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Tauseef Ullah
Department of Zoology,
Government College University
Faisalabad, Pakistan

Tayyaba Sultana
Department of Zoology,
Government College University
Faisalabad, Pakistan

Luqman Khan
(A). Department of Zoology,
Government College University
Faisalabad, Pakistan
(B) Division of Neurogenetics,
Graduate School of Life Sciences,
Tohoku University, Japan

Khurram Feroz
Department of Zoology,
Government College University
Faisalabad, Pakistan

Qazi Adnan Ahmad
Department of Zoology,
Government College University
Faisalabad, Pakistan

Sarfraz Hussain
Department of Zoology,
Government College University
Faisalabad, Pakistan

Correspondence
Luqman Khan
(A). Department of Zoology,
Government College University
Faisalabad, Pakistan
(B) Division of Neurogenetics,
Graduate School of Life Sciences,
Tohoku University, Japan

Histopathological alteration in Gill, Kidney and Liver of *Cirrhinus mrigala*, *Catla catla*, *Hypophthalmichthys molitrix* and *Labeo rohita* due to sub-lethal exposure of textile industries effluents in Faisalabad, Pakistan

Tauseef Ullah, Tayyaba Sultana, Luqman Khan, Khurram Feroz, Qazi Adnan Ahmad and Sarfraz Hussain

Abstract

The objective of present study was to determine the effect of textile industries effluents on water and aquatic organisms particularly fish. The experiment was performed in glass aquaria containing fingerlings of approximately ~25-28 gm. Histopathological changes in the gill, kidney and liver of four species were observed. Severe degeneration of the secondary gill filaments, degenerative changes in the primary gill filament were observed. Short fusion of secondary lamellae, less changes in the integrity of lamellae, less proliferation of mucus cell and severe hemorrhage showed in the gill of *C. catla*. Histopathological changes were glomerular shrinkage, increased spaces between glomerulus and Bowman's capsule, increased tubular lumen in the kidney of *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla*. Histopathological changes in the liver were focal area of necrosis, intravascular hemorrhage, vacuolar degeneration, severe hemorrhage, hepatocytes degeneration, necrosis, and hemolysis, erythrocyte infiltration in blood sinusoid, eccentric nuclei, cytoplasmic vaculation and dilation of the vein.

Keywords: *Cirrhinus mrigala*, *Catla catla*, *Labeo rohita*, *Hypophthalmichthys molitrix*, Histopathology

1. Introduction

Fish are comparatively sensitive to fluctuations in their surrounding environment containing pollution. Pollution of the aquatic system is known as a possible threat to all living organisms. Fish health may reveal the position of a particular aquatic ecosystem^[1]. Water quality is being reliably affected by man actions towards development. Fish are considered as a significant source of high superiority animal protein as they have great quantity of essential amino acids. In the beginning harmful effect of pollution may be obvious upon cellular or tissue level before important variations can be known in fish behavior or external appearance. Problem of ecological contamination, industrial discharges restraining toxic substances, including heavy metals extremely destructive to aquatic ecosystem^[2]. Discharge of heavy metals into the water environment can alter both aquatic species diversity and ecosystem due to their poisonous and accumulate in different tissues in significant amount^[3].

Heavy metals are non-biodegradable when once disposed into water ecosystem can either imbed in sediments elements or collected aquatic organisms. Some quantity of these heavy metals settled in the bottom of the aquatic reservoir and some amount accumulated in freshwater organisms. Wastewater of different sources has been verified that it is dependable for the degradation of aquatic life in different water bodies. Water factors change the physicochemical qualities for instance, nitrates, sulphide, also including BOD, COD, TSS and existence of metals, consequently harmful variations occur in different tissue^[4].

Histopathological investigations have been applied to define the health position of fish from contaminated sites in contrast to those from uncontaminated places. Histological indicators have been suggested to measure the influence of environmental stress in freshwater fish. Histopathological examination be a very susceptible parameter and critical in decisive cellular alteration that may happen in target organs, such as the gills, liver and kidney^[5]. Histopathological biomarkers are significant indicators of the general physical condition of

fish and reflect the effects of different range contaminants [6]. Histopathological biomarkers are directly linked to other biomarkers of stress for many pollutants have to endure and catalyze metabolic actions in order to provoke cellular damages in the affected organism [7]. Sub-lethal exposure to the aquatic environment, contaminants may result in variations histological structure of cells and pathological variations, which importantly change the function of tissues and organs. Histopathological changes are good indicators by which revealed the effect environmental stress and result in physiological and biochemical changes in the body tissues of organisms [8].

In Pakistan, most of rivers are not sufficiently monitored in order to conserve healthy and harmless supply of fish to customers. The present work was an effort to assess the Textile Industries toxic impact of heavy metals on Gill and Liver of *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla* and sub-lethal concentration of textile industries effluent and its effect on different organs of the fish, including gill, liver and kidney. This work was aimed to study the effects of textile industrial waste water on water chemistry through water quality parameters and to check and compare the level of sensitivities against textile mill effluents in different fish species.

2. Material and Methods

2.1 Fish Selection

Fingerlings of *C. mrigala*, *L. rohita*, *C. catla* and *H. molitrix* were collected from Fish Hatchery, Satyana Road, Faisalabad. These fishes are selected because of their different niche i.e. (surface, column and bottom feeder) of water based on differences in their food niche like herbivores (*L. rohita* and *C. mrigala*), omnivore (*H. molitrix*) and carnivore (*C. catla*).

