

Full Length Research Paper

Biochemical markers and histopathology of the target tissues of *Labeo rohita* reared in freshwater lakes of Bangalore, Karnataka, India

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The present study was aimed on the assay of the biochemical composition, activity of acid and alkaline phosphatase (as biomarkers) and histopathological changes in liver, kidney and brain tissues of *L. rohita* reared in the freshwater lakes of Bangalore. This study was conducted for a period of 15 months and fish sampled from Yellamallappa chetty lake (B) and Vengai lake (A) to record the variations in the organic constituents of the tissues. It revealed a significant reduction in the levels of proteins and glycogen and a marked decrease in enzymatic activities whereas cholesterol levels showed a marked increase in the tissues of test fish sampled from lake B compared to control fish from Hebbal fish farm. The acid and alkaline phosphatase enzymatic activities are known to be involved in several cellular metabolic reactions. Investigations conducted earlier in the fish sampled from similar conditions of sewage (Hebbal lake) and industrial (Chowkalli lake) polluted lakes showed necrotic condition of hepatocytes and hepatopancreatic cells in liver and shrunken glomerulus and degenerated proximal and distal tubular epithelial cells in kidney with similar changes in biochemical profile. Therefore the present study also suggests that the industrial effluents from lake B caused cellular damage which might have in turn induced hypoglycemia and metabolic acidosis. The tissues obtained from the lake A did not show significant variation when compared to those of control from farm.

Keywords: *L. rohita*, biochemical composition, industrial pollutants, acid and alkaline phosphatase, histopathology.

INTRODUCTION

Water pollution is recognized globally as a potential threat to both human and other animal populations which interact with the aquatic environments (Biney *et al.*, 1987; Svensson *et al.*, 1995). A number of organic and inorganic wastes in industrial and domestic effluents are responsible for water pollution. Fish in natural and aquaculture freshwater and marine systems are exposed to pollutants, pathogens and xenobiotic substances which causes stress to these aquatic organisms. The conventional laboratory toxicity studies cannot be extrapolated to the natural environment because they lack ecological realism (Benson and Black, 1990). Variability and interaction of environmental factors in

natural habitats complicate the responses of organisms to contaminants (Adams *et al.*, 1992 and 1996). The water parameters such as, high BOD, pH, COD, TDS, nitrates, phosphates and free ammonia besides toxic metals cause deleterious effects on the aquatic biota. This complicated interaction between all these parameters affects the pathological and immunological homeostasis of the fish. Thus it requires a detailed understanding of the immunological responses of fish to known multiple stressors and it has been a question mark for the biologists. Various scientists have showed considerable interest as to how these stressors interact to affect and disrupt metabolic activities at the biochemical level causing changes in biologically important enzymes (Verma *et al.*, 1981). Fish are excellent subjects for the study of various effects of contaminants present in water samples since they can metabolize, concentrate, and

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Table 1. Changes in physico-chemical parameters of Water of Control fish farm, Vengai lake (A) and Yellamallappa Chetty lake (B).

Analysis	Standard (Maximum Limits)	Control	Lake A	Lake B
Temperature (°C)	22-28	22	21	22
pH	06.50 - 08.5	7.37	7.8	8.84
Turbidity (NTU)	05 to 25	12	22	28
TDS	500-2000	842	954	1800
TSS	200	150	220	380
COD	2	2.66	2.8	7.14
BOD	6	13	22	112
DO	4-6	4	3	2
Nitrates	45	11	16	40
Nitrites (ppm)	10	12	18	30.1
Chlorides	200	252.2	280	442.4
Salinity	200	400	470	800
Phosphates	2	2.4	2.7	3.6
Total alkalinity	200	270	420	900
Total acidity	20	22	38	60
Iron	0.3	0.36	0.88	2.2
Zinc	5	6	6.42	9

Except Temperature, pH, conductivity, Turbidity, Nitrates all values are in mg/l.

store water borne pollutants. The development of biological monitoring techniques based on fish offers the possibility of checking water pollution with fast responses on low concentrations of direct acting toxicants (Poele and Strik, 1975; El- Shehawi *et al.*, 2007).

Adequate energy reserves are required by organisms to mediate the effects of stress (Lee *et al.*, 1983) and to serve as energy buffers during periods of harsh environmental conditions and food shortages (Adam and Mclean, 1985). Any disturbance in the external environment is first reflected in the biochemical composition of cells. Cell inclusions such as glycogen, proteins and lipids are the energy reserves available for a cell for its activities. Changes in these components indicate a change in the metabolic activities due to an external stress. Pollution monitoring methods using enzyme inducement or enzyme dispersion in fish or other aquatic organisms have been proposed for studying polluted environment (Verma *et al.*, 1979). Studies on acid and alkaline phosphatase activities in *Channa punctatus* exposed to mercuric nitrate (Jeelani and Shaffi, 1989), *Heteropneustes fossilis* exposed to fenvalerate (Johal *et al.*, 2002) and *Labeo rohita* exposed to domestic sewage (Rajan, 1990) have been reported. Phosphatases are good indicators of stress condition in the biological systems (Verma *et al.*, 1980). Though number of literature is available regarding the effect of some pollutants and pesticides on enzyme systems in different fishes, studies on the effect of industrial effluents on the phosphatases system in wild fishes reared in lakes are very scarce.

