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Histopathological alterations in the gill, liver and kidney of *Labeo rohita* on exposure to plywood effluent

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Abstract
Study was conducted to assess the histopathological damage of Gill, Liver and Kidney in the fresh water fish, *Labeo rohita* after sublethal exposure to plywood effluent. A parallel control was maintained simultaneously along with the fishes exposed to sublethal concentration of plywood effluent for short term (24, 48, 72 and 96 hours) and long term (10, 20 and 30 days) durations. Gill, Liver and Kidney of exposed individuals exhibited some remarkable changes in their histology in comparison to control. Prominent changes include degeneration of epithelial lining, epithelial proliferation in gills, clumping of nucleus, degenerated hepatocyte cells, fatty degeneration in liver and in kidney shrinkage of the glomerulus and dilation of tubular lumen. Duration of exposure appears to have a profound effect on gill, liver and kidney. With increasing duration of exposure, histopathological damages become more severe.

Keywords: Plywood effluent, LC_{50}, *Labeo rohita*, histopathology

1. Introduction
More and more of our habitats are being deteriorated day by day due to increased environmental pollution by means of various anthropogenic activities. The industrial effluents that contain toxic substances like heavy metals, pesticides and other chemicals are discharged into the water bodies. Any pollutant which is discharged into water will change the features of water like surface tension, thermal properties, conductivity, density and pH value. The acidic and alkaline pollutants destroy most of the invertebrates and microorganisms.[5] As a result, the aquatic fauna and flora are adversely affected which lead to bioaccumulation in aquatic organisms and bioconcentration in higher vertebrates.[1] Eco-friendly environment is a necessary condition for the well-being of human race. The degree of contamination in aquatic environment is frequently assessed by comparingcontaminent concentration in associated biota. Water pollution induces pathological changes in fish.
The assessment of histopathological changes in animal tissues is an invaluable tool in the determination of environmental pollution[16, 17]. It serves as an early warning sign of injury to cells, tissues and organs. One of the great advantages of using histopathological biomarkers in environmental monitoring is that this category of biomarkers allows examining specific target organs including gills and liver that are responsible for vital functions such as respiration, excretion, accumulation and biotransformation of xenobiotics in the fish[7] and serve as warning signs of damage to animal health[11]. The plywood industry involves the production of plywood from thin layers of wood veneer. The effluent from the plywood industry affects the surface water bodies and ground water adversely. The present study was performed to evaluate the sublethal effects of plywood effluent on histopathological alterations in the vital organs such as gill, liver and kidney of fresh water fish, *Labeo rohita* which was selected as a laboratory animal model. The *Labeo rohita* was selected for the bioassay experiments because it is one of the most economically important fresh water fish that is extensively cultured in India, Nepal, Bangladesh and other countries.

2. Materials and Methods
Active specimens of *Labeo rohita* (10.50± 0.10 in length and 16.85 ± 1.040 gms in weight) of both sexes were used for the experiments. All fishes used were procured from local aqua agri farm. Fish was treated with 0.02 % $KMNO_4$ for two minutes to avoid any dermal infection. The fishes were then acclimatized under laboratory conditions in static conditions for 15 days

and kept in rectangular glass aquaria of capacity 200 liters. They were fed with commercial fish food *ad libitum.* The faecal matter and other waste material were siphoned off daily to reduce ammonia content in water.

Thirty fishes were exposed to sublethal concentration of plywood effluent for the study. Simultaneously two control groups, one each for short term and long term were also maintained. The experimental set up was maintained properly by the daily renewal of effluent and provided fish feed daily. After the duration of experiment, fishes were taken from the respective concentrations and subsequently the vital organs (gill, liver and kidney) were removed by live dissection and fixed in Davidson fluid for 24 hours. Then organ tissues were washed with 70% ethanol and dehydrated through a graded series of ethanol [15, 24]. Subsequently, the tissues were embedded in paraffin sectioned at 4 to 5 mm thickness stained with haematoxylin and eosin and finally examined under photomicrography [14].

2.1 Determination of LC_{50}

LC_{50} for plywood effluent comes out to be 1.27 mg/L. Based on the sublethal concentration of 0.127 mg/L ($1/10^{th}$ of LC_{50}) fishes were exposed to short term (24, 48, 72 and 96 hours) and long term (10, 20 and 30 days) durations to examine the histopathological changes in the organs like gill, liver and kidney.

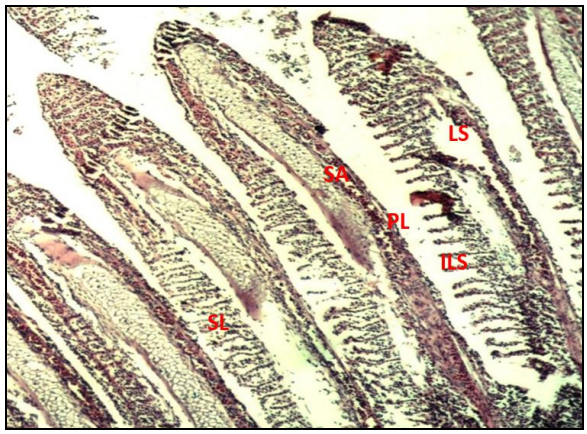
3. Results

3.1 Histopathology of Gill:

The untreated gills showed an arrangement of filaments in double rows and the secondary lamellae arise from these filaments (Plate I-A). The present investigation on the histology of gills in *Labeo rohita* exposed to plywood effluent revealed severe damages due to the nature of the effluent. Marked histopathological changes including necrosis and degenerative changes in the epithelial cells and pillar cells of the gills. The capillaries in the secondary lamellae were dilated. The epithelial cells as well as pillar cells were exfoliated. Consequently haemorrhage occurred in the damaged areas. Partially empty rachis with sloughed tissues indicated disorganization of gill structure. Original shape of the secondary gill filaments was distorted and curling of these filaments were observed. At some places the lamellae appeared like swollen bulging sacs filled with blood cells. Epithelial wall of secondary lamellae seemed to be ruptured. Loss of cellularity and fragmentation of gill lamellae were the most common feature indicating serious impairment in the respiratory function of the gills (PlateII B-E). It was observed that the histopathological alterations in the gill of fish, *Labeo rohita* increased with the increase in the days of exposure (Plate III F-H).

