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RESEARCH ARTICLE

HISTOPATHOLOGICAL CHANGES AS ALTERATIONS, A BIOMARKER STUDY IN THE FISH CATLA CATLA EXPOSED TO SUBLETHAL CONCENTRATIONS OF CYFLUTHRIN, A SYNTHETIC PYRETHROID

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Abstract

The fish Catla catla exposed to a synthetic pyrethroid Cyfulthrin, type II category with a cyano group in sublethal concentration ($1/10^{th}$ of LC_{50} values of 96 hrs) 0.14 mg/L for 10 days. The toxicant had effected on the entry point gill, rendering the fish not to have any normal intake of oxygen and finally the fish succumb to death. The important metabolic organ, liver got effected wherein the metabolism is impaired and the growth of the fish is also not normal. At the exit point, the aminotellic fish kidney, both haemopoietic and excretory organ got also effected and that resulted impairment of excretion function apart from haemogenesis. All will be discussed with the available literature.

Introduction:-

Thanks to Marie Francois Xavier Bichat a French Anatomist and Pathologist the father of histology who had invented the study and technique. Yanchova et al (2015) referred such studies on one of the biomarker targeted on fish which served as tool in the ecotoxicology study of the aquatic pollution. The architectural damage as lesions that are going to be observed in the vital organs, gills, liver and kidney, the entry, metabolic and exit points of the fish because they perform the important life process of respiration metabolism and excretion, which resulted damaged by the toxicant action.

According to one estimate the current world population may reach by 2050 by 9.8 billion and the use of pesticides may reach 1000 billion tonnes in terms of their use (Divya and Vinetha, 2022). The utilization and production and variety formulations are many that are introduced into the choices of the agricultural formers and aqua farmers for their use and in fact they are the commercial formulations only in the pesticide market (Aysha Parvin and Umesh Sukla, 2022). They are reiterated as fact by the study of histopathological variations as a tool serves as the biomarker study in the fish.

The scenario of pesticides that contaminated the Ganga waters was given by Zeeshan and Parveen (2022). Protein rich flesh, fish serves as good source of the study, the first vertebrates with closed circulatory system. First the tissues of the fish, due to repository, pave the way of the changes in its internal structures. Sinha et al (2022). Due to increase in the use of the chemicals as pesticides, the priceless ecosystems to human beings got effected Saravali

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et al (2022). Shefali et al (2021) cautioned about the impact of aquatic environment due to pesticide use (about 300 billion kilograms consistently), 10% of it reach open spill over that is utilized, universally. Rajmohan et al (2020), while reviewing the pesticide contamination as a defilement act in the aquatic environment, all depends upon mobility, bioavailability of pesticides in soils/atmosphere/water bodies all with reference to the chemical absorption and desorption mechanisms from soils only and ultimately effect the biological molecules and tissues primarily which can be revealed only by study familiarly as histopathological changes.

Even Pradip Kumar Maurya et al (2019) too had an emphasis of the ecosystems damage particularly of aquatic nature and all exposed organisms got effected by the pesticides.

Sana Ullah et al (2019a) mentioned that biomarker study of the one, like histopathology serves as better tool in the assessment of the toxic stress action in fish. The same was reiterated by Prusty et al (2015), Ahrar Khan et al (2010 & 2014), Anilava Kaviraj and Abhik Gupta (2014), for synthetic pyrethroids and for pesticides in general, Ullah and Zorriezahara (2015) and Krishna Murty et al (2013).

Such histopathological studies of the toxicant pesticides were reported in the above mentioned review articles as well as by individual studies by Naquash Khan et al (2021), Dhivya and Devadason (2021), Shehezad et al (2021), Padmini et al (2021), Rachele et al (2020), Kadam et al (2019), Arumugan Stalin et al (2019), Sezgi Sezgi Arman and Sema (2019), Mohammad et al (2019), Greeshma et al (2019), Sucitha et al (2018), Japamalai (2017), Bhat et al (2016), Nannu et al (2015), Nidhi Srivastava (2014), Kaya et al (2013), Manju Singh and Santosh Kumar (2013), Tamil Selvi and IIlavazhana (2012), Yogita Devi and Abha Misra (2012).

However, not of much work with the present studied toxicant Cyfluthrin 10% WP was carried to the fish Catla catla, by taking $1/10^{th}$ of LC_{50} value .of 96h of continuous flowthrough system for the first time such study is attempted.