The present investigation is an attempt to elucidate the comparative effect of different pollutant sources on liver, kidney and brain of wild Indian major carp, *Labeo rohita* reared in the Vengai lake (receiving domestic sewage) and Yellamallappa chetty lake (receiving pharmaceutical industrial effluents) for commercial purpose. Histological examination of liver and kidney tissue of *L. rohita* sampled from Hebbal lake (sewage polluted) and Chowkalli lake (industrial effluents polluted) showed remarkable structural changes as was observed in the studies conducted in our lab. *L. rohita* being the prime cultured species in poly-culture practices in India, occupy a prominent position in the aquatic system, hence the impact of sewage and industrial effluents polluted lake on the phosphatases system and at cellular level of this candidate species was chosen for the study.

MATERIALS AND METHODS

Water samples were collected from Hebbal fish farm (control site) and Vengai lake (receiving domestic sewage - A) and Yellamallappa chetty lake (receiving pharmaceutical industrial effluents - B). The water was collected in the morning at about 09.00 to 10.00 AM once every fortnightly for a period of 15 months and the physico-chemical parameters like temperature, pH, BOD, COD, DO, TDS, chloride, free ammonia, phosphates, sulphates, nitrates and alkalinity were determined by standard method of APHA, (2005). The water parameters of the three lakes Hebbal fish farm (Control), Vengai

Table 2. Levels of protein, glycogen and cholesterol in the different tissues of *Labeo rohita* from Fish farm (control), Vengiah lake (A) and Yellamallappa Chetty Lake (B)

Tissue	Biochemical constituent	Control	Lake A	Lake B
Liver	Protein	41.66 ± 1.75	39.00 ± 1.41 (-6.39%)	30.33 ± 1.63 ^b (-27.2%)
	Glycogen	4.26 ± 0.23	4.05 ± 0.23 (-4.93%)	2.42 ± 0.26 ^a (-43.19%)
	Cholesterol	2.6 ± 0.17	2.8 ± 0.13 (+7.6%)	4.1 ± 0.17 ^c (+57.6%)
Kidney	Protein	26.50 ± 1.52	23.40 ± 1.09 (-11.70%)	17.83 ± 0.75 ^c (-32.72%)
	Glycogen	2.58 ± 0.16	2.31 ± 0.21 (-10.4%)	1.93 ± 0.14 (-25.19%)
	Cholesterol	1.9 ± 0.08	2.0 ± 0.11 (+5.2%)	3.2 ± 0.09 ^c (+68.4%)
Brain	Protein	11.6 ± 1.03	10.17 ± 0.75 (-12.33%)	8.9 ± 0.73 (-23.27%)
	Glycogen	1.81 ± 0.08	1.68 ± 0.08 (-7.18%)	1.32 ± 0.12 (-27.62%)
	Cholesterol	1.3 ± 0.17	1.4 ± 0.15 (+7.7%)	1.7 ± 0.18 (+30.76%)

Values expressed mg/g wet weight of tissues,
 Values are expressed as mean ± S.E; sample size (n) = 6,
 Values given in parenthesis are % change over control,
 (-) indicates % decrease over control and (+) indicates % increase over control,
 Super scripts **a**, **b** and **c** indicate significance at 5%, 1% and 0.1% levels respectively,

Table 3. Levels of Acid Phosphatase and Alkaline Phosphatase in the different tissues of *Labeo rohita* from Fish farm (control), Vengiah lake (A) and Yellamallappa Chetty Lake (B)

Tissue	Enzyme activity	Control	Lake A	Lake B
Liver	ACP	8.46 ± 0.28	8.30 ± 0.09 (-1.89%)	7.26 ± 0.21 (-14.18%)
	ALP	16.61 ± 0.33	15.75 ± 0.37 (-5.18%)	12.9 ± 0.30 ^c (-22.34%)
Kidney	ACP	5.85 ± 0.35	5.43 ± 0.10 (-7.18%)	4.55 ± 0.18 (-22.22%)
	ALP	12.96 ± 0.12	12.03 ± 0.15 (-7.17%)	10.48 ± 0.31 ^b (-19.13%)
Brain	ACP	3.58 ± 0.25	3.33 ± 0.29 (-6.98%)	2.78 ± 0.12 (-22.34%)
	ALP	9.62 ± 0.25	9.07 ± 0.18 (-5.82%)	7.76 ± 0.15 ^c (-19.33%)