Plate I Histopathology of Gill of *Labeo Rohita*

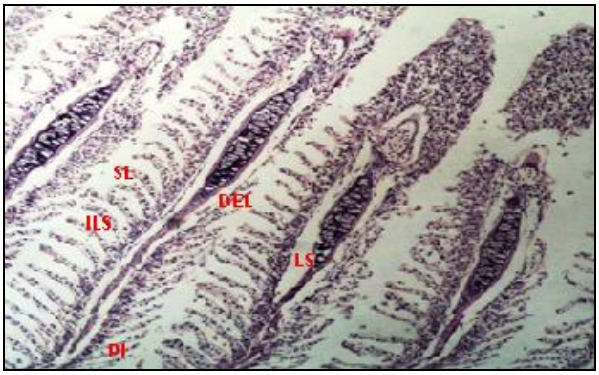
A. Control Gill section of *Labeo Rohita*



PL - Primary Lamellae, SL - Secondary Lamellae
LS – Lamellar Space, ILS - Inter Lamellar Space
SA - Supporting Axis

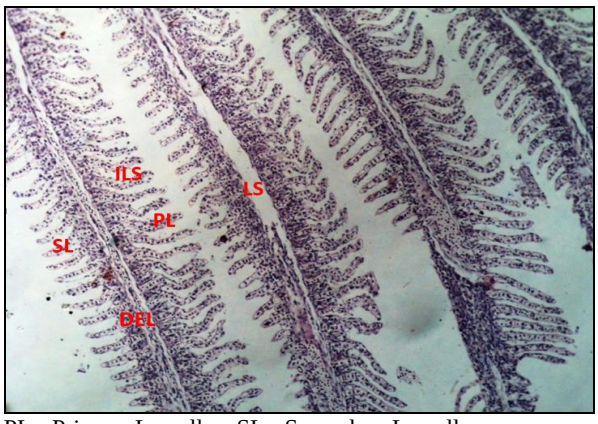
Plate II

B. Gill section of fish exposed to 24 hours of plywood effluent



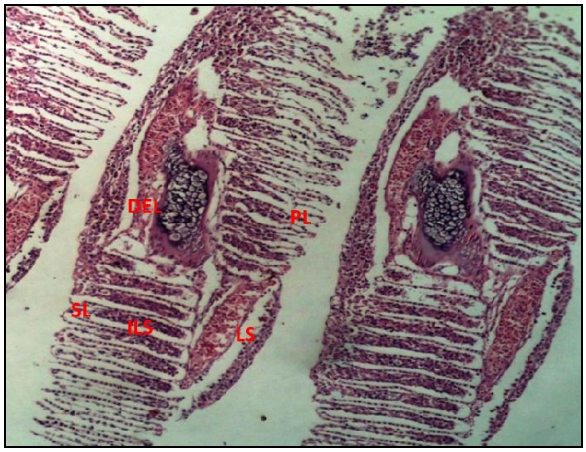
PL - Primary Lamellae, SL - Secondary Lamellae
LS - Lamellae Space, ILS - Inter Lamellar Space
DEL-Degeneration of Epithelial Lining

C. Gill section of fish exposed to 48 hours of plywood effluent



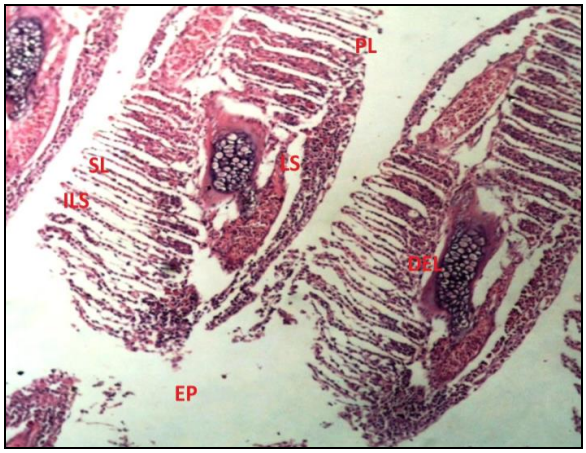
PL - Primary Lamellae, SL - Secondary Lamellae
LS - Lamellar Space, ILS - Inter Lamellar Space
DEL - Degeneration of Epithelial Lining

D. Gill section of fish exposed to 72 hours of plywood effluent



PL - Primary Lamellae, SL - Secondary Lamellae
LS - Lamellar Space, ILS - Inter Lamellar Space
DEL - Degeneration of Epithelial Lining

