# BIFENTHRIN INDUCED HISTOPATHOLOGICAL ALTERATIONS IN KIDNEY OF *CYPRINUS CARPIO*

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**ABSTRACT:** Bifenthrin is used as insecticide in agriculture. Present manuscript deals with the poisonous outcome of bifenthrin on histology of kidney of fish *Cyprinus carpio*. Freshwater fish *Cyprinus carpio* was used in this experimental study. An aggregate of 30 fish were arbitrarily separated into 3 gatherings A, B and C. Group A was kept as control while Group B and C were treated as exploratory. All fish were kept into glass aquaria at room temperature. During experiment, group A (control) was kept in normal water while bifenthrin was added in water of experimental groups B & C. The water of all groups was changed on daily basis. Kidney samples were collected after 48 & 96 hours exposure and processed for histopathological study. Behavioural pattern and histopathological changes of kidney of *Cyprinus carpio* were noted. Major changes, for example, Glomerular constriction, extreme degeneration of the tubules, hypertrophy, necrosis and pyknosis were observe under the microscope. Thus, bifenthrin as pesticide should be used carefully to protect the health of aquatic food animals.

**Keywords:** Bifenthrin, toxicity, Cyprinus *carpio*, Kidney.

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## INTRODUCTION

The FAO (Food and Agriculture Organization of the United Nations) describes pesticide as any element or combination of elements of biological or natural ingredients intended for preventing, abolishing or monitoring any pest, or regulating plant growth (FAO, 2020). Public and manufacturing wastes rich in pesticides also cause water pollution. Maximum insecticides eventually discover their approach into rivers, lakes and ponds (Lackmann *et al.,* 2019) and have been found to be extremely lethal to non-target animals that live in natural atmospheres close to cultivated lands, foremost to death of fish (Allen *et al.,* 2019).

A pyrethroid insecticide used against red imported fire ant which affects its nervous system is known as bifenthrin. It is highly toxic for aquatic animals. The bifenthrin is white waxy solid with a pale sweet odour and easily soluble in water. It is chemically manufactured in several arrangements such as powder, granules and pellets. It is not found naturally (Awoyemi, 2019). It is used as a contact insecticide and acaricide on diversity of crops, on stored grain and as a preconstruction termite obstruction (Bai *et al.,* 2019). Bifenthrin is a type 1 pyrethroid (Souza *et al.,* 2019). Almost 70% of all hops and rasberries cultivated in the United States are treated with bifenthrin (Perry *et al.,* 2019). It is used in textile industry to protect woollen products from insect attack. In aquaculture, pyrethroids are used to control some parasitic infections (Bhatt, 2019).

Histopathological examination is a valuable tool to evaluate the influence of ecological contaminant on fishes and toxicological studies (Georgescu *et al.,* 2017). Fish showed biochemical and histopathological alterations in different target tissues such as gills, liver and kidney (Hassan *et al.,* 2018). The exposure to chemical contaminants can induce a number of wounds and damages to different fish organs (Mustafa *et al.,* 2019) but gills, liver and kidney represent important target organs suitable for histological examination to study damages to tissues and cells (Al-Samawi *et al.,* 2017). The present study was conducted to investigate toxic effects of bifenthrin on kidney of common carp (*Cyprinus carpio*) with the help of histopathological examination.

## MATERIALS AND METHODS

A total of 30 fish (12.13±0.04 gms, 7.9±0.04 cm) were obtained alive Manawan Hatchery, Lahore and maintained in aquaria for a period of 7 days to acclimatize in the new environment before the experimental study. The fish were casually distributed into 3 groups A, B, and C. They were fed on

commercial feed two times daily. Group B and C were exposed to 0.58mg / L and 1.16 mg / L concentrations of bifenthrin respectively for 96 hours while group A was kept as control. Death rate was noted after 24, 48, 72, and 96 hours exposure to bifenthrin. Each group was kept in a 65-litre capacity glass aquarium containing 45 litre of water treated or untreated. The Each aquarium was cleaned and filled with fresh tap water 2 times per day. The experiment continued for 8 days to determine the effect bifenthrin on kidney of fish. The fish were anaesthetized with 40% ethyl alcohol before sampling. Kidney samples were collected following execution and fixed in 10% formalin. The immovable tissues were processed to prepare slides. The slides were stained using hematoxylene and eosin staining technique as described by Drury and Wallington et al., 1980. The stained slides were examined under oil immersion lens to record the histopathological changes.