Values expressed as μ moles PNP/mg of protein/30min wet. weight of tissues,
 Values are expressed as mean ± S.E; sample size (n) = 6,
 Values given in parenthesis are % change over control,
 (-) indicates % decrease over control,
 Super scripts **a**, **b** and **c** indicate significance at 5%, 1% and 0.1% levels respectively,

lake (A) and Yellamallappa chetty lake (B) is given in Table 1. In the present investigation the test fish were

sampled from lake A and B with the help of drag net and brought to the lake bank alive from the three water bodies

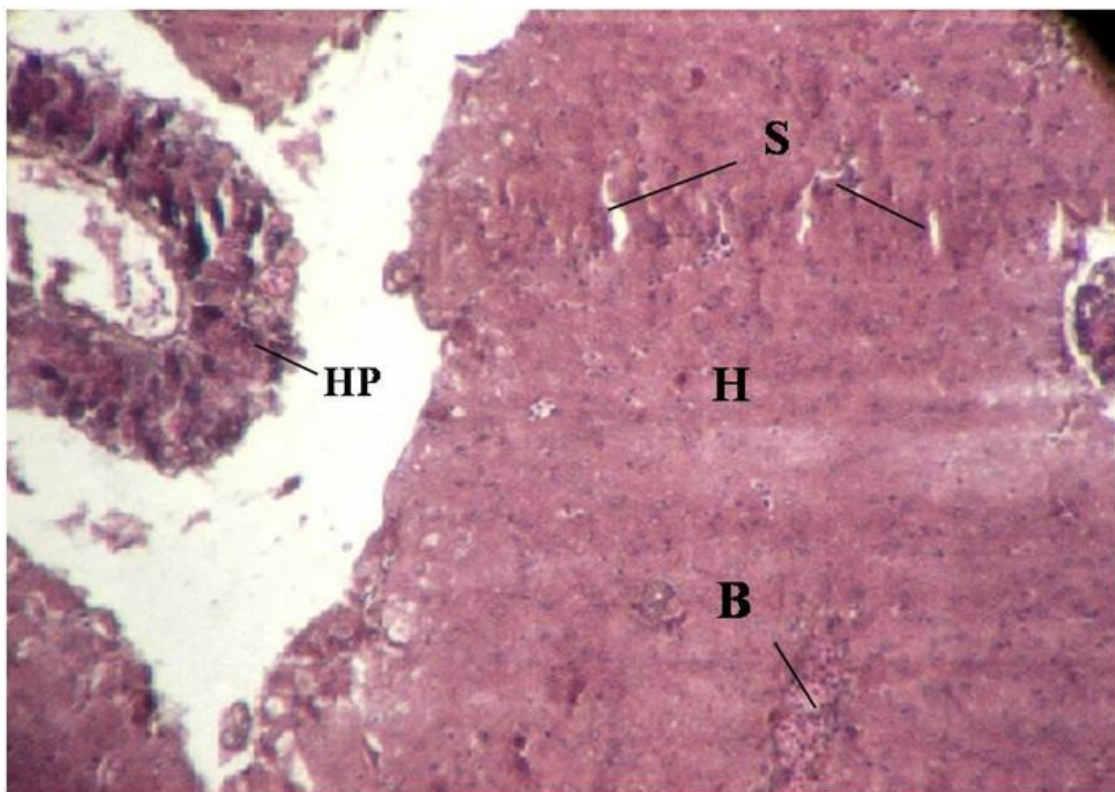


Figure 1. Microphotograph of liver of *L. rohita* from fishfarm showing hepatocytes (H), hepatopancreas (HP), sinusoids (S) and blood cells (B). HandE X400

simultaneously along with water samples. They were then anaesthetized using MS222 at the site itself so as to retain the properties of enzymes. After dissecting, the fish tissues such as liver, kidney and brain were carefully removed and transferred to a suitable media for biochemical estimations and for recording the activities of acid and alkaline phosphatase enzymes. In our initial studies for histopathological experiment, test fish sampled were from Hebbal lake (sewage polluted) and Chowkalli lake (industrial effluents polluted).

For biochemical estimations, the proteins were estimated by Lowry *et al.*, (1951) method using bovine serum albumen (BSA) as a standard, glycogen content was estimated by Anthrone reagent (Seifer *et al.*, 1949) using standard glucose solutions and cholesterol was estimated by Zlatkiis *et al.* (1953) method using standard cholesterol solution in FeCl_3 - acetic acid reagent. Acid and alkaline phosphatase activity (ACP and ALP) was determined by following the method of Bergmeyer *et al.* (1974) wherein Sodium p-nitrophenylphosphate was used as substrate. Tissue homogenate (2% w/v) were prepared in ice cold 0.9% saline and centrifuged at $5000 \times g$ at 0°C for 15 minutes. Buffer solution (0.5M citrate buffer, 0.0055 M p-nitrophenylphosphate, pH 4.8) was added to 10mL of homogenate. The reaction mixture was incubated for 30 min at 37°C . 4mL of 0.1M NaOH

was then added to stop the enzymatic reaction. The absorbance was measured at 420nm. The activity of acid phosphatase was expressed as units per mg protein). Alkaline phosphatase activity was also determined with the similar method as of acid phosphatase using Glycine buffer at a pH of 8.8.