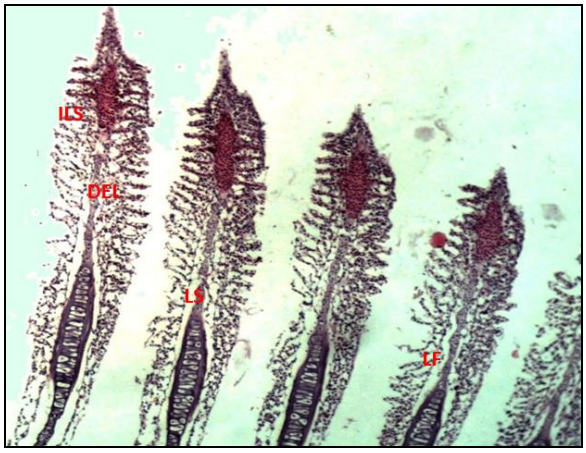
E. Gill section of fish exposed to 96 hours of plywood effluent



PL - Primary Lamellae, SL - Secondary Lamellae,
LS - Lamellar Space, ILS - Inter Lamellar Space,
DEL- Degeneration of Epithelial Lining,
EP - Epithelial Proliferation

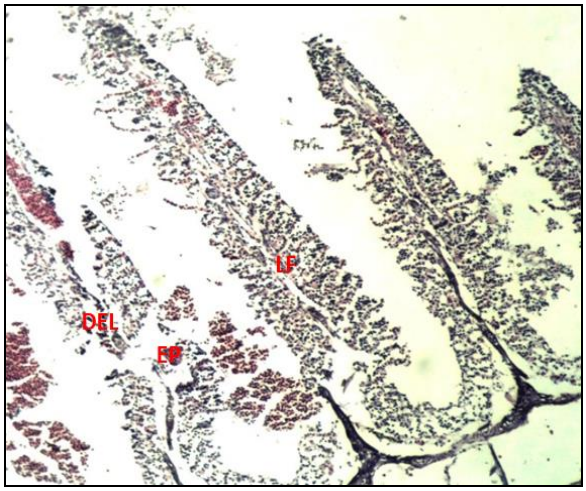
Plate III

F. Gill section of fish exposed to 10 days of plywood effluent



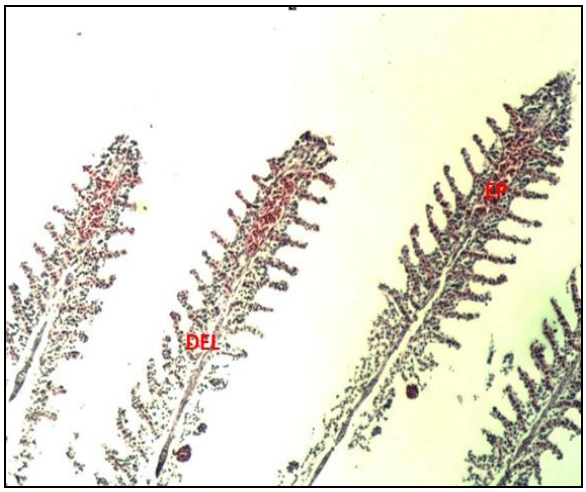
LF - Lamellar Filament, LS - Lamellar Space,
DEL - Degeneration of Epithelial Lining
ILS - Inter Lamellar Space

G. Gill section of fish exposed to 20 days of plywood effluent



LF - Lamellar Filament, DEL - Degeneration of Epithelial Lining, EP - Epithelial Proliferation

H. Gill section of fish exposed to 30 days of plywood effluent



EP - Epithelial Proliferation,
DEL - Degeneration of Epithelial Lining

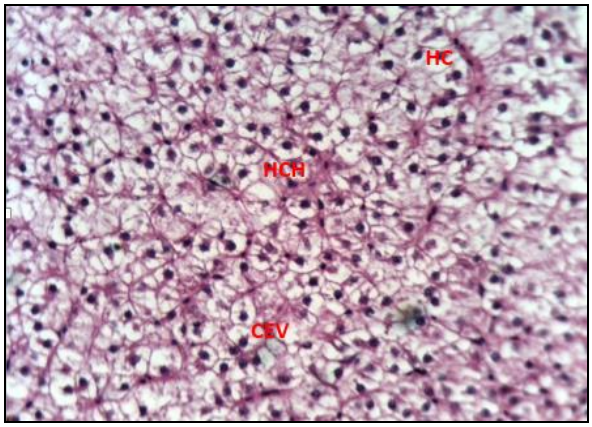
3.2 Histopathology of Liver

The unexposed fish shows intact liver structure with unaltered hepatocyte cells, hepatic cords and central efferent vein (Plate IV-A). In the study, several histological alterations were observed in *Labeo rohita* on exposure to short term duration. These pathological alterations included dilation in hepatic cells, focal areas of necrosis, hepatic cells degeneration and vacuolar degeneration with infiltration of lipid, blood vessels degeneration, haemorrhage and severe dilation in liver cells (Plate V B-E). The damage was severe in those fishes exposed to long term period (Plate VI F-H).

Plate IV

Histopathology of Liver of *Labeo rohita*

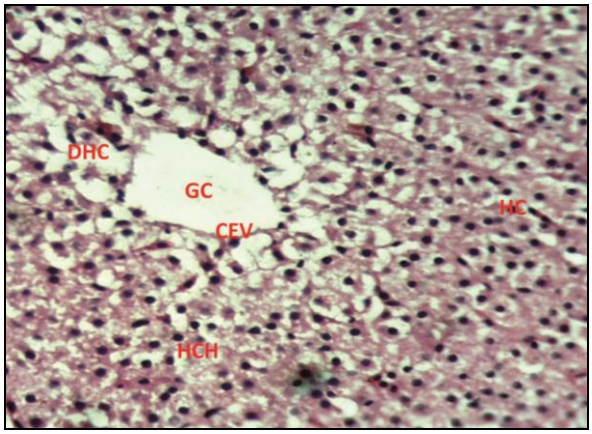
A. Control Liver Section of *Labeo rohita*