2.2 Textile Industries effluents water samples

Textile industries effluents were collected from five wastewater drains of Faisalabad- drains of Mustafa Abad, Bawa Chak, Abdullahpur, Satyana road and Sumandhri road) containing different textile industries effluents of processing, dyeing, printing and other textile corporation mills. Representative water samples from each site were collected in 1.5 L capacity of polypropylene bottles with polyethylene caps from the five selected sites. The bottles were thoroughly washed with water before taking samples and transported to the Research Laboratory, Department of Zoology, Faisalabad for physicochemical analysis and treatment. The Fish fingerling's specimen (approx.8-10 g) were procured alive from Government Fish seed hatchery, Satyana Road, Faisalabad and transported in fresh water containing plastic bags filled with oxygen to Fisheries Research Laboratory, Department of Zoology, Government College University, Faisalabad. The fingerlings of fish were bathed in dilute solution of potassium permanganate to remove any disease or infection and kept under laboratory conditions in glass aquarium for acclimatization. The experimental aquaria were aerated continuously with an air pump through capillary system. After acclimatization fish was divided into two groups one as experimental and other as control group. The fish were fed commercial diet @ 3-4% wet body weights. The wet weight, fork and total lengths of all fish in both groups were measured prior to the start of experiment. Each group was consisted of three replications to see the statistically significant differences. Growth parameters were recorded on monthly basis.

2.3 Water Sampling

Water sampling were done randomly by depth; center and sides (right and left banks) as water calmly flows through the drain due to the fact that drain is excavated at considerable depth allowing organic matter to sediment. Representative water samples from each site were collected in 1.5 L capacity of polypropylene bottles with polyethylene caps from the five selected sites. The bottles were thoroughly washed with water before taking samples.

2.4 Physico-chemical Parameters

Water quality parameters were observed for control and treated fingerling's tanks like, pH, temperature, dissolved oxygen (DO) on daily basis, while rest of WQPs of biological oxygen demand (BOD), Chemical oxygen demand (COD), Total dissolved solids (TDS-mg/L), Total suspended solids (TSS-mg/L), Salinity (mg/L) electrical conductivity, salinity, were monitored fortnightly with the help of HI 9828 Multi-Parameter HANNA meter. Samples were analyzed according to the standard procedures [10].

2.5 Toxicity Test (LC50)

The percentage concentration of sample water was prepared on the basis of volume to volume (v/v) ratio. After determining the LC50, three sub lethal dilutions of the textile effluents were tested for three months trial. LC 50 values for these fish species were determined separately according to their resistance and sensitivity. The concentration of textile waste water given separately to all four fish species tanks and killed 50 percent of the fingerlings for 96 hours, then the lowest sub-lethal concentration was selected in order to kept all these four species in the same aquariums. After determining the LC50, the fish survival sub-lethal concentrations of water from textile mill effluent were tested for three months trial introducing these fish species in the same glass aquaria as poly-culture. The exposure mediums were continuously be monitored to check the fish survival and to maintain the sub-lethal concentrations of drain sample in appropriate manner. The fish were fed commercial diet 3-4% of wet body weight. Fish mortality data were obtained against each concentration during 96 hours test duration. Each test concentration was tested in triplicate. The acute toxicity bioassay procedure, based on standard method was conducted to determine 96-hours LC 50 [9]. Growth parameters were recorded on monthly basis. During the exposure experiments, the water quality parameters viz. water temperature, pH, dissolved oxygen on routine basis, while rest of WQPs were monitored fortnightly. Detection of selected heavy metals was performed once in each dilution in every month.

2.6 Heavy Metals Analysis

Metals analysis was performed in the Research Lab, Zoology Department. Textile waste water collected in acidified water bottles with the capacity of 1500 ml brought to the laboratory for heavy metals examination and stored below -20 °C prior to analyses. Heavy metals analysis for each sample determined with the help of atomic absorption spectroscopy (AAS) [10].

2.7 Preparation of heavy metals standards

To prepared calibrated solution, stocked solution was used which is commercially available in the form of 1000 ppm. Apparatus carefully washed with de-ionized water for the preparation of standard solutions. This stock solution was used for making working solutions of 5 ppm, 10 ppm, 15 ppm, 20 ppm, 25 ppm and 30 ppm.

2.8 Atomic absorption spectrometry

Water samples examined for heavy metals concentration using Atomic Absorption Spectrometer. The solutions were then aspirated into an atomic absorption spectrophotometer (Hitachi polarized Zeeman Atomic Absorption Spectrophotometer AAS, 2000 series) by using an air acetylene flame [11]. The protocol of AOAC (1990) was used with little modification.

2.9 Histological analysis

Ten fish from each treatment group were anesthetized and gills, livers and kidneys were removed. Tissues were kept in pre-labeled, individual glass vials in which Bouin's fixative was added already. After 24 hours immersion fixation, tissues were transferred to coded glass vials containing 70% ethanol. Tissues dehydrated in concentrations of alcohol, and embedded in paraffin wax for microtome sectioning (5 μ m) [12]. Sections were mounted onto slides and after hydration in ethanol series of descending concentration, were stained with standard haematoxylin and eosin (H and S) stain. 10 sections of each tissue from each fish were examined by light microscope [13].

2.10 Histopathological Studies

For histological study of fingerlings muscles tissues (Kidney, Liver and Gill) were taken in the research laboratory. Muscles tissues were fixed in sera for 4 to 6 hours. Constitution of sera contained of absolute alcohol, formaldehyde and glacial acetic acid with quantity of 60, 30 and 10 ml respectively. Then dehydration of tissues was done with ethanol (70%) for 12 hours at 20 °C, ethanol (80%) for 2 hours at 20 °C, ethanol (90%) for 2 hours at 20 °C and finally tissues were dehydrated with ethanol (100%) for 2 hours at 20 °C. After the dehydration fixed tissues were shifted into cedar wood oil until became clear and translucent at 20 °C. Then embedding of tissues was with the help with benzol-1 for 15 minutes at 20 °C, benzol-2 for 15 minutes at room temperature, benzol-1 + paraplast-1 for 25 minutes at 50 °C, and Paraplast-1 for 15 hours. at 50 °C, Paraplast-2 for 15 hours at 50 °C and Paraplast-3 for 15 hours at 50 °C. Embedded tissues were placed into molten wax in a boat and 3-4 μ m thick finest sections sliced with the help of microtome for these blocks were trimmed from the paraffin wax with the help of knife or scalpel and fixed on wooden block for sectioning. The ribbons with tissue were enlarged and poured on cleaned albumenized glass slides on fisher slides warmed at 60 °C. Then these glass slides were kept in incubator for a night for completion of stretching and elimination of the remaining bubbles. Tissues were stained with the help of Haematoxylin and Eosin stain staining solution. Stained slides were fixed with Canada balsam and slides were incubated for 12 hours and were

covered with cover slip and extra Canada balsam was removed by xylene.