Each assay was replicated six times and the values are expressed as (mean \pm SE) in tables 2 and 3. The results were then statistically analyzed with the help of ANOVA and students "t" test was applied to find out significance at 5%, 1% and 0.1% levels.

For histopathological examination small pieces of the tissues (liver and kidney) were rinsed in physiological saline and fixed in formal-saline (10% neutral buffered formalin) for 24 hours. They were dehydrated in an absolute alcohol series of ascending concentrations, and processed to form paraffin blocks (60°C – 62°C). The tissue were sectioned at $5\mu\text{m}$, stained with haematoxylin-eosin (HE) and examined under Zeiss compound binocular microscope fitted with a photomicrography attachment.

RESULTS

During the present studies the physico-chemical charact-



Figure 2. Microphotograph of liver hepatocytes of *L. rohita* from Hebbal lake showing scanty (S) granulation of hepatocytes and mildly granulated hepatopancreatic cells (GC). HandE X400.

ics of control Hebbal fish farm, Vengai lake (A) and Yellamallappa chetty lake (B) were analyzed and were compared with each other as well as with the standard are presented in Table 1. The parameters of water showed the level of pollutant load in the two water bodies (lake A and B), which in turn affected the growth and metabolic profiles of fishes present in the lake. Histopathological examination revealed structural changes in the liver and kidney tissue of the fish collected from Hebbal and Chowkalli lake, which was a pilot study for the present investigation. Significant differences in the degree of such changes were recorded according to the degree of pollution in the site. Histological structure of liver of the test fish revealed a homogeneous hepatic parenchyma and polygonal-shaped hepatic cells. The hepatocytes arranged as cords, usually are two cells thick. Pancreas extends into the liver forming a hepatopancreas in *Labeo* species. It is a pronounced feature in liver and is clearly visible as darkly stained tissue around the hepatic portal veins. It can be differentiated from hepatic tissue by its acinar arrangement. Thin septa of connective tissue separate the hepatocytes from the exocrine pancreatic cells. The photomicrograph of normal liver in the fish sampled from fish farm (control) revealed granulated hepatocytes with distinct centrally located nuclei. Hepatopancreatic cells showed granulation with zymogen granules (Figure 1). But those from Hebbal lake showed vacuolization in hepatocytes. Few hepatocytes showed scanty granulation with pyknotic nuclei. Mildly

granulated hepatopancreatic cells were observed (Figure 2). The hepatopancreatic cells showed cytoplasmic vacuolation and degeneration of variable magnitude, clumping of nuclear materials and destruction of nuclear membrane leading to complete lysis of hepatocytes, was observed in liver tissue of fish sampled from Chowkalli lake. Haemorrhage of blood vessels and hepatic lesions was also noticed which may be the result of exposure to xenobiotic substances in the polluted water (Figure 3). The trunk kidney of fish from Hebbal fish farm showed a number of well vascularised glomerulus and proximal and distal tubules. The haematopoietic tissue occupied inter-tubular space in the trunk kidney. (Figure 4). Severe histopathological changes such as vacuolation of tubular epithelial cells and disruption in its alignment were noticed in the kidney tissue of the fish from Chowkalli lake. Glomeruli showed degenerating condition and some had lost their structural organization. Haematopoietic tissue showed large number of degranulated parenchymatous cells and were in necrotic condition (Figure 5).

Biochemical studies are good parameters, which help to study the effect of aquatic pollution on biochemical composition of vital tissues of fish. In the present investigation there was a marked decrease in the protein and glycogen content and an increase in the cholesterol content in all the three tissues of the fish collected from lake B. The variations in the level of protein, glycogen and cholesterol and the activities of acid phosphatase