Hence, in the present study, the fish Catla catla was taken, which is a food of many, and palatable one of the majority human beings among the freshwater fish. It is tested, the histopathological aspects of the three organs gills, liver and kidney, (the entry point, for metabolic point and exit point of the toxicant).

Material and Methods:-

Fresh water fish major carp, Catla catla was first acclimatized to the laboratory conditions for 10 days. 50 numbers of the fish are exposed to 10% WP Cyfluthrin for 10 days by taking into consideration of LC_{50} value of 96 hours ($1/10^{th}$ of the 96 h LC_{50}) value 0.14 µg/L as per the APHA guidelines (1998, 2005 & 2012 and OECD, 2019).

The fish are randomly selected for histopathological examinations after 10 days of exposure. The exposed fish tissues viz., the gill, liver and kidney tissues are separated and also not exposed fish to the toxicant which serve as control.

The method for fixation and processing of Humason (1972) is followed wherein the physiological saline solution of 0.85% NaCl was only used for rinsing and also for cleaning. After fixing in Bouins solutions for 48 hrs, then processed through graded series of different alcohols, cleared in xylol. The tissues/organs are embedded in paraffin wax.

According to Humason (1972) thickness of 6 µm sections were made to cut and stained by Ehlrich haematoxylin/Eosin dissolved in 70% alcohol and mounted on Canada balsam. The sections were observed in digital microscope (Intel ply - Q x 3 at 200 x magnification). By the Q x 3 computer Intel Pentium attached microscope of 400x lens (made in China), the changes as manifested in the respective tissues are examined and photographed.

Observations and Results:-

Gill : Normal structure as the control (Plate-1A)

The fish, Catla catla, the teleost group of the fish which is one among the major carp have four pairs of gills made of cartilage the principle respiring organ through which it filters the waters including the contaminated one apart from food the zooplankton. Each gill apart from paired, has double rows of the gill arches termed primary base of it from which radiate the transverse rows of double, the secondary lamellae and the number of it is more. The entire gills

articles are covered by operculum and the primary lamellae are well protected by the epidermis as a thick covering and in its mucus cells are more in number whose function is to secrete the protein mucin. Below to this external lining of covering lymphoidal tissue whose arrangement is specific. The lamellar arch of the gill containing the primary lamellae part has cuboidal epithelium which contains predominantly chloride cells for transportation of respiratory gases as well as for secretion of ions. The whole component as a compound tissue of several layers squamous epithelium, epidermal cells, cuboidal cells and finally columnar cells. The secondary lamellae play the role in respiration where it provides the facility of diffusion. The secondary lamellae are in turn have microvilli for adsorptive processes and as such provides many functions.

Pathological manifestations observed in the gills (1, B, C & D)
Marked pathological lesions viz., proliferating epithelium (PE), Aneurism (AN) and Fused lamella (FL) as per the plate-1, fig. B, C and D. When compared to the control (Fig. A). In terms of pathological aspects this can be called, hyperplasia of the squamous epithelium, secondary lamellae got the effect of necrosis. The raising abnormally the epithelium apart from swelling that resulted joining of all the secondary lamellae (Figs C, B, and D).

As per the chapter 3, the Oxygen consumption of fish is effected and the same can be taken into cognigence as the tissue damage of the gill which impairs metabolism due to lack of oxygen and the fish was likely to be retardation of the growth. In aquacultural practices, the carp Catla catla is one of the candidate species, if the growth is impaired there is a economical loss in the profitable venture of aquaculture.

General Histology of Liver (Plate-2A)
The entire surface of the liver is covered by a serous membrane (squamous Epithelium) derived from endodermal derivative. It is permeated by the contective tissue that is extended internally as thick cords called by name hepatic cords (Mesodermal derivative). It has many fibers reticular, of both yellow and white and as a whole the entire thing is in the forms of a sinusoid (lobular nature) permeated by canalculi which carry bile Juice at the central position and which chemical contains glycolipid granules of larger quantities.

Liver reddish in colour with polygonal shape with a prominent spherical nucleus which by location centrally positioned. The chromatin part of the tissue is at the periphery and nucleolus is prominent because of its role in synthesis of RNA. The whole tissue/organ has both rough as well as smooth endoplasmic reticular (RER and SER) with acinor cells that serve in the secretory as well as detoxification aspects respectively. It is with numerous mitochondria for energy synthesis because the liver performs several metabolic functions.

