

Research Article**Impact of heptachlor on haematological and histopathological indices of fish *Catla catla*****B. Kiran Kumar, D. Vineela, S. Janardana Reddy***

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Abstract

Background: It is well known that pesticides have become an important tool of modern agriculture to protect standing crops, stored grains and human belongings from pests and also help in preventing diseases. As a whole or in residual form, these pesticides make their entry into the aquatic ecosystem and pose a serious threat to the aquatic organisms in general and fishes in particular. The LC50 of Heptachlor was identified as 1.60 mg/L for 96h. **Objective:** The present investigation was undertaken to evaluate the effect of sublethal concentration of heptachlor on haematological and histopathological indices of *Catla catla*. **Material and Methods:** The fish were exposed to 3, 7, 15, 30 and 45 days and observed the significant modulations in haematological and histopathological indices in fresh water fish, *Catla catla* for 45 days exposure period. Modulations are discussed and highlighted with advanced literature. **Results:** In the present study LC50 of Heptachlor was determined as 1.60mg/L for 96 hours. The present study revealed that the sublethal concentration of heptachlor caused variations in haematological indices and Histomorpho and structural integrity of liver and kidney tissues of *Catla catla* for 3,7,15,30 and 45 days exposure periods. The RBC was significantly lower at for 45 days (2.67 ± 0.21) when compared to control (4.32 ± 0.14) and other treated groups. The maximum decrease in Hb% was recorded at 45 days (7.11 ± 0.17) and higher value is observed in control group (11.86 ± 0.24). The PCV % was recorded the maximum decrease was observed on day 45 (16.11 ± 0.11) compared to control group (27.48 ± 1.18). The sublethal concentration of heptachlor caused drastic histomorphological variations in Liver and Kidney tissues of *Catla catla*. The Melanomacrophage Centers in Liver and kidney tissues are also significantly varied. Melanomacrophage Centers (MMC) also play an important role in the fish response to foreign materials including infection causing agents. **Conclusion:** It is obvious that the sub lethal concentration of heptachlor caused significant variations in haematological indices and irreversible modulations in histoarchitecture of liver and kidney of fish.

Keywords: *Catla catla*, haematology, heptachlor, histopathology, melano macrophage centres

Introduction

Many new pesticides introduced into the market every year to combat increasing in pest resistance. These pesticides persist in the soil, water and food, with toxicity related outcomes to both humans and animals (Ntow et al., 2008; Kumari, 2008). Modern agricultural practices result in indiscriminate use of various agrochemicals which usually enters into the aquatic environment. These agrochemicals affect the non-target aquatic biota including fish (Omitoyin et al., 2006). Injudicious and indiscriminate use of agrochemicals have caused great concern among health and environmental scientists because records of

filed application of pesticides even in developed countries revealed that less than 0.1% of pesticides applied to crops reach target pest, thus over 99% moves into ecosystem to contaminate the land, water and air (Chapman et al., 1998).

These pesticides can reach natural water either via transfer of the chemicals from soil (or) directly by spraying against target organisms. Aquaculture apart from agriculture is common in India. The non-target organisms are directly exposed to pesticides used for the control of insects and pests. The pesticides affect the survival growth rate, fecundity and reproductive activity of fish (Singh and Singh, 2006).

Catla catla is the Indian major carp is an economically important South Asian freshwater fish in the carp family Cyprinidae. It is native to rivers and lakes in Northern India, Nepal, Myanmar, Bangladesh, and Pakistan, but has also been introduced elsewhere in South Asia and is commonly

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farmed. *Catla catla* is a fish with large and broad head, a large protruding lower jaw, and upturned mouth. It has large, greyish scales on its dorsal side and whitish on its belly. It reaches up to 182 cm (6.0 ft) in length and 38.6 kg (85 lb) in weight.

Catla catla is a surface and mid water feeder. Adults feed on zooplankton using large gill rakers, but young ones on both zooplankton and phytoplankton. *Catla catla* attains sexual maturity at an average age of two years and attains an average weight of 2 kg.

Heptachlor is an organochlorine (cyclodiene) insecticide which was first isolated from technical chlordane in 1946. Heptachlor epoxide is a man-made compound that looks like a white powder. Heptachlor enters in to the aquatic environment cause serious threatening to various aquatic organisms and also cause severe metabolic abnormalities in non-target species like fish and freshwater mussels. Heptachlor epoxide degrades more slowly and, as a result, is more persistent than heptachlor (Black et al., 2000; Firouzbakhsh et al., 2014).