HC - Hepatocyte Cells, HCH - Hepatic Cords
CEV - Central Efferent Vein

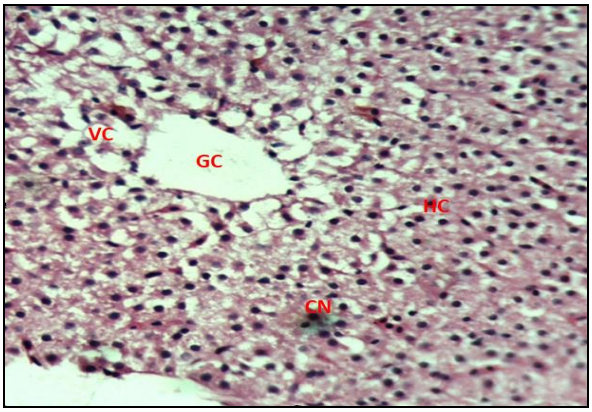
Plate V

B. Liver section of fish exposed to 24 hours of plywood effluent



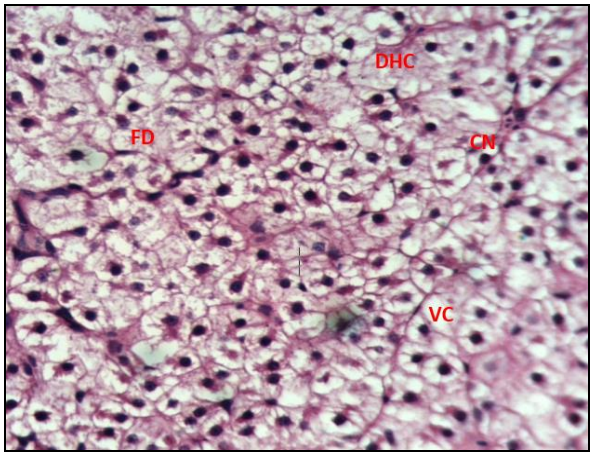
GC - Gilssen's Capsule
DHC - Degenerated Hepatocyte Cells
HC - Hepatocyte Cells, HCH - Hepatic Cords
CEV - Central Efferent Vein

C. Liver section of fish exposed to 48 hours of plywood effluent



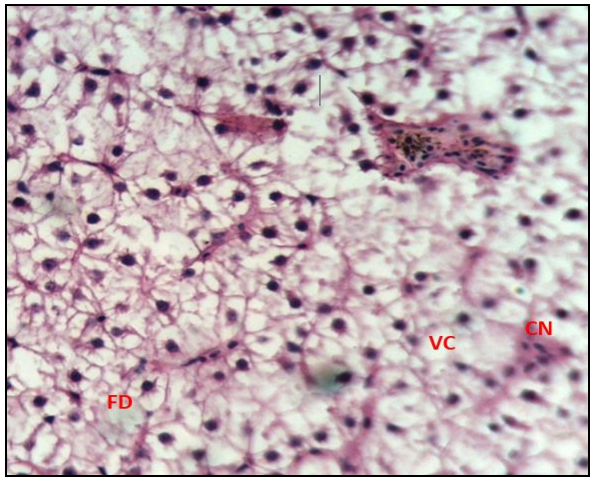
HC - Hepatocyte Cells, GC - Gilssen's Capsule
VC – Vacuoles, CN - Clumping of Nucleus

D. Liver section of fish exposed to 72 hours of plywood effluent



DH - Degenerated Hepatocyte Cells
VC - Vacuoles
CN - Clumping of Nucleus
FD - Fatty Degeneration

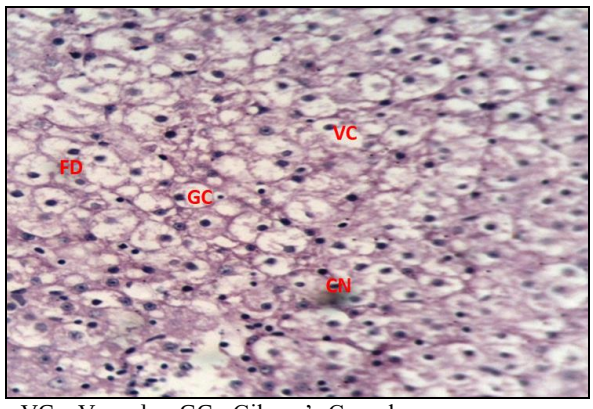
E. Liver section of fish exposed to 96 hours of plywood effluent



VC – Vacuoles, GC - Gilssen's Capsule
FD - Fatty Degeneration, CN - Clumping of Nucleus

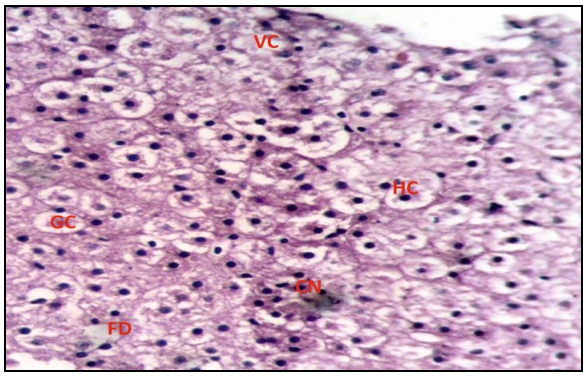
PLATE VI

F. Liver section of fish exposed to 10 days of plywood effluent



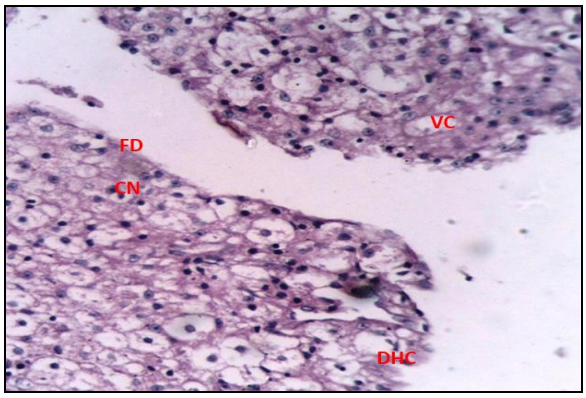
VC – Vacuoles, GC - Gilssen's Capsule
FD - Fatty Degeneration, CN - Clumping of Nucleus