## RESULTS

The bifenthrin induced behavioural and histopathological changes in fish of group B and C (Table.1 and Table 2). The results of present study indicated different toxicity level in tissue histology with increase in dose and duration of toxicant. The progressive degenerative changes in kidney histology increased with an increase in dose and exposure time period.

**Group – A (Control group):** The control group showed a normal architecture of kidney that comprises of many functional units called nephrons. Each nephron comprises renal corpuscle and a renal tubule. The renal corpuscle contains glomerulus and Bowman's capsule. The renal tubule contains proximal, distal and accumulating tubules (Figure 1).

**Experimental Group:** In group B and C kidney of fish shows histopathological changes such as glomerulus constriction, hypertrophy, necrosis and pyknosis were produced after exposure to bifenthrin for 8 days.

**Group – B (Low dose):** After 48 hours pyknosis, the earlier stage of necrosis which is defined as the degeneration of the cell was derived (Figure 2 and 3). In this condition, the cells nuclei appeared to be shrunken and deeply stained. Necrosis was seen in both epithelial and hematopoietic tissue. Localized disorganization and dilation of renal tubules was also observed after 96h (Figure 4 and 5).

**Group – C (High dose):** 1.99µg/L dose of bifenthrin produced severe changes in kidney histology. 80 to 90% glomeruli and renal tubules were disorganized after 48-hour exposure. The entire tissue was disintegrated and separated into patches. Hypertrophy which is defined as the enlargement of an organ or tissue from the increased in size of its cells, and vacuolization were also observed (Figure 6).

After 96 hour's focal necrosis and nuclear pyknosis were sharply increased and regular shape of tubules lost due to cytoplasm precipitation and karyolysis. The renal corpuscle was dilated and degenerated. The proximal and distal tubules were degenerated and tubular lumen is remarkably widend. Degenerative changes in tubular epithelium are enormous and the epithelium is desquamated in some tubules. Hydropic swelling and vacuolization are seen in most tubules. Nuclei show erosion, and deteriorating necrotic regions turn out to be visible and glomeruli show obvious reduction. (Figure 7).

**Table 1. Behavioral changes *Cyprinus carpio* during exposure time to bifenthrin.**

| Behavioral and physical changes | Concentration of bifenthrin | | |
|---|---|---|---|
| | 0.00 µg/L | 0.58 µg/L | 1.16 µg/L |
| Lack of stability | - | - | ++ |
| Level of spinning | - | - | +++ |
| Rate of opercular movement | - | + | ++ |
| Restlessness | - | + | ++ |
| Bottom to surface movement | - | - | ++ |
| Resting at bottom | - | + | ++ |

Foot note: Nope alteration (-), Slight alteration (+), Adequate alteration (++), Stark alteration (+++).

**Table 2. Histopathological variations in Kidney of *Cyprinus carpio* after 96 hour's exposure to bifenthrin during experimentation.**

| Histopathological changes in fish | Experimental groups with concentrations | | | | |
|---|---|---|---|---|---|
| | 0.00 µg/l control | 0.58 µg/l Low dose | | 1.16 µg/l high dose | |
| | | After 48h | After 96h | After 48h | After 96 |
| Enlargement of renal tubule | - | + | ++ | ++ | +++ |
| Pyknotic nuclei in the hematopoietic tissue | - | + | ++ | ++ | +++ |
| Degenerated glomerulus | - | - | -- | ++ | +++ |
| Contraction of the tubular lumen | - | + | ++ | +++ | +++ |
| Necrosis and degeneration | - | + | ++ | +++ | +++ |
| Focal necrosis | - | - | -- | ++ | +++ |
| Disorganization of renal tubules | - | + | ++ | +++ | +++ |
| Shrinkage of glomerulus | - | - | - | ++ | +++ |

Foot note: no change shown by (-), Mild changes (+), Moderate changes (++), Severe change (+++).