2.11 Light Microscopy

For histological examination prepared slides were examined under light microscope at magnifications power 10X, 20X, 30X and 40X. For Control and Treatment group all slides were studied and photographs were taken.

2.12 Statistical Analysis

Data collected were subjected to proper statistical analysis. Two-way complete randomized analysis of variance (ANOVA) was used and tested different variation of the water quality parameters with the 0.05 significance level [14]. Different factors used against all physical parameters, dissolved oxygen, pH, pH (mv), temperature °C, pressure (mbar), resistance ($M\Omega m$) electrical conductivity ($\mu S/cm$), actual electrical conductivity ($\mu S/qcma$), total dissolved solids (ppm), salinity (ppm) and redox potential (ORP). Correlation coefficient was calculated to test degree of relationship between the water quality parameter and Duncan Multiple Range tests to test difference among all possible pair of treatment means. Fingerling mortality rate was obtained by calculating number of death of fingerlings in each trail. The mortality percentage was also calculated abbot Formula (Abbot 1935). This concerned mortality percentage data was analyzed by using statistical approach, a software Minitab. A probit model was used to analyze the mortality of all different four fingerlings to each wastewater concentration and LC50, Slope, Standard error, Chui square, and probability were obtained.

3. Results

The comparative study of sub-lethal concentration exposure of textile industries effluents to *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla* was studied. Water physicochemical parameters, heavy metals analysis, histology and histopathology of gill, kidney and liver of control and treated fish species were determined. Water physico-chemical parameters were checked on daily basis for control and treatment groups. DO, BOD level and temperature was maintained during the exposure of sub-lethal concentration of textile effluents, dissolved oxygen was with mean of 5.216 (mg/l), pH mean was 7.359, pH (mv) mean was -108.2, temperature was with mean 29.460 °C determined. All other water quality parameters including pressure, resistance, electrical conductivity, TDS and salinity shown in tables (Table 1) for control and treatment group with their means, standard deviations, variance, minimum and maximum level.

Table 1: Physico-chemical Parameters of water

Water parameters	Mean	SD	Variance	Min.	Max.
Dissolved oxygen (mg/l)	5.2168	0.0576	0.0033	5.1200	5.3100
pH	7.359	0.2016	0.0406	7.1000	7.8000
pH (mv)	-108.2	52.9	2801.7	-126.9	128.4
Temperature (C ⁰)	28.639	0.503	0.253	27.890	29.460
Pressure (m Bar)	818.56	2.00	4.00	814.70	822.60
Resistance (m Ω cm)	0.001268	0.000048	0.000	0.001200	0.001300
Electrical conductivity ($\mu s cm^{-1}$)	769.95	16.77	281.09	732.00	791.00
TDS (ppm)	313.41	1.74	3.02	310.00	316.00
Salinity (ms)	3.1128	0.2730	0.0745	2.3620	3.4890
Salinity (per cm ²)	2.8715	0.0733	0.0054	2.7670	2.9860
Original Salinity	1.2677	0.0966	0.0093	1.1270	1.4640
BOD or ORP (Oxidative Redox Potential)	-205.22	28.63	819.63	-283.10	-118.70

3.1 Analysis of heavy metals

Random collection of textile wastewater from each selected sites was done and descriptive statistical analysis of wastewater analysis of heavy metals including Mn, Cr, Pb, Zn, Sn, Ni, Co and Cu with different concentration were observed. Analysis of heavy metals in the effluents samples recommended that there were distinct variations observed among all these heavy metals. In the textile effluents samples,

concentrations of Mn, Cr, Pb, Zn, Sn, Ni, Co and Cu were observed with its means and standard deviation, 22.317±4.33, 16.953±1.79, 19.172±2.80, 3.265±1.34, 4.917±1.70, 7.186±1.45, 3.029±2.25 and 5.163±3.20 mg/l were respectively observed (Table 2). Analysis revealed that there was heavy metals with high concentration values in the wastewater were Mn, Pb and Cr while Co, Zn, and Sn were determined in less concentration in the wastewater.

Table 2: Summary table of heavy metals.

Element	Mean (mg L ⁻¹) & SD	Max. (mg L ⁻¹)	Min. (mg L ⁻¹)
Mn	22.317±4.33	26.647	17.987
Cr	16.953±1.79	18.738	15.168
Pb	19.172±2.80	21.973	16.371
Zn	3.265±1.34	4.600	1.929
Sn	4.917±1.70	6.617	3.217
Ni	7.186±1.45	8.632	5.740
Co	3.029±2.25	5.274	0.784
Cu	5.163±3.20	8.364	1.962

3.2 Toxicity test for sub-lethal concentration (LC50)

The percentage concentration of sample water was prepared on volume to volume (v/v) ratio. LC50 for 96 hours were determined for *C. mrigala* (169.377 ml/L), *C. catla* (256.337 ml/L), *L. rohita* (194.902 ml/L) and *H. molitrix* (186.414 ml/L). Table 3 showed number of fish species (N), slope with

Standard deviation (Slope ± SE), chi-Square (X²), degree of freedom (df), LC50 or mean, Mean standard error, Mean of 95% Confidence Interval Lower Bound and Upper Bound Threshold (ML and MU) and significant level (P) for each species.