G. Liver section of fish exposed to 20 days of plywood effluent



HC - Hepatocyte cells, VC - Vacuoles
GC - Gilssen's Capsule, FD - Fatty Degeneration
CN - Clumping of Nucleus

H. Liver section of fish exposed to 30 days of plywood effluent

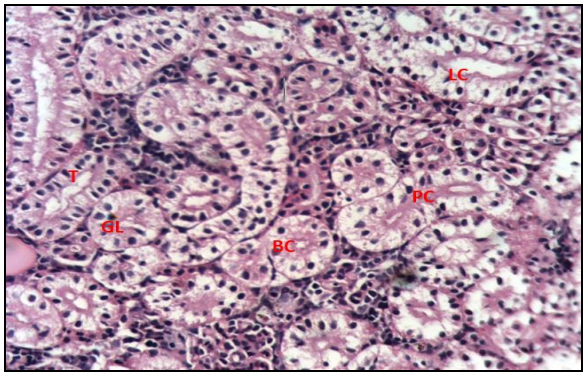


VC – Vacuoles, DHC - Degenerated Hepatocyte Cells
FD - Fatty Degeneration, CN - Clumping of Nucleus

3.3 Histopathology of Kidney

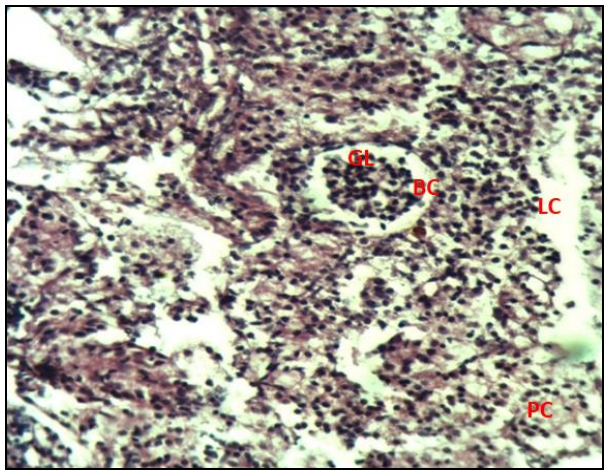
The unexposed fishes shows kidney with unaltered structures like glomeruli, lymphoid cells, parenchyma cells, Bowman's capsule and tubules (Plate VII A). Meanwhile, short term exposed fishes showed shrunkened glomerulus, damaged tubule nucleus and Bowman's capsule (Plate VIII B- E). The damages were severe in long term exposed fishes. These fishes showed highly altered kidney with almost all its structures degenerated (Plate IX F-H).

Plate VII
Histopathology of kidney of *Labeo Rohita*
A. Control Kidney Section of *Labeo rohita*



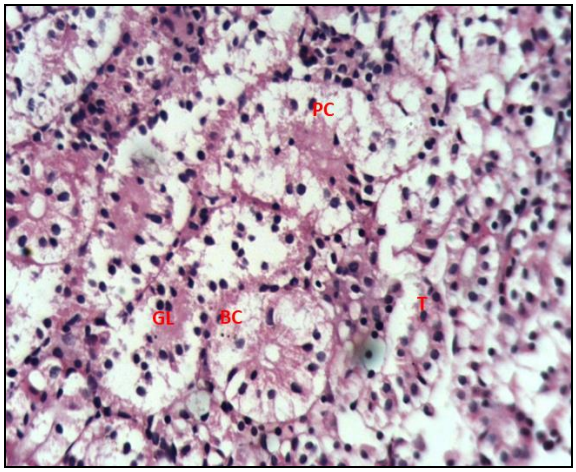
GL - Glomeruli
LC - Lymphoid Cells
PC - Parenchyma Cells
BC - Bowman's Capsule
T - Tubules

Plate VIII
B. Kidney Section of fish exposed to 24 hours of plywood effluent



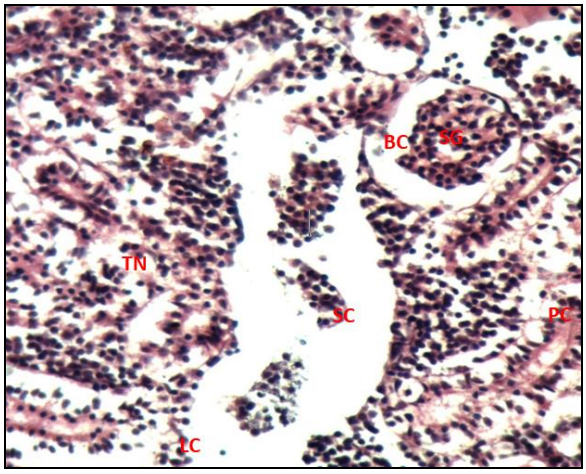
GL – Glomeruli, LC - Lymphoid Cells
PC - Parenchyma Cells, BC - Bowman's Capsule