Pathological observations (Plate 2, B,C & D)
The cytoplasm of hepatocytes got damaged as can be termed as degeneration wherein also the observation of vacuoles and apart all the blood vessels got narrowed (Figures Vi.2, B, C, & D). necrosis (N), Vacuoles (V) and sinusoids (B), all resulted that paved the way for metabolism to get impaired. The resultant effect of that the candidate species of the culture, Catla catla, when ventured as one of the species in Aquaculture, where growth parameter is impaired the purpose of the very culture is defeated. As a whole, metabolic organ liver got damaged and lesions are observed as above.

General Histology of Kidney (Plate 3 A)
Fish kidney, aminotellic is secretory in function, conservation of salts, aglomerular in its function apart from haemopoietic nature. It is reticulo-endothelial endocrine as well as excretory role, it has to perform. It has glomerular filteration as well as salts conservation of reabsorption that have with proximal as well as distal convoluted tubules (PCT & DCT). The renal corpuscle is near (PCT) and DCT ends into urinary bladder. Taller columnar cells and smaller columnar cells varied in its distribution. The glomalular tissues consists of inner and outer layer of a single flattened epithelium (Renal Epithelium-Squanous), and renal tubules too have covering. Mesangium is the space, that fills the gap between loops of the glomalar capillaries.

The renal tubules (RT) are thin and short in the neck segment which are divided into two parts, segment I and II consisting of cubodal epithelium, densely arranged. The segment of the RT is having microvilli for absorption whereas they are absent in the segment I (Brush border).

Pathology of the Kidney – observed (Plate-3 B, C & D)

The bowmans capsule that got extended in to convoluted tubules, had necrosis n aspect of disintegration (Plate 3, Fig. B). The followed substrates cannot reach the further parts of the kidney. The damage/lesions got extended even to glomerulus as a result, the space between within it increased (Plate 3, Fig. C) and even necrosis and damage as degeneration of the haemopoietic tube of the interstitial component was prominent (Plate 3, Fig. D). As we know the Aminotellic nature of the excretion get effected even in these subtle changes.

Discussion:-

With Synthetic Pyrethroid I & II

Sana Ullah (2022) in the fish Ctenopharyngodon idella reported on bifenthrin a multibiomarker study, in which due to oxidative stress resulted the damage of cells/tissues. Due to enhanced ROS (Reactive Oxygen Species) that resulted lipid peroxidation which had damage of the tissues/organs of the fish due to bifenthrin intoxication. Rapid opercular movements were resulted to in take more oxygen as a result of the damage of the epithelial tissues. The same aspect of the toxic action of bifenthrin visualized in the fish Clarias batracus by Saha et al (2021) and Saha and Saha (2020) and also in the fish Oreochromis mossambicus (Peters, 1952).

Juliat Selvaraj et al (2019) reported on the histopathological changes in the fish, Tilapia (Oreochromis niloticus) and reported on change of gill, liver due to exposure to Lambda Cyhalothrin. The severity of alterations differ with the present study.

Sana Ullah et al (2019a) reported biomarker studies of synthetic pyrethroids and referred histopathological changes given as table 1.

Sangeetha et al (2018) in the fish Poecilia latipinna using 25% EC of cypermethrin reported on the histopathological lesions in the gill. The destruction of the secondary lamellae, necrosis, and club shaped secondary gill lamellae all as of the present study observations were reported.

Carlos et al (2018) reported in the fish Prochilodus lecreatus that are induced by lambda cyhalothrin. Histopathological variations that are observed are of severity nature as of the present study, with Cyfluthrin 10% WP. The toxic effect of the both the toxicants differ hence variations.

Sahoo et al (2017) in the fish Labeo rohita, using deltamethrin as the toxicant reported that the branchial arch was swollen and all of it included cells that are infiltered.

Chandrima Dey and Saha (2016) reported on the fish Labeo rohita by lambda cyhalothrin 5% EC as well as Marshal (carbofuran 8.5% EC). Notable changes were noticed in the gill, liver and kidney as in the present study and both of them belong to same group of the synthetic pyrethroid.

Asma Karim et al (2016) reported, on histological variations that are observed in the fish, Hypophthalmichthys molitrix after acute exposure to pyrethroid – deltamethrin. Changes observed were severe than the present toxicant.

Mohammad et al (2016) in their study using Deltamethrin as the toxicant in the fish Nile tilapia too observed changes as lesions, hyperplasia of the secondary lamellae and joining of them termed as fusion which are also the same in agreement of the present study.