Table 3: Statistical analysis of LC50 for *C. mrigala*, *C. catla*, *L. rohita* and *H. molitrix*.

Fish Specie	Exposure duration	N	Slope ± SE	X ²	df	LC50	Mean SE	ML	MU	P
<i>Cirrhinus mrigala</i>	96	60	0.0826469±0.0055107	12.1511	12	169.377	0.829766	167.751	171.004	0.434
<i>Catla catla</i>	96	60	0.0599339±0.0033405	13.7577	22	256.337	0.967742	254.440	258.234	0.910
<i>Labeo rohita</i>	96	60	0.0538324±0.0031882	20.8594	15	194.902	1.07032	192.804	197.000	0.141
<i>Hypophthalmichthys molitrix</i>	96	60	0.0623696±0.0038324	21.7441	14	186.414	0.977826	184.497	188.330	0.084

3.3 Histology and Histopathology

Histological and Histopathological examination of selected control and treated fish species revealed that textile effluents consisted a large number of heavy metals has adversely profound effect on body organs and damaged their cellular organelles and tissues. Different changes and damages in the gill, liver and kidney were clearly marked separated as clinical symptoms at different stages of experiment. This histopathological examination for continuously three months was served as biomarkers and monitored textile wastewater toxicity with different concentration and its effect on different organs of the same and different fish species. Histopathological changes which linked to textile toxic effluents and accumulated in the fish liver examined and recommended that these toxic wastes created rigorous damage to the gills, kidney and liver cells.

3.4 Histology and Histopathology of Gill

Gill of control group, *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla* showed normal gills filaments of primary lamellae and secondary lamellae and proper arrangement of the gill filaments in double rows shown in figure 1-A, B, C, and D. Histopathological examination of gill due to sub-lethal

exposure of Textile Industries effluents, there were different variations and damages revealed, severe degeneration of the secondary gill filaments and degenerative changes in the primary gill filament in the gill of *C. mrigala*, *L. rohita* and *H. molitrix* (Figure 2-A, B and C) while short fusion of secondary lamellae, less changes in the integrity of lamellae, less Proliferation of mucus cell and severe hemorrhage showed in the gill of *C. catla* (Figure 2-D). There were great losses of gill lamellae, fusion of secondary lamellae, hemorrhage in primary and secondary lamellae, degenerative and necrotic changes in the epithelium of gill filament reported in the gill of *C. mrigala*, *L. rohita* and *H. molitrix* while curling of secondary gill lamellae and separation of gill filament from basement membrane showed in the gill of *L. rohita* and *C. mrigala* (Figure 2-B and D). Severe degenerative changes in the secondary lamellae were proliferation of mucus cell, atrophy of gill filament, loss of structural integrity of lamellae and congestion of blood vessels in gill filament in the gill of *C. mrigala*, *L. rohita* and *H. molitrix* were observed. Severe loss in histoarchitecture in the gill of *C. mrigala*, *L. rohita* and *H. molitrix* while less degenerative changes in the secondary lamellae examined in the gill of *C. catla*.

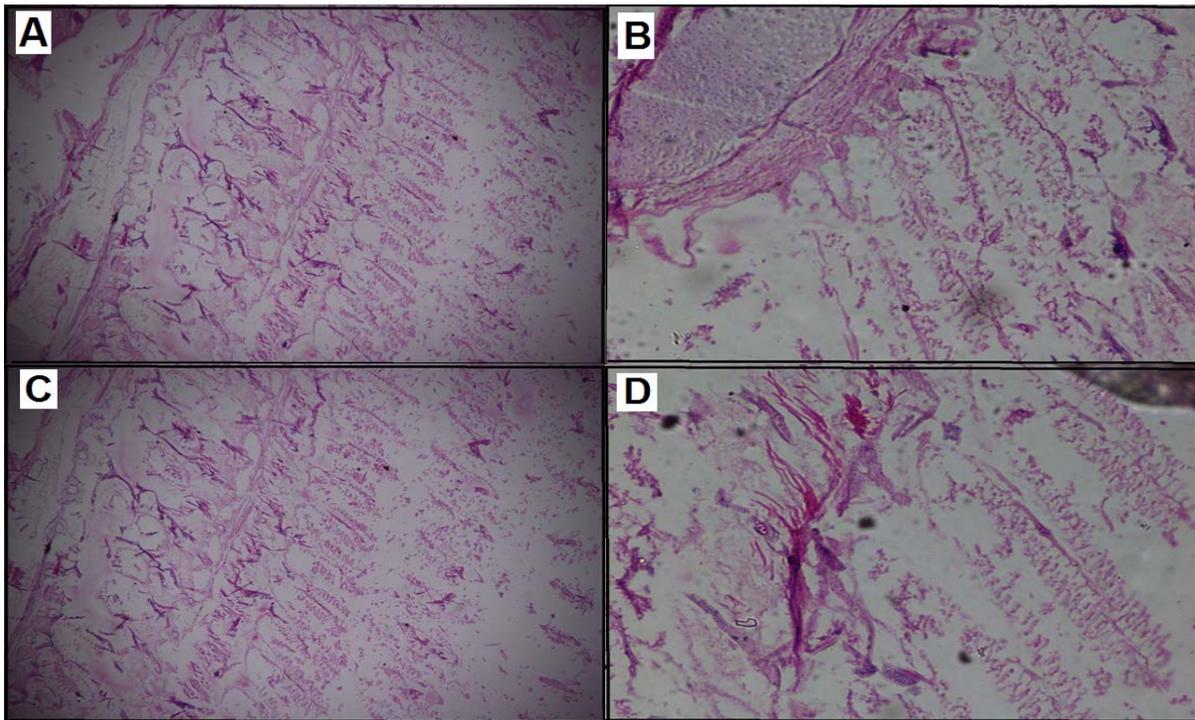


Fig 1: Gill of control group (A, B, C and D 20X)