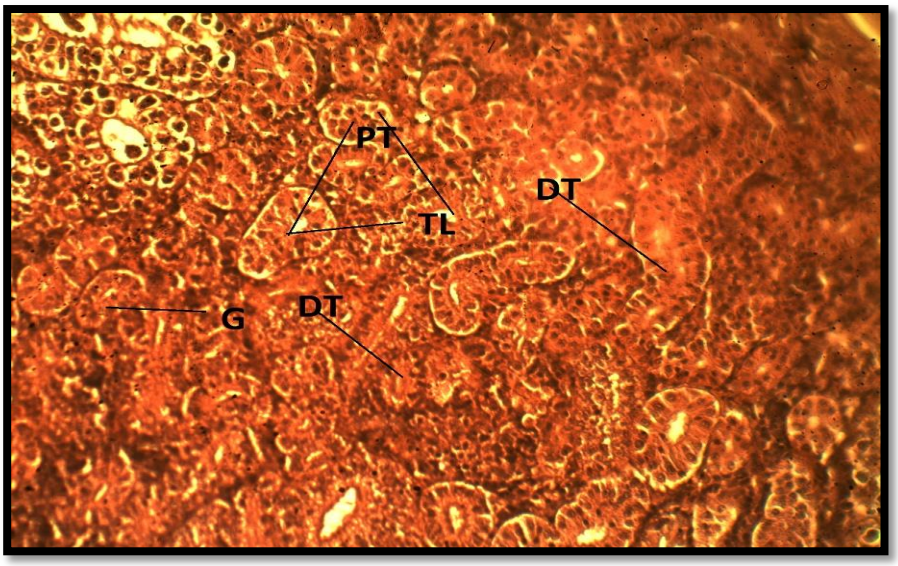


**Figure 1: Group A (control) showing normal histology. Glomerulus (G), normal hematopoietic tissues (H), tubules (T). (H &E) 40X.**

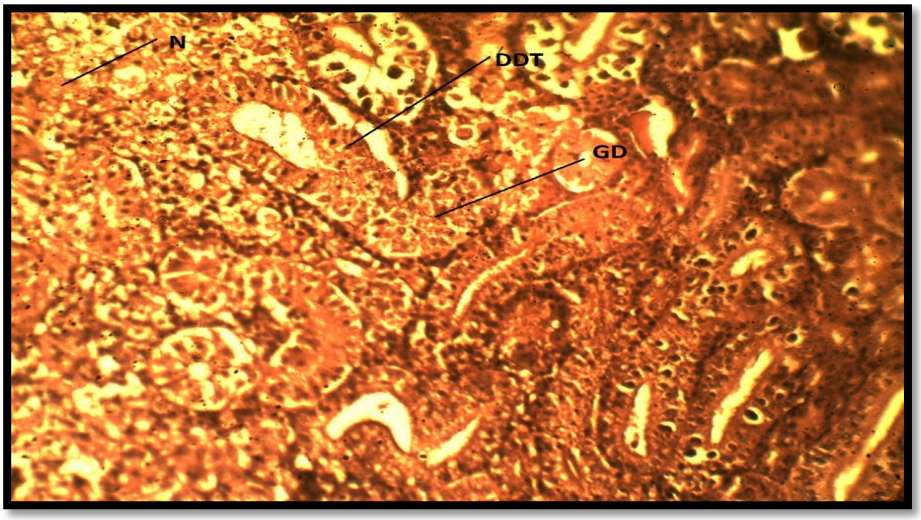


**Figure 2: Group B (low dose) after 48hours, showing degeneration of glomerulus (DG), degeneration of distilled tubule (DDT), necrosis (N). (H & E). 40X.**