Neelima et al (2015) reported in the fish Cyprinus carpio the exposure to cypermethrin 25% EC which caused changes in the gill tissues. They mentioned about the primary lamellae of necrosis, fusion of the secondary lamella clubbing and shortening wherein it had all the alterations that were also observed as the toxicant of the present study even though they belong to two different classes pyrethroids..

Singh et al (2015) using lambda cyhalothrin in the fish Clarias batracus, Velisek et al (2009a&b) using bifenthrin as the toxicant reported on the histopathological damages in the fish common carp Cyprinus carpio and Oncorhynchus mykiss.

Anitha Susan et al (2012) studied and reported in the fish of the three major carps Labeo rohita, Catla catla and Cirrhinus mrigala after exposure to the synthetic pyrethroid of type II, fenvalerate. The tissues they studied, include

gills also, a comparative study. The observations are in agreement of the present work even though the present tested toxicant belongs to the type I without a cyano group which imparts lesser toxicity. The Permethrin being adhered to the mud particles, (adsorption) had much effect on the bottom dwelling fish, such as Cirrhinus mrigala , including the Labeo rohita the present studied one due to water dwelling habit being teleost have five pairs of gills prone to have the damage by the toxicant tested that was reported for the fenvalerate and similar in the present work.

Velisek et al (2011), of the fish reported on the effects of the pyrethroid and triazine pesticides mentioned also about the histopathological damages that resulted after exposure to the cypermethrin, a synthetic type II pyrethroid, Bifenthrin and also the triazines. When cypermethrin was studied in the fish rainbow trout after exposure to 96h lethal concentration, caused a pathological conditions called 'Elamgiectasia' in the secondary lamellae of the gills and pillar /columnar cells of the compound epithelium. In the fish Cyprinus carpio hyperplacia of the respiratory epithelium of the chloride cells was also observed. Bifenthrin another toxicant as one of the triazines, severe Elamgiectasis (dialation of capillaries) was observed for the same fish.

Velisek et al (2007, 2009) observed and reported in the fish rainbow trout Onchorynchus mykiss, exposed to biferthrin, type I synthetic pyrethroid, the localized tissue damage and also the weakened cell walls known by the term aneurysm in secondary lamellae, hyperplasia of the epithelium and fusion of cells in the gills. This is also similar in line of the present observation and is also of similar of other studies of type II synthetic pyrethroids, (Velisek et al 2007 and also by Cengiz 2006a), in Cyprinus carpio after exposure to Deltamethrin).

Cyfluthrin induced histopathological alterations in the fish Cyprinus carpio (Aylin et al 2009) and some of the changes are similar of the present study.

Balu Velumurugan et al (2007) reported the study of the toxicant fenvalerate another synthetic pyrethroid, to the fish Cirrhinus mrigala. The study revealed that epithelial hyperplasia, necrosis of the epithelial surface desquamation and fusion of lamellar fusion hindering the diffusion pathway of the gases apart from curling. The present work even though it belongs to the different class of the synthetic pyrethroid is of similar line.

Diana et al (2007) opined that the damage of the chloride cells due to the exposure of the Deltamethrin at 2 µg/L concentration that was similar as in the present study and the resultant damage preclude the chloride cells to death, that make difficulty in the process excretion of the salts through the gills.

Yildrim et al (2006) study of the toxicant exposure of Deltamethrin to the fish Gambusia affinis for a long duration in sublethal concentration too revealed similar changes of fusion of the secondary lamellae of the present study. Observation as above and in addition were also reported as hyperemia and aneurysm by Cengiz, 2006b).

Thus the observations of the present study are of similar lines, but for present toxicant the studies are very few.

Discussion - Liver.

Liver with synthetic pyrethroids

Karim et al, (2016) the liver of silver carp Hypothalmichthys moltrix after acute exposure to pyrethroid deltamethrin. The study revealed that necrosis, nuclear pyknosis hypertrophy of hepatocytes vacuolization nuclear atrophy and congestion of blood vessels were noted in their study.

Any biochemical alterations in the organs of the fish Labeo rohita, liver metabolically active and kidney for excretion were due to intoxification of a synthetic pyrethroid deltamethrin the type II wherein histopathological changes finally manifested impact in the biochemical enzymes of tissues/organs was reported by Suvetha et al (2015).