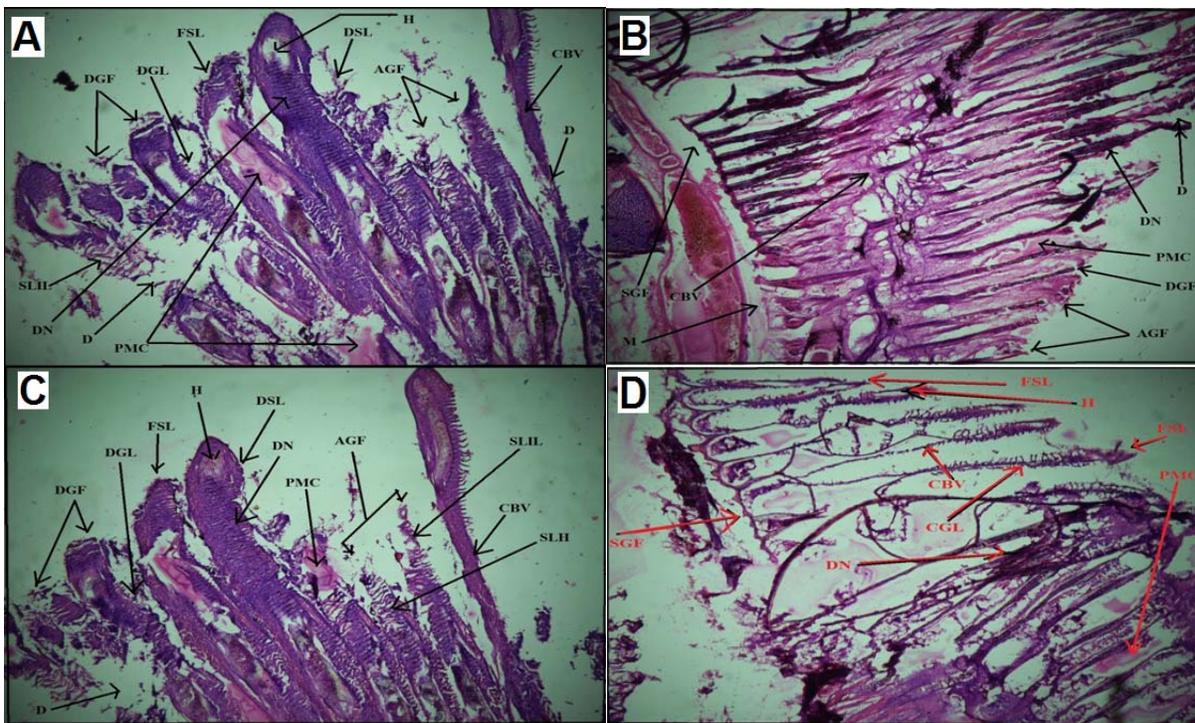


Fig 2: Gill of treatment group (A, B, C and D 40X), Degeneration of gill filament (D), Degenerative changes in gill filament (DGF), Degeneration of gill lamellae (DGL), Fusion of secondary lamellae (FSL), Hemorrhage in primary and secondary lamellae (H), Degenerative and necrotic changes in epithelium of gill filament (DN), Degenerative changes in secondary lamellae (DSL), Proliferation of mucus cell (PMC), Atrophy of gill filament (AGF), Loss of structure integrity of lamellae (SLIL), Congestion of blood vessels in gill filament (CBV), Severe loss in histoarchitecture (SLH), Curling of secondary gill lamellae (CGL) and Separation of gill filament from basement membrane (SGF).

3.6 Histology and Histopathology of Kidney

Kidney of control group of *Cirrhinus mrigala*, *Labeo rohita*, *Hypophthalmichthys molitrix* and *Catla catla* reported rich of renal corpuscles with well expand glomeruli and a system of renal tubules, showing Bowman’s capsule and capillaries in glomerulus. Kidney of control group showed well organized blood vessels and hematopoietic tissues (Figure 3-A, B, C,

and D). Due to sub-lethal exposure of textile industrial effluents different variations seen in the kidney of *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla*. Histological examination revealed that glomerular shrinkage occurred in the kidney of all treated group fish species, increase gap between glomerulus and Bowman’s capsule and increased tubular lumen in the kidney of *C. mrigala*, *L. rohita*, *H.*

molitrix and *C. catla* was reported (Figure 4-A, B, C and D). Pycnotic nuclei and hyaline degeneration of tubular epithelium also showed in the kidney of *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla*, less glomerulus shrinkage found in the kidney of *C. catla* (Figure 4-D), disorganized tubules, hydrophobic swelling, intact of tubules vaculation, and eccentric nuclei found in the kidney of *C. mrigala*, *L. rohita*

and *H. molitrix* (Figure 4-A, B, and C). Desquamation, collapsing of glomeruli and damage of blood vessels determined in the kidney of *C. mrigala*, *L. rohita* and *H. molitrix*. Diameter of lumen increased and nuclei of some epithelial cells became pycnotic in the kidney of *C. mrigala*, *L. rohita* and *H. molitrix*.