C. Kidney Section of fish exposed to 48 hours of plywood effluent



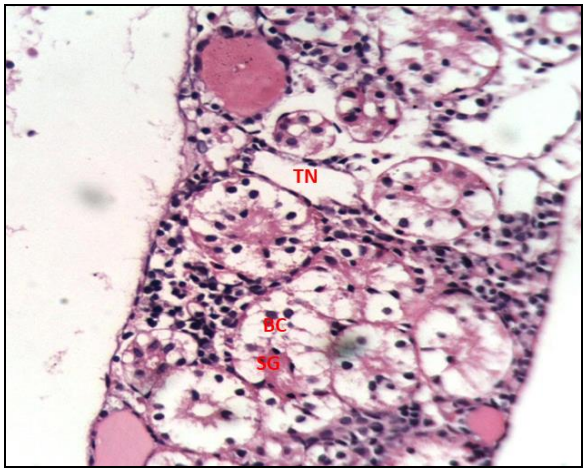
PC – Parenchyma Cells, BC - Bowman's Capsule
T – Tubules, GL – Glomeruli

D. Kidney Section of fish exposed to 72 hours of plywood effluent



LC - Lymphoid Cells, PC - Parenchyma Cells
BC - Bowman's Capsule, SG - Shrunkened Glomerulus
TN - Tubule Nucleus, SC - Shrunken of Cells

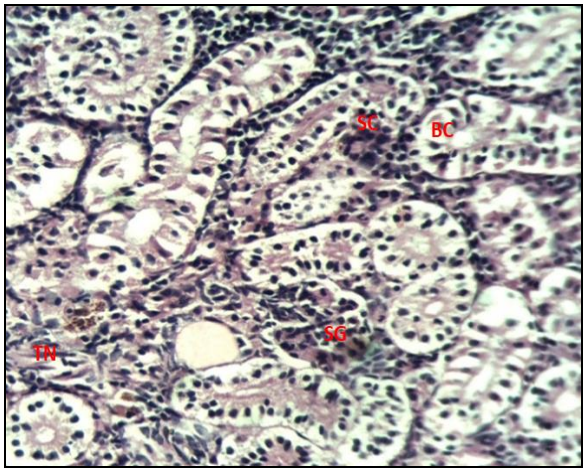
E. Kidney Section of fish exposed to 96 hours of plywood effluent



BC - Bowman's Capsule, SG - Shrunkened Glomerulus
TN - Tubule's Nucleus

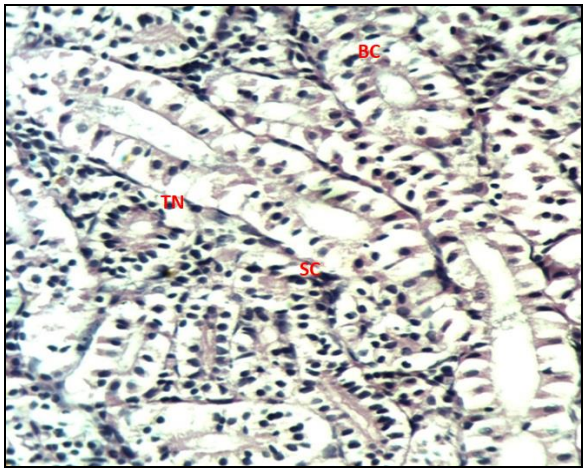
Plate IX

F. Kidney Section of fish exposed to 10 days of plywood effluent



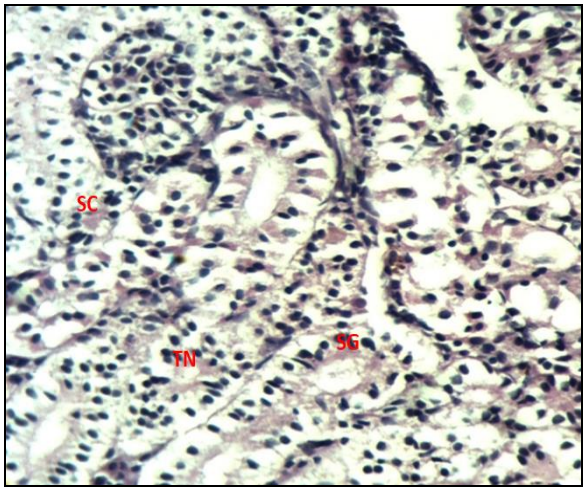
BC - Bowman's Capsule, SC - Shrunken of Cells
SG – Shrunkened Glomerulus, TN - Tubule's Nucleus

G. Kidney Section of fish exposed to 20 days of plywood effluent



BC - Bowman's Capsule, SC - Shrunken of Cells
TN - Tubule's Nucleus

H. Kidney Section of fish exposed to 30 days of plywood effluent



SC - Shrunken of Cells, SG - Shrunkened Glomerulus
TN - Tubule's Nucleus

4. Discussion

The extent of damage in the fish organs increases as the exposure period increases. The histopathological alterations in the study indicates the toxic nature of plywood effluent.