In the Cyprinus carpio after cypermethrin exposure and the result was of the similar lines of the present study by Neelima et al (2015) reported. Andem et al, (2017), Velisek et al, (2006a) too reported on the toxicant cypermethrin in the fish Clarias gariepinus and Onchorynchus mykiss respectively and their observations are also of similar nature of the present study.

Anitha Susan et al (2012) too reported damage to the liver tissue when fenvalerate toxicant is tested on the three Indian major carps and it also coincides results of the present work.

Velisek et al (2007) reported in the fish Onchorynchus mykiss, due to exposure to deltamethrin that showed pathological variations in the largest gland of the body, the liver and is also in the organ of ammonotellic excretory component.

In liver tissue of the mosquito fish Gambusia affinis after exposure to commercial deltamethrin (EC), hypertrophy of hepatocytes the kupffer – (immune) cells of the disturbance in circulation necrosis and also narrowing of the sinusoids, all are reported by Cengiz and Unlu (2006b). Some of the present studied, changes are also observed in the fish experimented.

Sree Vani and Veeraiah (2014) reported the effect of cypermethrin on the histopathology of the freshwater fish Cirrhinus mrigala. The toxicant induced changes, that cytoplasm of the hepatocytes degeneration, formation of the vacuoles and the rupture in the blood vessels and the disappearance of the hepatocytic cell walls and also the disposition of the hepatic cords.

Yildrim et al (2005) reported in the fingerlings of Oreochromis niloticus L., popularly known as nile tilapia after exposing them to type II synthetic pyrethroid, deltamethrin wherein liver damage of hydrophobic nature as being observed..

In the fish Gambusia affinis, mosquito fish and the reports of them as lesions that are observed also the same of the present study [Cengiz and Unlu (2002 and Cengiz 2003)].

The present study reveals that the xenobiotic compounds effect the fish organs like liver and kidney and the fish in pesticides in the environment, when our target is for monitoring of pollution and this type of biomarker study of histopathology as a tool of approach is a good one. Metabolically active liver organ and excretory function of kidney is impaired even in sub lethal concentrations on the survival of the non-target organism. The formulations of the pesticides used in then applications of both agri & aqua products must be viewed seriously.

Kidney – Discussion

Sana Ullah (2022) in the fish Ctenopharyngodon idella reported on bifenthrin a multibiomarker study, in which due to oxidative stress resulted the damage of cells/tissues. Due to enhanced ROS (Reactive Oxygen Species) that resulted lipid peroxidation which had damage of the tissues/organs of the fish due to bifenthrin intoxication. Rapid opercular movements were resulted to in take more oxygen as a result of the damage of the epithelial tissues. The same aspect of the toxic action of bifenthrin visualized in the fish Clarias batracus by Saha et al (2021) and Saha and Saha (2020) and also in the fish Oreochromis mossambicus (Peters, 1952).

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Yildrim et al (2006) study of the toxicant exposure of Deltamethrin to the fish Gambusia affinis for a long duration in sublethal concentration too revealed similar changes of fusion of the secondary lamellae of the present study. Observation as above and in addition were also reported as hyperemia and aneurysm by Cengiz, 2006b).

Shalini Anand and Harender (2016) reported the pathological changes in the kidney fish of the fish Clarias batracus due to cypermethrin. Shrinkage of the glomerulus and degeneration were observed due to toxic action.

Neelima et al (2015) also reported in the fish Cyprinus carpio after exposure to the cypermethrin a commercial formulation in the kidney and present work coincides with their report.

Velumurugan et al (2009) also reported that the kidney of the fish was effected with Cypermethrin as the toxicant in the fish Clarias gariepinus. Much of the observations were similar to the present study. Anitha et al (2012) in the three major carps using fenvalerate a synthetic pyrethroid of type II and Sree Veni and Veeraiah (2014) in the fish Cirrhinus mrigala after the exposure to the cypermethrin of 10% reported the degenerative changes in the kidney.

Velisek et al (2009a&b) reported on histopathological changes that are observed in the fish Onchorhyncus mykiss, exposed to bifenthrin the present tested toxicant degenerative changes were observed.

In the fish, Cirrhinus mrigala, on exposure to two sublethal concentrations of lambda cyhalothrin of 0.3 and 0.6 ppb by Velumurugan et al (2007) reported necrosis, pyknotic nature of nuclei in the haemopoietic tissue, the hypertrophy of the cells in the epithelium of tubules, the its lumen was narrowed and the space in the Bowman's capsule and glomerulus was expanded. Except the severity of the situation was not that much in the present study as described by them.