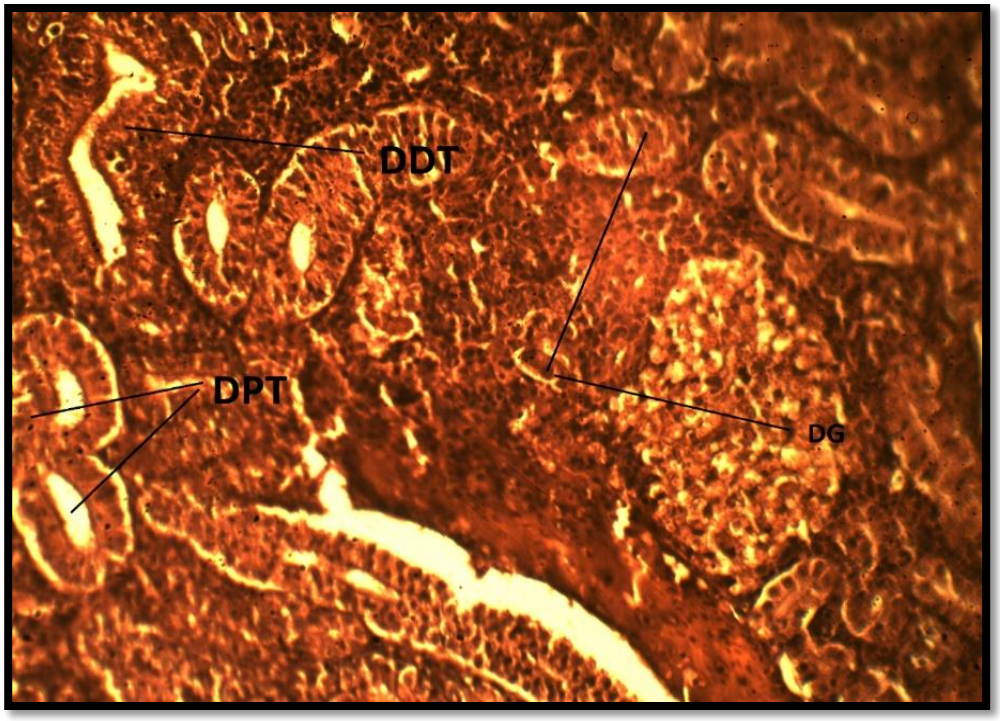


**Figure 3: Group B (low dose) after 48hours, showing dilation of distilled tubule (DDT), degeneration of Glomerulus (DG), Dilation of proximal tubule (DPT). (H & E).**