The present study was in the line of degenerative changes in respiratory epithelium of the fishes exposed to aquatic pollutants metals [6]. The noticed clubbed shaped lamellae indicating progressive degeneration in the gill due to fusion of secondary lamellae. As a result of lamellar fusion, the lamellar surface area may be reduced minimizing oxygen delivery to the tissues [18]. They further noted that after chronic exposure to the pollutants progressive degenerative changes resulted into complete disorganization of the gill lamellae. The present study also agreed with the findings of severe damages in the respiratory epithelium of gills exposed to heavy metals [8, 13]. Damages occurred at cellular level in the gills of *Cirrhinus mrigala* exposed to metallic salts [21]. In the present study, fishes exposed to very low concentration of effluent resulted in remarkable histopathological changes which lead to decreased efficiency of gill surface for gaseous exchange. Mucous secretion mainly from the base of the primary lamellae and proliferated primary lamellar epidermis form a tenacious outer layer of different consistency leading to obstruction of respiratory exchange. Another characteristics pathological change of the gill observed in the present study was lamellar telangiectasias recognized grossly by the presence of the small red spots on the secondary lamellae. The lesion had its genesis in the rupture of the retaining pillar cells which normally joins the dorsal surface of secondary lamellae to the ventral. The result was dilation of the lamellar capillary and pooling of the blood which thrombosed and eventually fibrosed. Multiple telangiectasias occurred when the fishes exposed to metals [22].

The damages observed in the present study is explained by the fact why liver plays an important role in vital function, basic metabolism and accumulation, transformation and excretion of contaminants [3, 28]. Moreover, the liver as the major organ of metabolism comes in close contact with xenobiotics absorbed from the environment and liver lesions are often associated with aquatic pollution. Deteriorations of the liver can be due to the storage of heavy metals in the body [23]. Histopatholgical alterations observed in the liver of *Gymnocephalus cernua* collected from the Elbe Estuary

contaminated by domestic, industrial and agricultural pollutants [26]. The liver anomalies in this work are similar to those observed in *Clarias gariepinus* after exposure to lead. These authors observed cells hepatic degeneration after 3 days and necrosis after two weeks exposure to lead [2]. The present results are in agreement with those observed in other fish species under the influence of different pollutants [4, 19, 31].

The kidney is a vital organ of body and proper kidney function is to maintain the homeostasis. It is not only involved in removal of wastes from blood but is also responsible for selective reabsorption which helps in maintaining volume and pH of blood and body fluids and also makes erythropoiesis [12]. The teleostean kidney is one of the first organs to be affected by contaminants in the water [29]. Most common alterations found in the kidney of fishes exposed to water contamination are tubule degeneration (cloudy swelling and hyaline droplets) and changes in the corpuscle such as dilation of capillaries in the glomerulus and reduction of Bowman's space [27]. Exposure to metals frequently causes alterations in the tubules and glomerulus in the perch (*Lates calcarifer*) exposed to cadmium [29]. Bowman's capsule cells and melanomacrophages in the kidney of trout (*Salmo trutta*) and tilapia (*Oreochromis mossambicus*) found swollen exposed to mercuric chloride [9]. Similar alterations were found in fishes exposed to organic contaminants [30] and mixed environmental contaminants [20, 25]. In the present study, kidney of the fish often showed cloudy swelling in tubule cells. This alteration can be identified by the hypertrophy of the cells and the presence of small granules in the cytoplasm which takes on the appearance of a net. This initial stage in the degeneration process can progress to hyaline degeneration characterized by the presence of large eosinophilic granules inside the cells. These granules may be formed inside the cells or by the reabsorption of plasma proteins lost in the urine, indicating damage in the corpuscle [10, 27]. In more severe cases, the degenerative process can lead to tissue necrosis [27]. The presence of tubule degeneration coupled with the absence of necrosis in the kidney in the present study indicates that the kidney suffered damage after exposure to the water of the plywood industry.

5. Conclusion

The present study shows the harmful effect of pollution due to industrialization. Even though the development and industrialization are the two sides of the same coin, we should be very responsible to protect our environment; its flora and fauna. The results of the present study have proven remarkable damages to gills, kidney and liver of fishes exposed to plywood effluent. The industrial effluents discharged into the water bodies is the main reason of depleting fish fauna among water bodies.

6. Acknowledgement

The author is deeply indebted to Department of Zoology, Kongunadu Arts and Science College, Coimbatore for carrying out this research work.