Similarly, Cengiz (2006) reported in the fish Cyprinus carpio on exposure to deltamethrin of synthetic pyrethroid deltamethrin of 50% and 70% of 96h LC_{50} value. As observed by them that showed a regional damage and degeneration of the cells of the renal tubules, pyconotic nuclei in the haemopoitic tissue, dilation of glomerullar capillaries and intra cytoplsmic vacuoles in the epithelial cells apart from hypertrophied and narrowing of the part and the results were not observed in the present study.

Yildrim et al (2006), studied histopathological effects of deltamethrin and also apart from Staicu et al 2007 too reported in the fish Carassu sauradus using the same toxicant.

Table 1:- Histopathological-morphological toxicity.

S.No.	Fish species	Changes observed	References
1	Hypophthalmichthys molitrix	Deltamethrin exposure resulted in different histomorphological alterations in the brain. e.g. hemorrhage, infiltration. Neuronal degeneration. and spongiosis; liver. e.g. congestion. hernosiderosis. increased sinusoidal spaces. fibrosis. and pycnosis; gills. e.g. disruption of arch. epithelium. and lamellae. Atrophy of lamellae and desquamation of goblet cells; and intestine. e.g. shredding of mucosal cells, disruption of mucosal cells and intestinal mucosa and increased goblet cells	Ullah et al (2019)
2	Danio rerio	Deltamethrin induced different developmental malformations	Parlak (2018)
3	Calossoma macropomum	Deltamethrin induced different histopathological damages in the gills	Cunha et al (2018)
4	Danio rerio	Deltamethrin led to different morphological alterations such as shorter body length, larger head-body angle. and smaller eyes	Liu et al (2018)
5	Pangasianadon hypophthalmus	Cypermethrin exposure induced different changes in the histological architecture of gills and liver	Cunha et al (2018)
6	Tor putitara	Cypermethrin induced different histopathological damage to the brain, liver and gills	Ullah et al (2015)
7	Clarius batracus	λ-Cyhalothrin induced different histo-architectural damages in the testes. liver. and kidneys	Singh et al (2015)
8	Sparus aurata	Deltamethrin resulted in deleterious morphological	Guardiola et al

		changes in the liver	(2014)
9	Aphanius dispar	Deltamethrin exposure led to different histopathological changes in the gills such as secondary lamellae fusion, epithelium lifting (lamellar), vacuolization and desquamation	Al-Ghanbousi et al (2012)
10	Oreochromis niloticus	Deltamethrin mediated different histopathological lesions in the liver, e.g. necrosis. pycnosis, and hypertrophy and gills. e.g. epithelial lifting, hypertrophy of mucous cells. and hyperplasia	Kan et al (2012)
11	Oncorhynchus mykiss	Bifenthrin induced different histopathological alterations	Velisek et al (2009b)
12	Heteropneustes fossilis	Cypermethrin induced different histological alterations in the gonads, liver and gonadotrophic cells	Singh and Singh (2008)
13	Oreochromis niloticus	Deltamethrin exposure resulted in different histopathological lesions in the gills. e.g. fusion of secondary lamellae, gills, hyperemia and telangiectasis and liver. e.g. hydropic degenerations	Yildrim et al (2006)

Source: Sana Ullah Z. Li, Amin Zuberi, Md Zain UL, A. Mirza Md Faran and Asif Baig (2019a). Biomarkers of synthetic pyrethroid toxixcity fish. Environmental Chemistry letters 17: 945-973.

Plate-1
Histopathological Alterations in the Gill of the fish Catla catla
Exposed to CYFLUTHRIN 10% WP