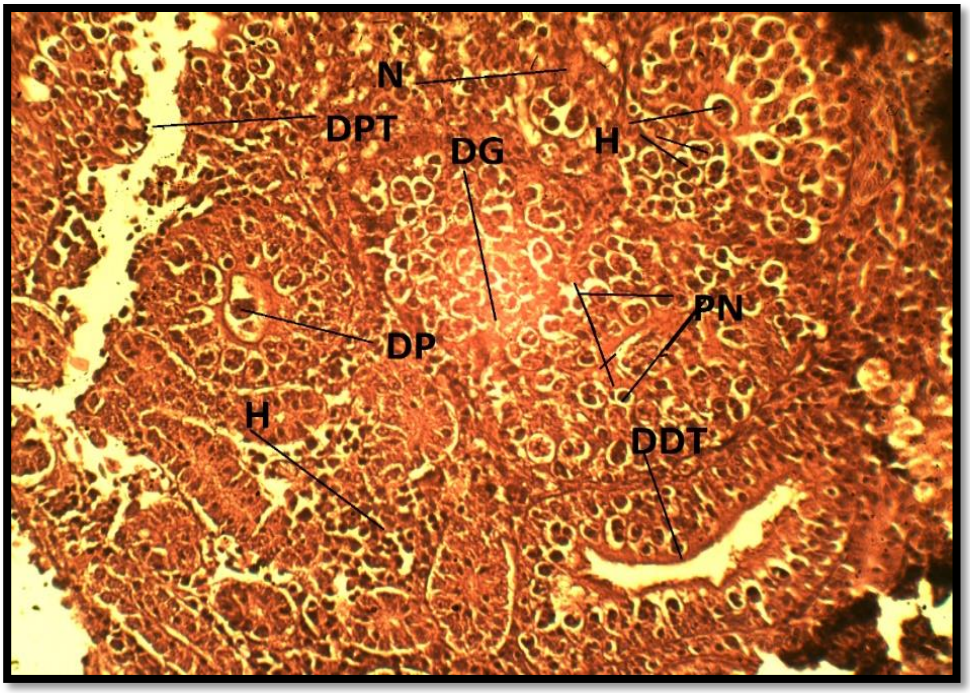


**Figure 4: Group B (low dose). After 96 hours, showing dilation of distilled tubule (DDT), degeneration of hematopoietic tissue (DHT), degeneration of glomerulus (DG), necrosis (N), hypertrophy (H), Pyknotic nuclei (PN), degeneration of proximal tubule (DPT). (H & E). 40X**

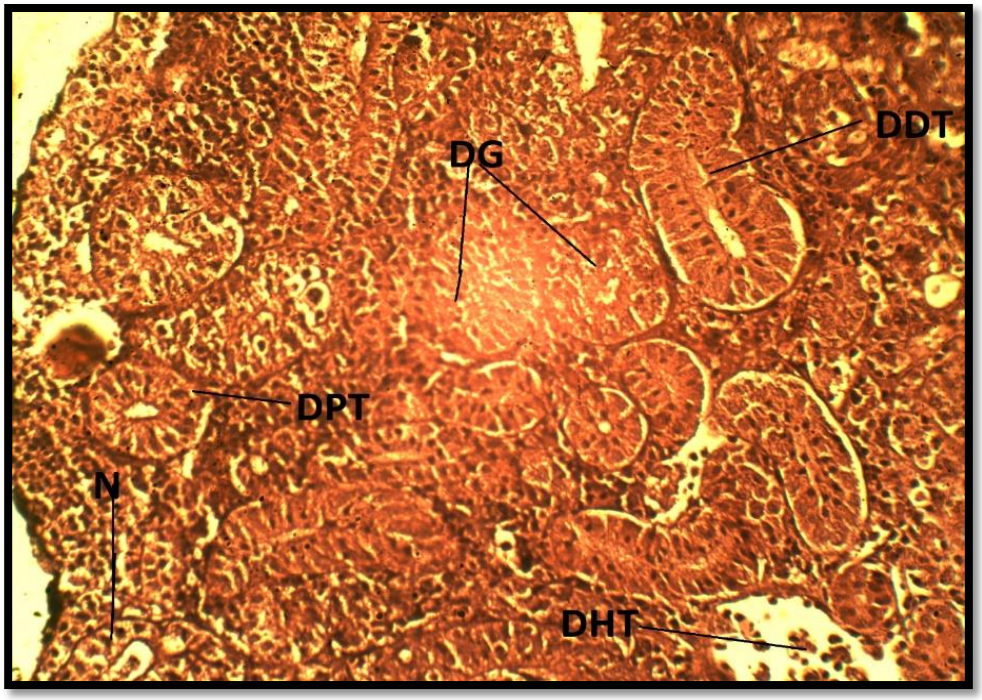


**Figure 5: Group B (low dose). After 96 hours, showing dilation of distilled tubule (DDT), depletion of hematopoietic tissue (DHT), degeneration of glomerulus (DG), necrosis (N), dilation of proximal tubule (DPT). (H & E). 40X.**