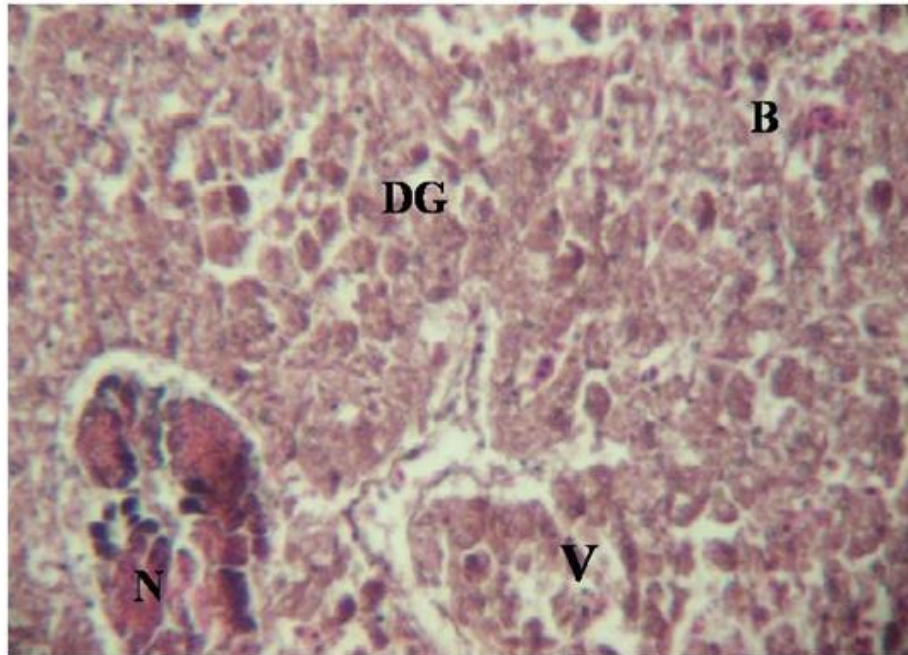


Figure 3. Microphotograph of liver of *L.rohita* from Chowkalli lake showing cytoplasmic degranulation and degeneration (DG), vacuolation (V), nuclear disintegration, haemorrhages of blood vessels (B) and clumping of hepatopancreatic nuclear material (N). HandE X400

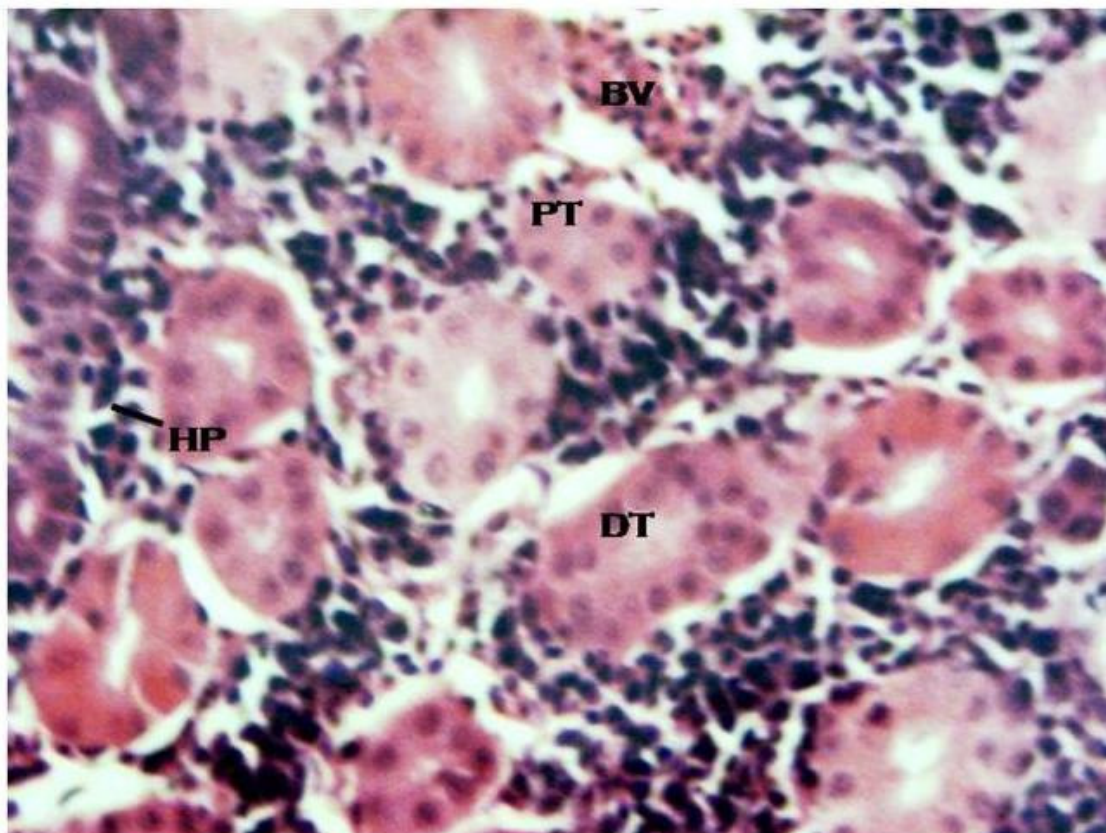


Figure 4. Microphotograph of kidney of *L.rohita* from fishfarm showing proximal tubules(PT), distal tubules (DT), haematopoietic tissue (HT) and blood vessel (BV). HandE X400