7. References

1. Ackerman PA, Iwama GK. Physiological and Cellular Stress Responses of Juvenile Rainbow Trout to Vibriosis. Journal of Aquatic Animal Health. 2001; 13:173-180.
2. Aly SM, Mona SZ, Halam ME. Pathological, biochemical, haematological and hormonal changes in cat fish (*Clarias gariepinus*) exposed to lead pollution. Journal of the Egyptian Veterinary Medical Association. 2003; 63:331-342.
3. Athikesavan S, Vincent S, Ambrose T, Velmurugan B. Nickel induced histopathological changes in the different tissues of fresh water fish *Hypophthalmichthys molitrix* (Valenciennes). Journal of Environmental Biology. 2006; 27(2):391-395.
4. Atif MEN, Soaad AM, Safaa IT. Bioaccumulation of some heavy metals and histopathological alterations in the liver of *Oreochromis niloticus* in relation to water quality at different localities along the river Nile, Egypt. World Journal of Fish and Marine Sciences. 2009; 1(2):105-114.
5. Barber D, Shrma MS. Experimentally induced bioaccumulation and elimination of cadmium in freshwater fishes. Pollution Research. 1998; 17: 99-104.
6. Dhanapakiam P, Sampoornai V, Kavitha M, Ramasamy VK, Chandrakala A, Aruna KC. Gill lesions in the major carp, *Labeo rohita* exposed to lethal and sublethal concentrations of tannery effluent. Journal of Environmental Biology. 2004; 25:331-336.
7. Gernhofer M, Pawet M, Schramm M, Muller E, Triebskorn R. Ultrastructural biomarkers as tools to characterize the health status of fish in contaminated streams. Journal of Aquatic Ecosystem Stress and recovery. 2001; 8:241-260.
8. Gupta N, Dua A. Mercury induced architectural alterations in the gill surface of a fresh water fish, *Channa punctatus*. Journal of Environmental Biology. 2002; 23(4):383-386.
9. Handy RD, Penrice WS. The influence of high oral doses of mercuric chloride on organ toxicant concentrations and histopathology in rainbow trout, *Oncorhynchus mykiss*. Comparative Biochemistry and Physiology (C). 1993; 06:717-724.
10. Hinton DE, Lauren DJ. Liver structural alterations accompanying chronic toxicity in fishes: potential biomarkers of exposure. Biomarkers of Environmental Contamination. (Lewis Publishers, Boca Raton), 1993, 51.
11. Hinton DE. Cells, Cellular Responses and their Markers on Chronic Toxicity in Fishes. In Aquatic Toxicology: Molecular, Biochemical and Cellular Perspectives. Eds. (Lewis Publishers, Boca Raton), 1995, 207.
12. Iqbal F, Qureshi IZ, Ali M. Histopathological changes in the kidney of common carp, *Cyprinus carpio* following nitrate exposure. Journal of Research in Science Teaching. 2004; 4:41-418.
13. Jagadessan G. *In vivo* recovery of gill tissue of fresh water *Labeo rohita* after exposure to different sublethal concentration of mercury. Pollution Research. 1999; 19(3):289-291.
14. Kaneko JJ. Clinical Biochemistry of Domestic Animals. 4th ed. (Diego Academic Press Inc. California), 1989, 132.
15. Kelly WR. Veterinary Clinical Diagnosis. 2nd ed. (Balliere Tindall, London), 1979, 266.
16. Malik GM, Raval HV, Ahmed Khali, HK. Toxic effects of effluent on mortality and behaviour changes in fresh water fish, *Poecilia reticulate*. Journal of Environmental Research and Development. 2012; 7(2A):1036-1039.
17. Moore MJ, Myers MS. Pathology of chemical associated Neoplasia in Fish. Aquatic Toxicology. 1994; 24:327.
18. Muller ME, Sanchez DA, Bergman HL, Mcdonald DG,

Wood CM. Nature and time course of acclimation to aluminium in juvenile brook trout (*Salvelinus fontinanils*) Histology. *Canadian* Journal of Fisheries and Aquatic Science. 1991; 48:2016-2027.
19. Olfat MW, Zenab AEG. Comparative impact of different waste sources on the reproductive parameters and histology of Gonads, Liver and Pituitary gland of *Siganus rivulatus.* Journal of Applied Sciences Research. 2007; 3:231-236.
20. Pacheco M, Santos MA. Biotransformation, genotoxic and histopathological effects of environmental contaminants in European eel (*Anguilla anguilla* L.). Ecotoxicology and Environmental Safety. 2002; 53(3):331-347.
21. Palaniappan PLRM, Karthikeyan S, Sabhanayakam S. Studies on the effects of heavy metal nickel on gills of fingerlings of an edible fish, Cirrhinus mrigala. Pollution Research. 2003; 22(2):247-250.
22. Palanisamy P, Sasikala G, Mallikaraj D, Bhuvaneshwar N, Natarajan GM. Histopathological lesions in gill of air breathing cat fish *Mystus cavasius* exposed to electroplating industrial effluent nickel. International Journal of Applied Biology and Pharmaceutical Technology. 2011; 2(2):150-155.
23. Rodrigues EL, Fanta E. Liver histopathology of the fish *Brachydanio rerio* after acute exposure to sublethal levels of organophosphate Dimetoato 500. Revista Brasileira de Zoologi. 1998; 15:441-450.
24. Schalm OW, Jain NC, Carrol EJ. Veterinary Haematology, 3rd Ed. (Philadelphia, Leaard Febiger), 1995, 15.
25. Schwaiger J, Wanke R, Adam S, Pawert M, Honnen W, Triebskorn R. The use of histopathological indicators to evaluate contaminant related stress in fish. Journal of Aquatic Ecosystem, Stress and Recovery. 1997; 6(1):75-86.
26. Sorensen EM. Metal poisoning in fish: Environmental and life Sciences Associates, Austian Texas. (CRC Press Inc., Boston), 1991.
27. Takashima F, Hibya T. An atlas of fish histology: normal and pathological features, 2nd ed, (Tokyo, Kodansha), 1995.
28. Tayel SI, Yacoub AM, Mahmoud SA. Histopathological and responses to fresh water pollution in the Nile cat fish, *Clarius gariepinus*. Journal of Egyptian Academic Society for Environmental Development. 2008; 9(4):43-60.
29. Thophon S, Kruatrachue M, Upathan ES, Pokethitiyook P, Sahaphong S, Jarikhuan S. Histopathological alterations of white seabass, *Lates calcarifer* in acute and subchronic cadmium exposure. Environmental Pollution. 2003; 121(3):307-320.
30. Veiga ML, Rodrigues EL, Pacheco FJ, Ranzani Pavia MJT. Histopathological changes in the kidney tissue of *Prochilodus lineatus*, 1836 (Characiformes, Prochilodontidae) induced by sublethal concentration of Trichlorfon exposure. Brazilian Archives of Biology and Technology. 2002; 45:171-176.
31. Zahra K, Amin M, Reza K. Bioaccumulation of some heavy metals and histopathological alterations in liver of *Euryglossa orientalis* and *Psettodes erumei* along North Coast of the Persian Gulf. African Journal of Biotechnology. 2010; 9(41):6966-6972.