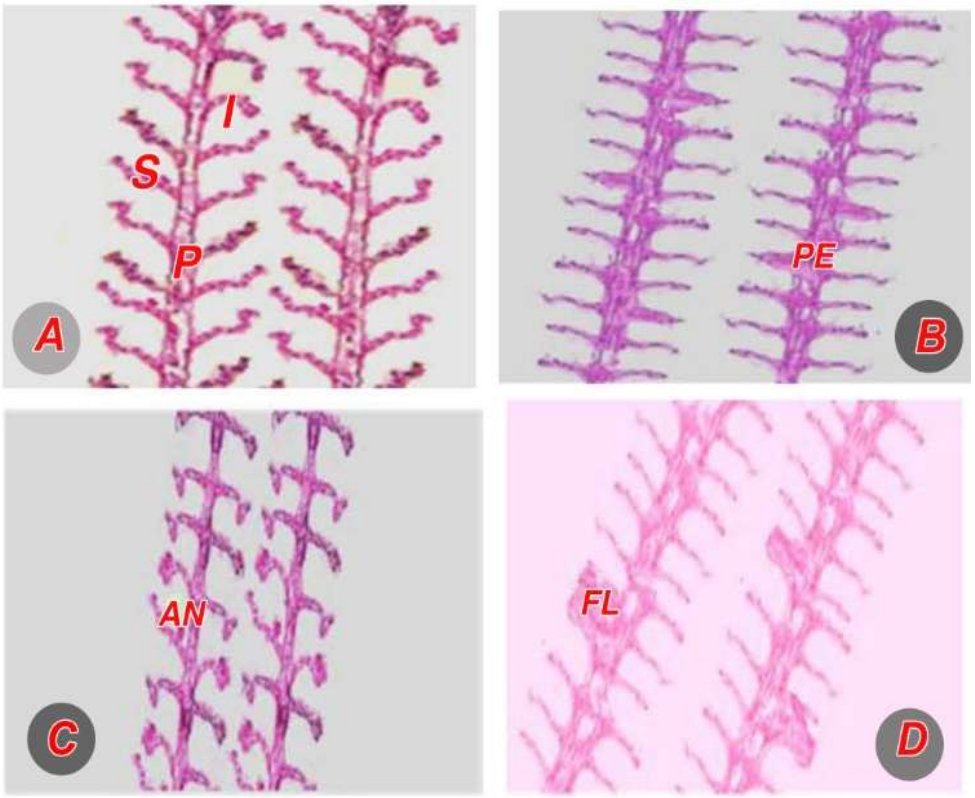

Fig.-A: Control Gill (S-Secondary Lamellae, P-Primary Lamellae, I - Interlamellar Space) (H&E 400x)
Fig,-B: Effect of CYFLUTHRIN 10% WP in sublethal concentration (PE-Proliferating Epithelium) (H&E 400x).
Fig.-C: Effect of CYFLUTHRIN 10% WP in sublethal concentration (AN-Aneurism) (H&E 400x).
Fig.-D: Effect of CYFLUTHRIN 10% WP in sublethal concentration (FL-Fused Lamellae) (H&E 400x).

Plate-2
Histopathological Alterations in the Liver of the fish Catla catla
Exposed to CYFLUTHRIN 10% WP