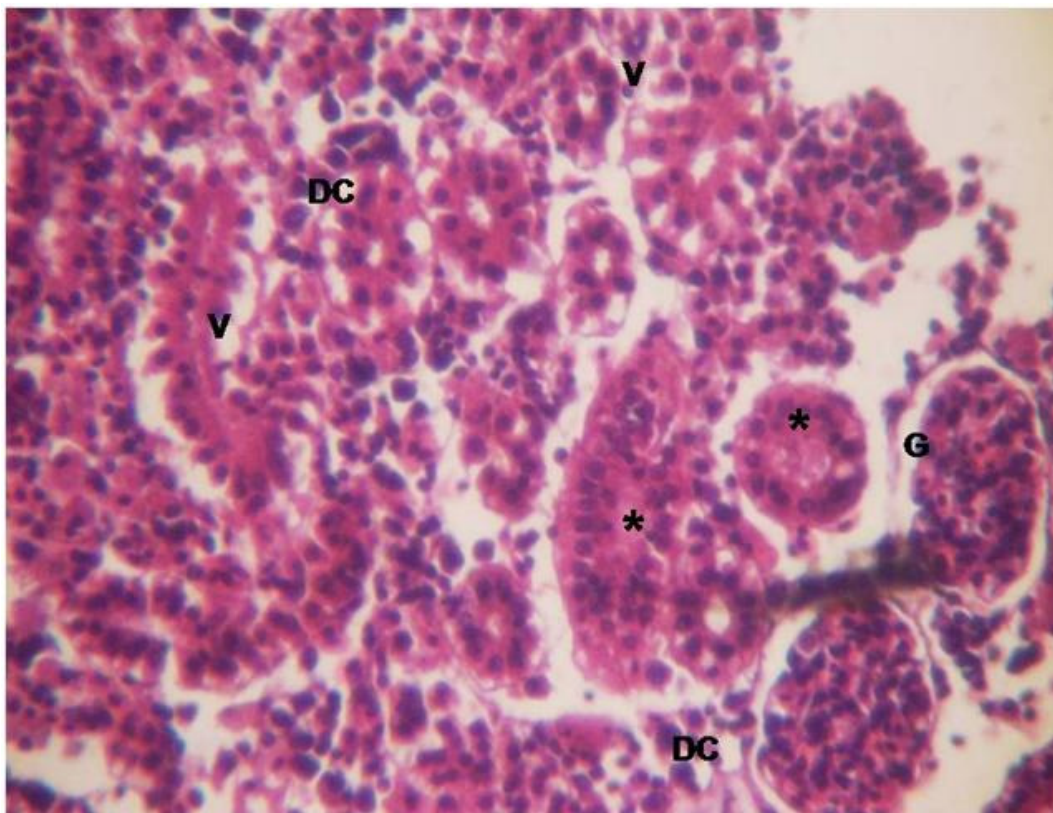


Figure 5. Microphotograph of kidney of *L. rohita* from Chowkalli lake showing vacuolated (V) and degenerated tubular epithelial cells with disruption in its alignment, tubule lumen with extensive infiltration of cells (*), glomerulus (G) with increase in glomerular space and large number of degranulated haematopoietic cells (DC) HandE X400

and alkaline phosphatase in liver, kidney and brain of freshwater fish *L. rohita* sampled from the fish farm (control), lake (A) and lake (B) is presented in Table 2 and 3

In the present study the protein content showed high level in liver (41.66 ± 1.38) when compared to those of kidney (26.50 ± 0.61) and brain (11.6 ± 1.92) in the control fish. A significant reduction of 27.2%, 32.72% and 23.27% respectively of protein content was recorded in the tissues of fish collected from lake (B) when compared to those of control. Maximum reduction in percentage of protein content was recorded in kidney (32.72%), which was followed, by liver (27.2%) and brain (23.27%). But protein content in fish sampled from lake A (39.00 ± 2.11 to 10.17 ± 1.22) did not show much variation when compared to those of control.

Glycogen content in the control fish was recorded highest in liver (4.26 ± 0.35), followed by kidney (2.58 ± 0.15) and brain (1.81 ± 0.22). Fish sampled from lake B showed significantly less glycogen content when compared to those of control ones in the order as liver > kidney > brain (2.42 ± 0.12 , 1.93 ± 0.19 and 1.32 ± 0.21 , respectively). An insignificant reduction of glycogen was recorded in liver (4.93%) followed by brain (7.18%)

and then kidney (10.4%) from the fish of lake A.

Cholesterol content showed a marked increase in all the tissues in the fish from lake B. The control fish collected from the farm showed highest cholesterol content in liver (2.6 ± 0.39) when compared to those of kidney (1.9 ± 0.24) and brain (1.3 ± 0.28). The fish sampled from lake B showed a remarkable increase of percentage in kidney cholesterol content (68.4%), followed by liver (57.6%) and brain (30.76%), whereas fishes from lake A showed a mild increase in kidney cholesterol content 5.2%, in liver 7.6% and brain 7.7% when compared to those of control ones.