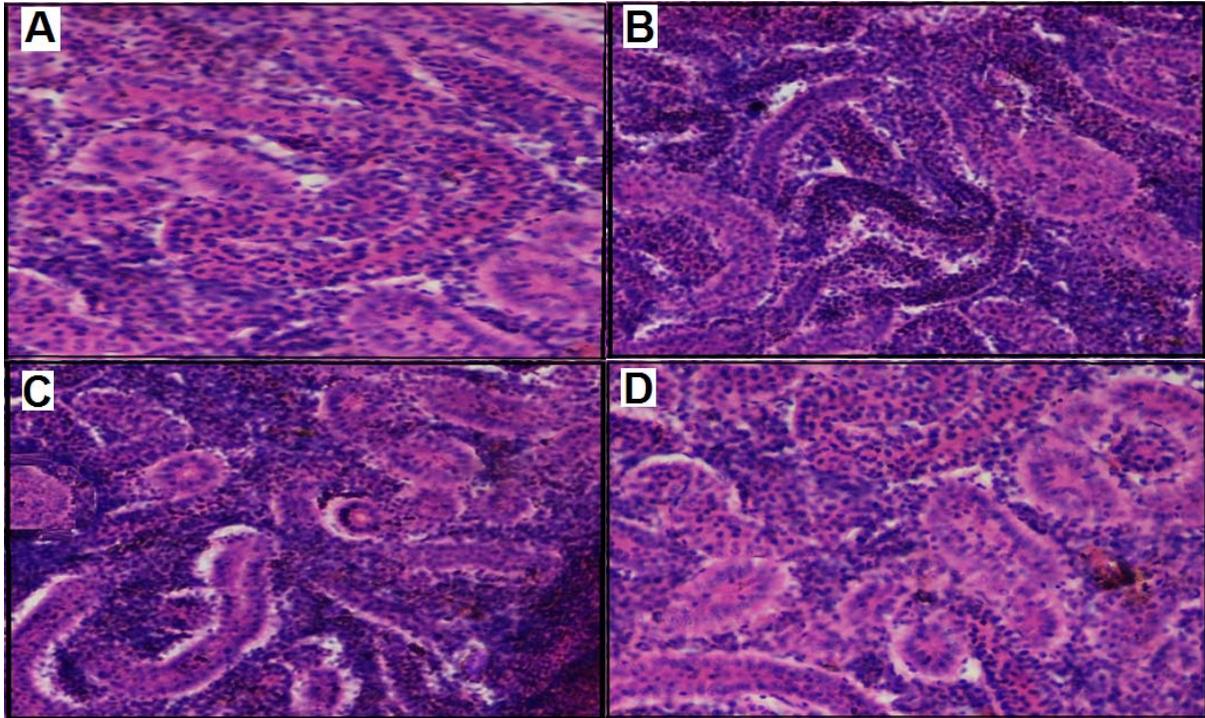


Fig 3: Kidney of control group (A, B, C and D 20X)

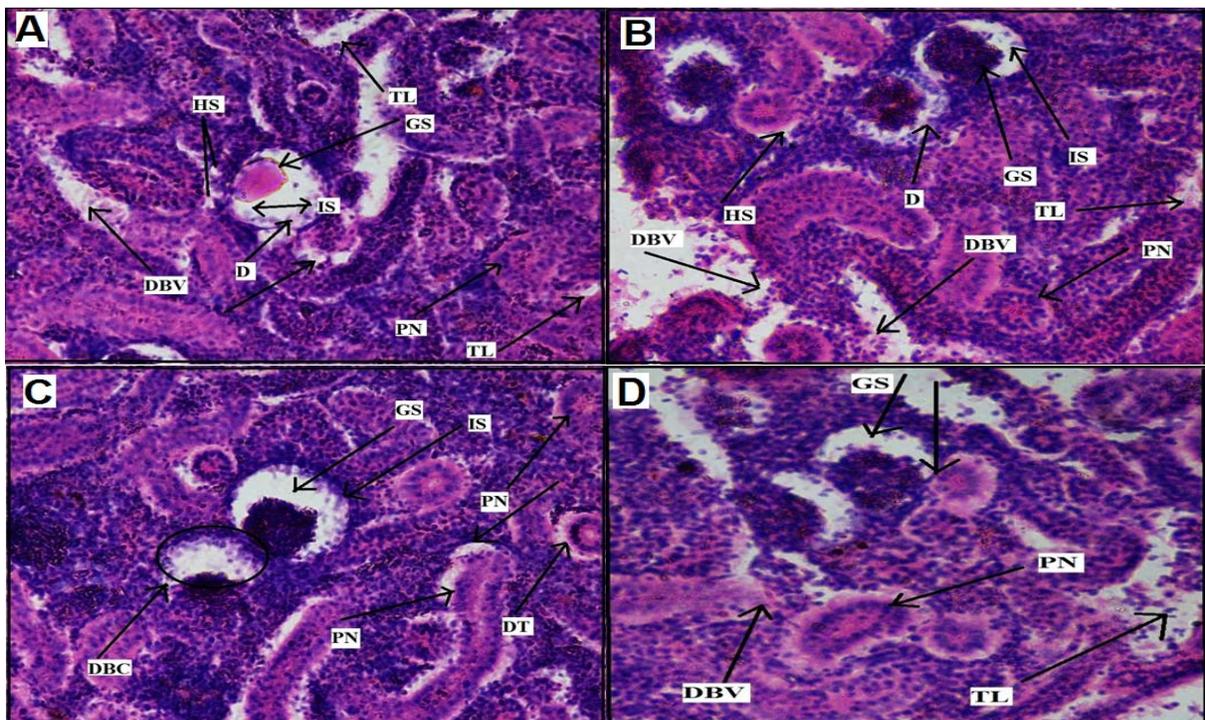


Fig 4: Kidney of treatment group (A, B, C and D 40X), Glomerular shrinkage (GS), Increase space between glomerulus and Bowman's capsule (IS), Increase tubular lumen (TL), Pycnotic nuclei (PN), disorganized tubules (DT), Hydropic swelling (HS), Desquamation (D) and Damage blood vessels (DBV).

