biochemical experiments carried on fish *Cyprinus carpio* and *Mystus vittatus*, a major inhabitant of river in this region shows great adaptability and resilience. I hope, this will provide current health status of this river and its animals for making some conservation programmes in spreading awareness towards our natural resources and sustainable development for future generations.

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