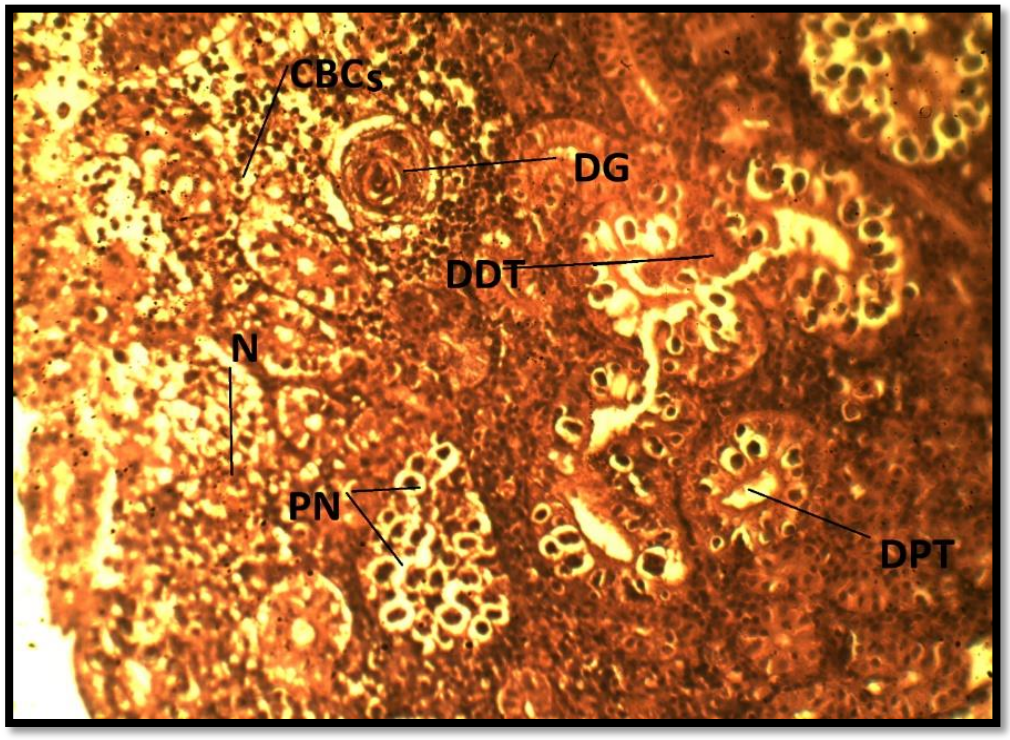


**Figure 6: Group C (high dose) after 48 hours, showing degeneration of glomerulus (DG), degeneration of proximal tubule (DPT), necrosis (N), depletion of distilled tubule (DDT), congestion of blood cells (CBCs), Pyknotic nuclei (PN). (H & E). 40X.**