3.7 Histology and Histopathology of Liver

Liver of control fish showed distinctive dense designed feature, distribution and morphology of cells and organ (Figure 5-A, B, C and D). Histopathological study of fish liver due to sub lethal concentration of textile effluents revealed severe damages of cellular structure, high scale necrosis and hemolysis due to damage of erythrocytes and inflammation of the hepatic cells, eccentric nuclei. Vacuole were emerged and commonly found in the liver of *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla* (Figure 6-A, B, C and D). Liver of *C. mrigala* showed severe and less severe hemorrhage, focal area of necrosis, intravascular hemorrhage, vacuolar degeneration, hepatocytes degeneration, necrosis, hemolysis, erythrocyte infiltration in blood sinusoid, eccentric nuclei, cytoplasmic vaculation and dilation of vein (Figure 6-

A). Histopathological alteration in the liver of *H. molitrix* showed vacuolar degeneration, dilation of vein, Eccentric nuclei, infiltration of erythrocytes, congestion of blood vessels, mild hemosiderin, congestion of sinusoid and separation of hepatocytes (Figure 6-C). Liver of *L. rohita* showed hemorrhage, infiltration of erythrocytes, focal area of necrosis, intravascular hemolysis, eccentric nuclei, vacuolar degeneration, congestion and dilation sinusoid and dilation of vein (Figure 6-B). Histopathological changes in the liver of *C. catla* showed less severe hemorrhage, short focal area of necrosis, less intravascular hemolysis, infiltration of erythrocytes, vascular degeneration, and rough shape of hepatocytes, hepatocytes hypertrophy and separation of hepatocytes (Figure 6-D).

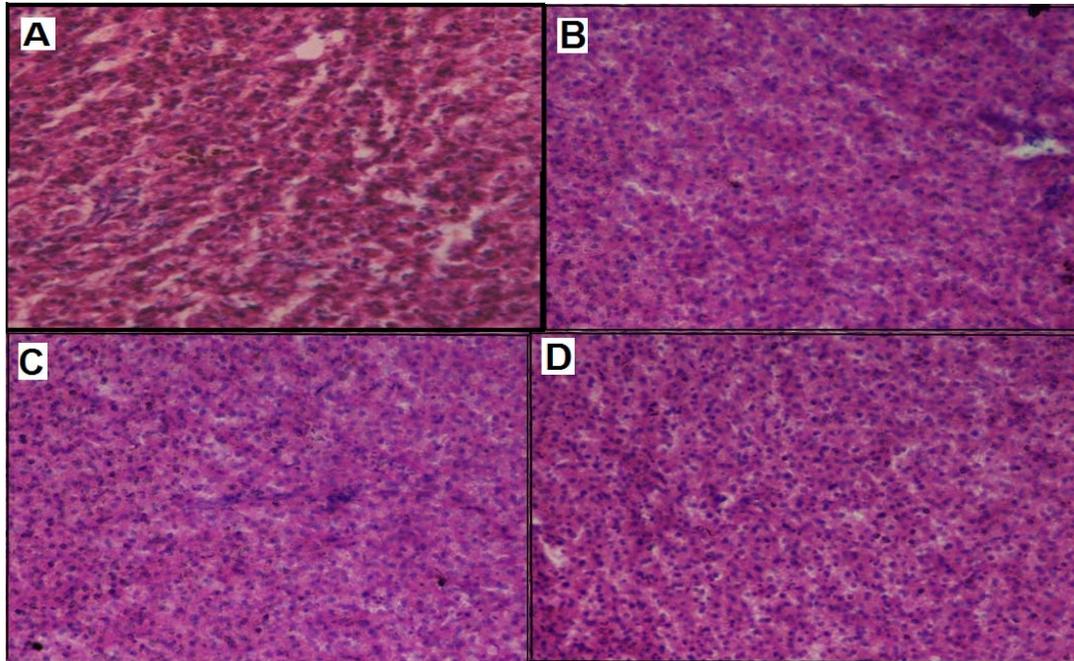


Fig 5: Liver of control group (A, B, C and D 20X)

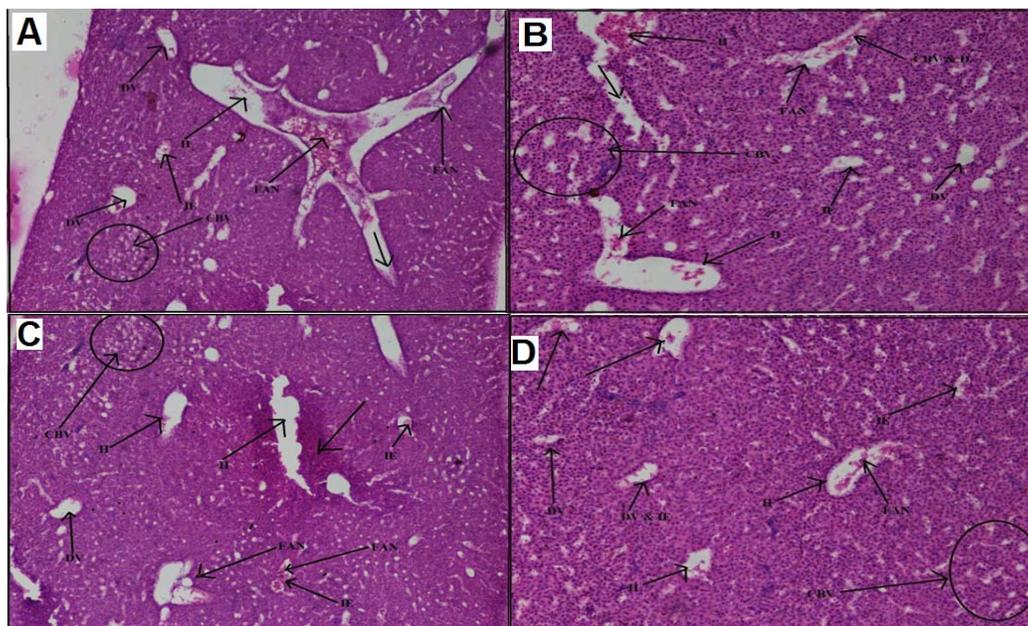


Fig 6: Liver of treatment group (A, B, C and D 40X), Focal area of necrosis (FAN), Dilation of vein (DV), Severe hemorrhage (H), Congestion of blood vessels (CBV) and Infiltration of erythrocytes in blood sinusoid.