The acid phosphatase activity (ACP) in the brain showed a minimum value of 3.58 ± 0.18 and liver showed the maximum value of 8.46 ± 0.26 in the control fish sampled from fish farm. Decrease in the ACP level was noticed in the tissues as brain < kidney < liver in the fish sampled from lake B (2.78 ± 0.12 , 4.55 ± 0.19 and 7.26 ± 0.22 , respectively). The ACP percentage showed maximum reduction in brain (22.34%) followed by kidney (22.22%) and liver (14.18%) of the fish from lake B compared to control ones. The alkaline phosphatase (ALP) content in the liver of fish sampled from fish farm showed a maximum content (16.61 ± 0.30) than those of

kidney (12.96 ± 0.11) and brain (9.62 ± 1.0). Fish sampled from lake B showed comparatively less alkaline phosphatase content (12.9 ± 0.31) when compared to those of control fish and the trend of reduction was recorded in the order as liver > kidney > brain. The reduction in the percentage of enzyme activity was highest in liver followed by brain and kidney (22.34%, 19.33%, and 19.13%). The fish sampled from lake A, did not show significant variation in the protein, glycogen, cholesterol, ACP and ALP parameters compared to the fish obtained from the fish farm and lake B.

DISCUSSION

The analyses of physico-chemical parameters of water are essential to evaluate water quality, as they provide important data about the variations caused by the different seasons and due to discharges into the water body from adjacent industries and residential areas. The liver parenchyma of the fish reared in Hebbal lake showed scanty granulation of hepatocytes and hepatopancreatic cells. Similar observations were reported in the siluriform *Corydoras paleatus* contaminated by organophosphate pesticides by Fanta *et al.*, (2003). These alterations are more severe when associated with the exposure of the fishes to contamination by metals. An increase in destruction of nuclear membrane and cytoplasmic degeneration leading to complete lysis of hepatocytes, was observed in the liver of *L. rohita* from Chowkalli lake. Melanomacrophages, degenerative and necrotic hepatocytes was observed in *Pleuronectes americanus*, to contamination with PAHs (polycyclic aromatic hydrocarbons) and pesticides in urban areas on the USA coast (Chang *et al.*, 1998). In fishes from Chowkalli lake which was industrially polluted showed the degenerative process leading to tissue necrosis (Takashima and Hibya, 1995). The high incidence of histological alterations in the kidney of *L. rohita* is an evidence of the poor environmental quality of this lake, while interrenal cells hypertrophy indicates that these fish are chronically exposed to stressors in their environment (Bela *et al.*, 2007). Although the specific causative factors for the observed alterations are unknown this study demonstrates the application of kidney histopathology as a general quality indicator of the aquatic environment (Silva and Martinez, 2007) and hepato-renal syndrome' condition in the stressed fishes (Narain and Srivastava, 2006).

The biochemical studies on the tissues as liver, kidney and brain of the test fish exposed to various pollutants in sewage- and in industrially polluted lake (Vengaiiah - A and Yellamallappa chetty - B) revealed a significant decrease in the levels of protein, glycogen, ACP and ALP and an increase in the levels of cholesterol in lake B when compared with those of control fish collected from

fish farm (Control) and those of lake A. Hymavathi and Rao, (2001) reported a decrease in the total proteins, carbohydrates and lipids in the fish, *Channa orientalis* collected from the habitat polluted by slaughterhouse wastes in comparison to an unpolluted habitat of Mudasarlova stream of Visakhapatnam. This is in agreement with the present investigations on *L. rohita*. The significant decrease in protein and glycogen content in tissues of fish from lake B may be an adaptive response to resist persistent stress of the pollutants. The results are in agreement with the depletion of protein content which is a physiological strategy adopted by the animal to adjust itself to the changed metabolic systems due to the exposure of fish to various pollutants. This leads to degradation processes like proteolysis and utilization of degraded products for increased metabolism (Jha and Jha, 1995a). Glycogen is the primary and immediate source of energy for all animals. The decrease in glycogen content in tissues of fish from lake B indicates the exposure of pollutants might have stimulated the mobilization of glycogen in the liver, resulting in its usage to overcome the stress by the animal. This could have happened by rapid glycogenolysis and inhibition of glycogenesis through activation of glycogen phosphorylase and depression of transferase (Kabeer *et al.*, 1984). It is suggested that liver reacts with the toxic substances by glycogen depletion. Since glycogen reserves in the liver tissue of fish under stress are used as an emergency energy supply, changes in the glycogen levels in these tissues could indicate the health status of fish population. The protein undergoes hydrolysis and oxidation through TCA cycle to meet the increased demand for energy caused by the stress (Somnath, 1991). The quantity of protein is dependent on the rate of protein synthesis or on the rate of its degradation. The inhibition of protein level may be due to the decrease in the alkaline phosphatase activity as it plays an important role in protein synthesis (Pilo *et al.*, 1972) and other secretory activities (Ram *et al.*, 2003). Due to pollutants, liver experiences metabolic stress and in such situations, the excretory mechanism is disturbed leading to an increase in cholesterol content as observed in the tissues of test fish from lake A and B. Goks*yr, *et al.*, (1994) reported increased lipid contents and accumulation due to the effect of aromatic and chlorinated hydrocarbons in juvenile Atlantic cod (*Gadus morhua*) caged in a polluted fjord due to the increased bioconcentration of lipophilic toxicants. Lipid deposits may exert protective effects by removing and inactivating organic chemicals from the metabolism or actively sequestering them, thus improving toxicity tolerance and resistance (Kruzynski, 1979). Krishna *et al.*, (1994) reported high levels of phospholipids and cholesterol content in the tissues of *Tilapia mossambica* subjected to acclimation in sub lethal acidic water (pH 4.0). These results can be correlated to the higher energy demands and impairments in cell membrane organization.