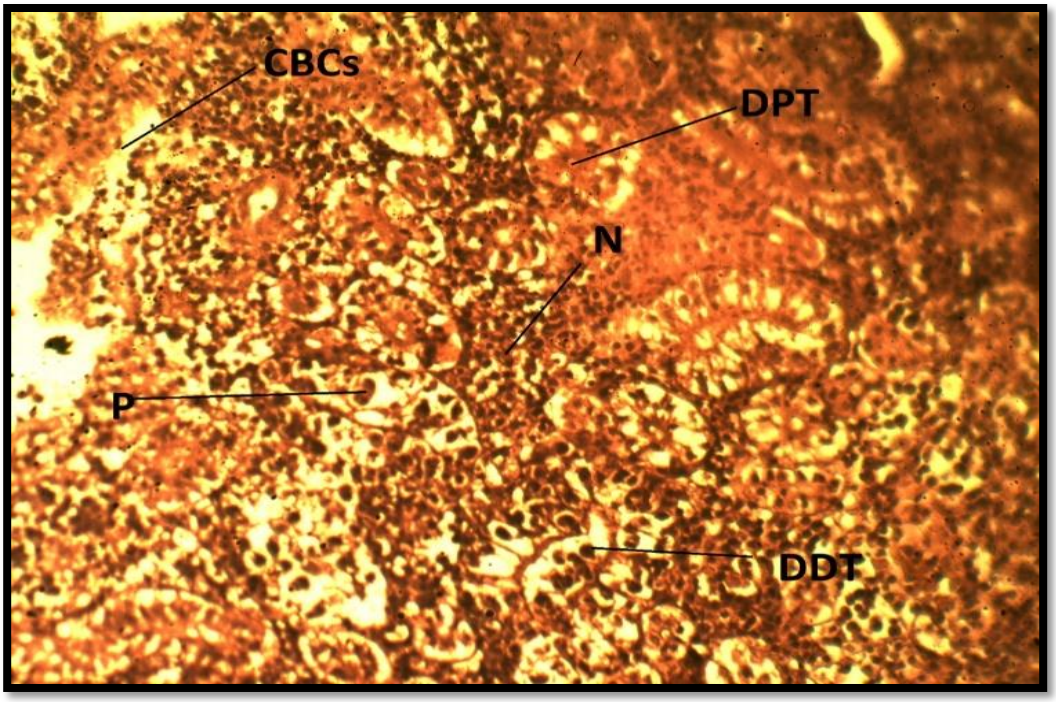


**Figure 7: Group C (high dose). After 96 hours, showing depletion of distilled tubule (DDT), disorganization of proximal tubule (DPT), necrosis (N), congestion of blood cells (CBCs), Pyknosis (P). (H & E). 40X. (H & E). 40X.**

## DISCUSSION

Miniscule examination of normal tissue to watch the presence of sickly cells and tissues is perceived as histopathology. Practically like our investigation, Chapdense *et al.,* 2009 depicted the behavioural changes all through exposure period and assessed the lethal impacts of atrazine on fish tambaqui.

They noted unpredictable conduct like torpidity, loss of balance, increment inside the recurrence of opercular developments, and increment inside the width of the mediocre lips following 48 hours of demonstration to atrazine at the scope of 20-25 mg/L. Furthermore, they examined the existence of haemorrhages inside the eyes, lips, or even the entire form. Comparative advancements associated through clumsy conduct were additionally perceived in aquatic fish *Channa punctatus* following 96 hours introduction to sub-lethal concentrations of bifenthrin at the scope of 4.2-10.6mg/L (Nwani *et al.*, 2010).

At the crucial divulgence through this examination, fish were changed and left swimming; a little while later, they attempted to stay away from the poisonous impacts of bifenthrin with quick spinning and hopping and at the most noteworthy of trial; fish lost their equilibrium and got depleted and torpid. They likewise persistently released gigantic measures of bodily fluid i-e mucus from entire body constantly and in this way the body pigmentation was reduced.

Velmurugan *et al*., (2007) revealed pyknotic cores in cylindrical epithelium, hypertrophy of epithelial cells of renal tubules, reduction of the glomerulus and development of space inside the Bowman's container within the kidney of *Cirrhinus mrigala* presented to monocrotophos. Gill *et al.,* (1989) illustrated different histopathological variations like decay of cylindrical epithelium, atomic decrease like karyorrhexis and karyolysis, and deteriorating glomeruli inside the kidney of *Puntius conchonius* succeeding contact to cadmium. They additionally discovered reformist increment in seriousness of degenerative changes with expanding length of disclosure.

The modifications found in the kidney of fish in the current investigation were enlargement of refined tubule (DDT), degeneration of hematopoietic tissue (DHT), degeneration of glomerulus (DG), Necrosis (N), hypertrophy (H), Pyknotic cores (PN), degeneration of proximal tubule (DPT). The kidney of the fish is a blended organ involving hematopoietic, reticuloendothelial, endocrine and excretory component.

The kidney of fish is normally situated in retroperitoneal position facing the ventral part of the vertebral column. It is a light or dark brown coloured or black organ ordinarily broadening the length of the

body cavity. Seriousness in modifications extended in relation to extended dosage. Cengiz (2006) accounted Comparative outcomes in *Cyprinus carpio* after intense presentation to deltamethrin. Comparable modifications were found in fishes accessible to natural contaminants (Veiga *et al.,* 2002). Most regular modifications found in the kidney of fishes presented to water pollution are tubule deterioration (overcast enlargement plus hyaline precipitations) and modifications in the corpuscle, for example, enlargement of vessels in the glomerulus and decrease of Bowman's space (Takashima and Hibiya, 1995). Necrosis of cylindrical and encompassing hematopoietic cells, pyknosis and karyorrhexis of kidney tissue were seen when Coho salmon was accessible to Amitrole for 144 hour (Rand and Petrocelli, 1985).

**Conclusion:** It is concluded that bifenthrin is toxic to fish and calls for its cautious use as a pesticide to prevent water contamination for saving the lifetime of fish and other aquatic animals. It affects fish wellbeing by causing harmful changes in the kidney. Thus the farmers should be informed about harmful effects of these substances before their use.

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