4. Discussion

In this comparative study, responses of *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla* due to the exposure of sub-lethal concentration of textile industries effluents, heavy metals analysis, histology and histopathology were studied. In this study DO, pH, temperature, pressure, resistance, electrical conductivity, TDS, salinity, and ORP were observed of the treatment group with their average mean, 5.2168 mg/l, 7.359, 28.639 °C, 818.56 (m bar), 0.001268 (mΩcm), 769.95 (μs cm⁻¹), 313.41ppm, 3.1128 (ms) and -205.22 respectively. Textile industries effluents that had broad range of pH that different from 2 to 12 have great variation in the research work [15]. Alkalinity was also determined in excessive amount (8.10±4.33 mg/l) along total hardness of 330.00*60.00 mg/L. For dissolved oxygen different quantity of water samples were experimented, result in the absence of dissolved oxygen. Total dissolved solids were 1761±780.00mg/l. The quantity of COD was 650.0±168 mg/l, while Biological oxygen demand during three days at temperature 27 °C was confirmed as 306.0±175.00 mg/l. Heavy metal were determined in order of Fe > Pb > Mn > Cr and Al > Ni > Zn > Cu that was proceeded as pollutants under lethal exposure for the aquatic fauna. Histopathological variations of the gills and liver tissues of *L. rohita* have been studied earlier [16]. The marked histopathological variations for instance degeneration, necrosis and disorganization of lamellae and hyperplasia of epithelial cells was determined in the gills of the *L. rohita* captured from contaminated site and also compared to the fish species taken from a control site. The liver histopathology of *L. rohita* showed aggregation, severe hypertrophy of hepatocytes, cytoplasmic deterioration, and haemolysis between hepatocytes, focal necrosis and other degenerations of liver cells. Impure water has emerged high histopathological variations in *L. rohita*. Several basal cells and result revealed less severe necrosis, devastation of the mucosal cells was examined, chronic dose (0.9% for 30 days) of TME resulted in substantial damages in the structural design of *Labeo rohita* gill [15]. Due to the exposure of sub-lethal concentration surface of epithelium in the primary gill lamellae became apart. Gill arches detected fall apart mass of the spongy of spongy cartilage with indistinct arteries. Reliability of basal cells was not revealed, but some cells in the primary lamellae were showed less severe necrosis appearance. Secondary gill lamellae showed unusual thickness and full damage of the mucosal cells. In this study different variations and damages revealed severe degeneration of the secondary gill filaments, degenerative changes in the primary gill filament in the gill of *C. mrigala*, *L. rohita* and *H. molitrix*. Short fusion of secondary lamellae, less changes in the integrity of lamellae, less Proliferation of mucus cell and severe haemorrhage showed in the gill of *C. catla*. There was great loss of gill lamellae, fusion of secondary lamellae, haemorrhage in primary and secondary lamellae, degenerative and necrotic changes in the epithelium of gill filament reported in the gill of *C. mrigala*, *L. rohita* and *H. molitrix*. Severe degenerative changes in the secondary lamellae, proliferation of mucus cell, atrophy of gill filament, and loss of structural integrity of lamellae, congestion of blood vessels in gill filament in the gill of *C. mrigala*, *L. rohita* and *H. molitrix* revealed. Histological study of fish liver due to sub lethal concentration of textile effluents, revealed severe damages of cellular structure, high scale necrosis and haemolysis due to damage of erythrocytes and inflammation of the hepatic cells, eccentric nuclei, vacuole emergence were commonly found in the liver of *C. mrigala*,

L. rohita, *H. molitrix* and *C. catla*. High level of haemorrhage and focal area of necrosis, intravascular haemorrhage, vacular degeneration, severe haemorrhage, hepatocytes degeneration, necrosis, haemolysis and, erythrocyte infiltration in blood sinusoid, Eccentric nuclei, cytoplasmic vaculation and dilation of vein in the liver of *C. mrigala*, *L. rohita* and *H. molitrix*. Histological changes in the Kidney tissue of yellow *mystus*, *Hemibagrus filamentus*, and the dilation of both Bowman's gap and glomerulus examined and also showed kidney lesions of the treated kidney as in contrast with control kidney which have been no lesions. In the renal tubular cells necrosis was examined in few region of the kidney [17]. In this study Due to sub-lethal exposure of textile industrial effluents different variations seem in the kidney of *Cirrhinus mrigala*, *L. rohita*, *H. molitrix* and *C. catla*. Histological examination revealed that glomerular shrinkage occurred in the kidney, increase gap between glomerulus and Bowman's capsule, increased tubular lumen in the kidney of *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla*.

5. Conclusion

Study of histology and histopathology of different organs revealed that there was large scale of degenerations. Histopathological examination of gill due to sub-lethal exposure of Textile Industries effluents, different variations and damages revealed, severe degeneration of the secondary gill filaments and degenerative changes in the primary gill filament in the gill of *C. mrigala*, *L. rohita* and *H. molitrix* while short fusion of secondary lamellae, fewer changes in the integrity of lamellae, less proliferation of mucus cell and severe hemorrhage showed in the gill of *C. catla*. Glomerular shrinkage, damages of blood vessels, increase gap between glomerulus and Bowman's capsule, intact of tubules vaculation, and increased tubular lumen in the kidney of all treatment group. Histopathological study of liver reported severe damages of cellular structure, high scale necrosis, hemolysis due to damage of erythrocytes, inflammation of the hepatic cells, eccentric nuclei and vacuole emergence commonly found in the liver of *C. mrigala*, *L. rohita*, *H. molitrix* and *C. catla*

6. References

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