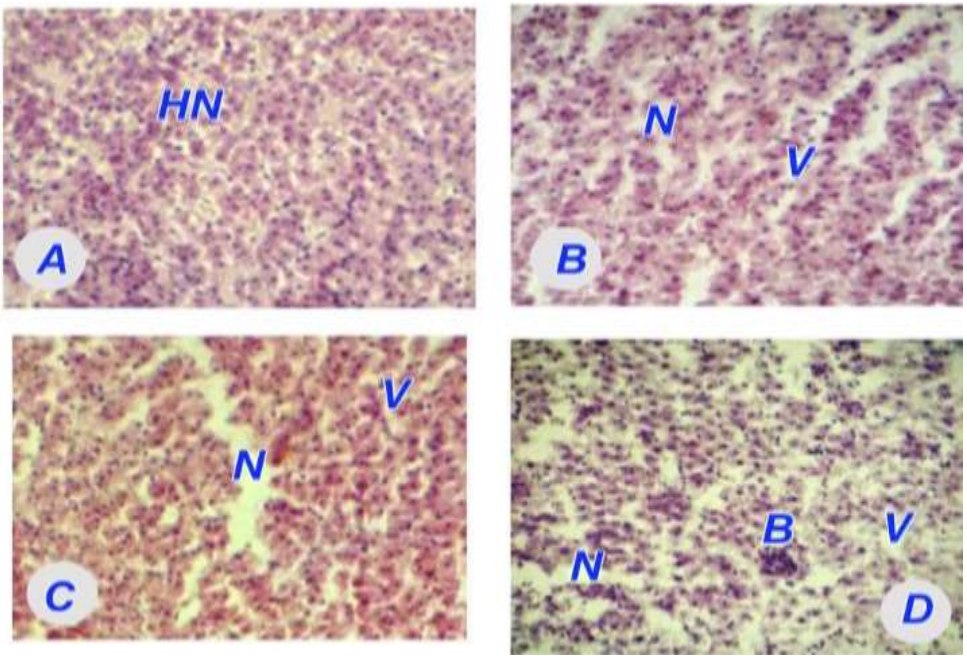

Fig.-A: Control Liver (HN-Hepatocyte with Nucleus) (H&E 400x)
Fig,-B: Exposed to CYFLUTHRIN 10% WP in sublethal concentration (N-Necrosis, V-Vacuoles)
Fig.-C: Exposed to CYFLUTHRIN 10% WP in sublethal concentration (N-Necrosis, V-Vacuoles) (H&E 400x).
Fig.-D: Exposed to CYFLUTHRIN 10% WP in sublethal Concentration (N-Necrosis, V-Vacuoles, B-Blood Sinusoids) (H&E 400x).

Plate-3
Histopathological Alterations in the Kidney of the fish Catla catla
Exposed to CYFLUTHRIN 10% WP

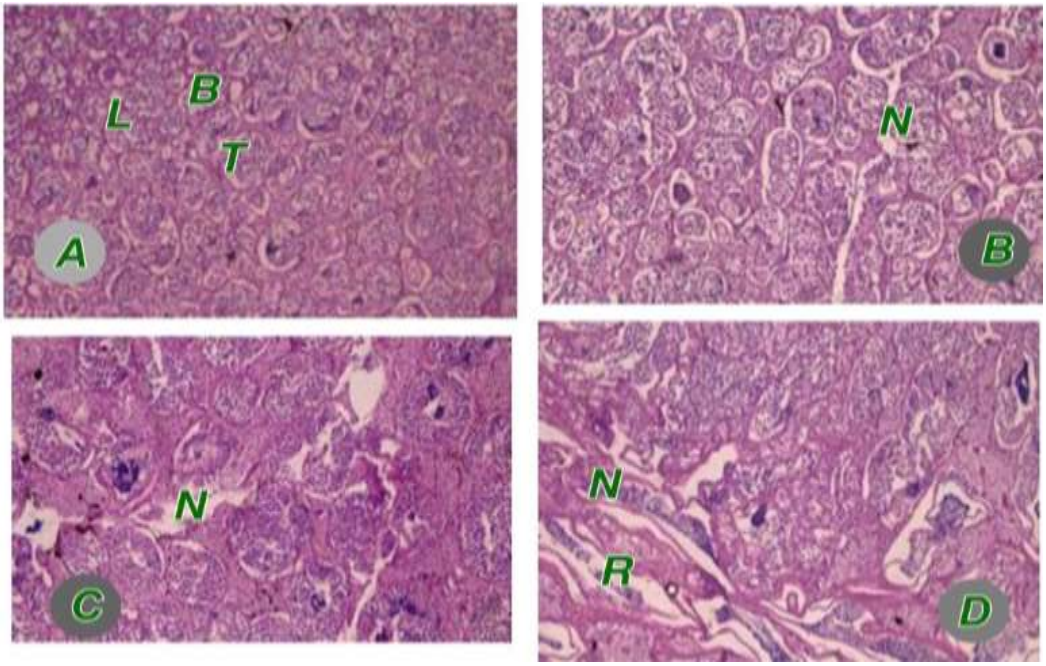

Fig.-A : Control Kidney (B-Bowman's Capsule, T-Tubule, L-Lumen) (H&E 400x).

Fig,-B : Exposed to CYFLUTHRIN 10% WP in sublethal concentration (N-Necrosis) (H&E 400x)
Fig.-C : Exposed to CYFLUTHRIN 10% WP in sublethal concentration (N-Necrosis) (H&E 400x)
Fig.-D: Exposed to CYFLUTHRIN 10% WP in sublethal Concentration (N-Necrosis, R-Reduction of Haematopoietic tube) (H&E 400x).

Conclusion:-

The important tissues gills, liver and kidney of the fish, can be termed as vital when exposed to the toxicants like Cyfluthrin 10% WP that showed pathological degenerative changes which culminate the biochemical changes and the very survival of the fish is questionable even in the sub-lethal concentrations and apprehend that sub lethal concentrations are really lethal to the fish. If any, representation of the toxicant is going to be given for its usage, the sublethal, chronic devastations that are going to manifested be thoroughly studied and the actual mechanisms of toxicant and such lesions needs to be understood. It is evident the work on bifenthrin on histopathological studies are very few.

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