Enzymes are fragile substances with a tendency to undergo denaturation and inactivation under unsuitable conditions. Acid and alkaline phosphatases are general and hydrolytic enzymes present in all the tissues. They are concerned with the process of transphosphorylation and are associated with the transport of metabolites, with metabolism of phospholipids, phosphoproteins, nucleotides and carbohydrate, and with synthesis of proteins (Srivastava *et al.*, 1995). In the present investigation a marked decrease in the activity of both the enzymes, particularly alkaline phosphatase was noticed in all the three tissues of the fish sampled from lake B when compared to those of fish farm. Acid phosphatase showed a significant decrease in liver tissue when compared to those of kidney and brain in the fish sampled from lake B. The decreased acid phosphatase activity in the liver was due to increased glycogenolysis or due to changes in the mitochondrial membrane function. The accumulation of toxicants beyond a tolerable level in the liver might cause such enzymatic changes. This is in conformity with the report by Karuppasamy and Parthasarathi, (1998) in liver, intestine and muscle tissues of *C. punctatus* when exposed to fenvalerate. The decrease in acid phosphatase in liver suggested the uncoupling of phosphorylation by toxicity. Similar decreasing trends were observed in *Ophiocephalus punctatus* exposed to copper (Srivastava and Pandey, 1982). Alkaline phosphatase splits various phosphate esters at an alkaline pH and mediates membrane transport. In the present study a decrease in alkaline phosphatase was noticed in all the tissues of the fish from lake B suggesting inactivation of phosphorylase enzymes, thus promoting glycogen synthesis. Therefore, inhibition in alkaline phosphatase activity may cause alterations in glycogen content. Decrease in the activity of this enzyme may result in altered transport and an inhibitory effect on the cell growth and proliferation (Goldfischer *et al.*, 1964). Shaikila *et al.*, (1993) opined that inhibition of alkaline phosphatase activity could also be due to severe acidosis or interaction of toxicant with co-factors and regulators. This may in turn be adaptive for the fish to meet the energy demand by the anaerobic breakdown of glycogen. Similar findings were reported in liver of *Oreochromis niloticus* when exposed to methyl parathion (Sarabadikary and Sur, 1992), in *Brachydanioreio* exposed to malathion (Kumar and Ansari, 1986), in *Nemachelius denisonii* exposed to phosphamidon (Rashatwar and Ilyas, 1985), and in *Cyprinus carpio* exposed to vegetable oil factory effluent (Ramesh *et al.*, 1994). Few scientist had a contradictory results such as an increase in the activity of both the enzymes reported by Sastry and Malik (1981) in *C.punctatus* exposed to diazinon, an increase in acid phosphatase and a decrease in alkaline phosphatase activity observed by Shrivastava and Shrivastava (1998) in *Mus musculus* treated with carbyl and also by Johal *et al.*, (2002) in *Heteropneustes fossilis* exposed to

fenvalerate. An increase in alkaline phosphatase and a decrease in acid phosphatase activity were also noticed by Ruparelia *et al.*, (1992) in *Sarotherodon mossambica* exposed to cadmium.

CONCLUSION

Alterations were observed in histology of liver and kidney tissue of *L.rohita* reared in Hebbal and Chowkalli lake where they were exposed to contamination by domestic sewage and trace metals from industrial effluents. An increase in destruction of nuclear membrane and cytoplasmic degeneration leading to complete lysis of hepatocytes, was observed in the liver of *L.rohita* from Chowkalli lake. The changes in the biochemical profiles of the test fish suggest a hypoxic condition. The study also showed that the disruption in metabolic processes is very high in the experimental lakes, especially lake B due to the industrial pollutant load. The decreased activities of ACP and ALP enzymes indicate disturbance in the structure and integrity of cell organelles, like endoplasmic reticulum and membrane transport system. (Karatat and Kalay, 2002; Roy, 2002).

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