Heptachlor can enter the body through various sources and Heptachlor is readily converted into its oxygenated metabolite, Heptachlor epoxide. Once it is converted, the Heptachlor epoxide works to block the GABA_A receptors, causing an inhibition of the chloride ion flux through the GABA_A receptor and overstimulation of the nervous system (Caudle et al., 2005). Heptachlor and heptachlor epoxide are readily stored in fatty, liver, and kidney tissues in mammals. Heptachlor also passes the placenta and is found in breast milk. Animals exposed to heptachlor epoxide during gestation and infancy are found to have changes in nervous system and immune function. Higher doses of Heptachlor when exposed to newborn animals caused decrease in body weight and death.

Since a major part of worlds food is being supplied from fish source, so it is essential to secure the health of fishes for sustainable food source (Tripathi and Harsha, 2002). Among biological changes, Haematological parameters are considered potential bio markers of exposure to chemical agents, since the latter can induce an increase or decrease in the various haematological components (Railo and Nikinmaa, 1985). Haematological indices such as RBC, HB and PCV analysis can provide valuable knowledge for monitoring the health and condition of the both wild and cluttered fish. Haematological indices changes depend of the fish species age the cycle of sexual maturity and health condition (Blaxhall, 1972). The present study is aimed to study the effect of sub lethal concentration of heptachlor on haematological indices and histopathological modulations in freshwater fish, *Catla catla* for a period of 45 days.

Materials and methods

Experimental Chemical

The experimental chemical Heptachlor an Organochlorine

pesticide purchased from Kisan Agrochemicals Ananthapur district, Andhra Pradesh, India.

Experiment fish collection and maintenance

Live and healthy species of *Catla catla*, with age of 2 – 3 months old, were collected from Andhra Pradesh Government fish Breeding and Hatchery center near to Tirupati, Chittoor district and immediately transported them through polypropylene tank of 500 Ltr. capacity filled with well aerated and dechlorinated bore well water.

These collected fish species were acclimatized to laboratory conditions for 15 days. In large cement tank of 500L capacity. During this period of acclimatization bore well water free from chlorine and well aerated. Analysis of physico-chemical properties of water was followed APHA (1998) method, (Temp 27±1°C PH 6.8±0.05) at 27°C, and DO 6.9 -7.4 mg/l).

During the maintenance period the fishes were fed five times a day viz, 6 am, 9 am, 12 noon, 3 pm and 8 pm by commercially formulated pelletized feed (contain 35% protein) (Janardanareddy et al., 2016). Before 24 hours of the commencement of experiment the animals were harvested. The water was renewed at every 48 hours.

Determination LC₅₀ value of Heptachlor

LC₅₀ value of heptachlor for 96hr is calculated by the static bioassay method (Finney, 1971). In this method acclimated fish period were grouped into five batches of each contains 20 animals and they were transfer into 50L tanks filled with 40L of dechlorinated and aerated water and add different concentrations of Heptachlor in 5 tanks and simultaneously a control group was also maintained without (0 mg) Heptachlor for 96hrs. Water with same concentration of Heptachlor renewed at every 24 hrs (Table 1).

The lethal concentration of heptachlor (LC₅₀/96 hr) was identified as 1.60 mg/L (Figures 1 & 2) and 1/5th concentration (0.32 mg/L) was selected as sublethal concentration for further analysis.

Sublethal Studies

120 healthy fishes from acclimatized were selected randomly and divided into 2 groups, one experimental and second controlled group with each aquarium contain 40 species. Experiments were conducted for 45 days with respect intervals. 1/5th of LC₅₀/96 days of heptachlor = 0.32 mg/L) and another aquarium maintained as control without heptachlor for 45 days.

At the end of 3, 7, 15, 30 and 45 days experimental periods fishes and also control fishes were randomly selected sacrificed and used for haematological and

histopathological analysis.

Haematological studies

Blood sample collection and preservation

The Blood samples were collected from control and *Heptachlor* treated fishes by cardiac vein puncture using non – Heparinized syringes and expelled into heparinised plastic vials and they were stored at 4 – 5°C for subsequent hematological analysis (Shah and Altindag, 2005). Heparin sodium (1%) was used as an anticoagulant (Svobodova et al., 2008).

Total RBC

The total RBC count was determined by an improved neubaur haemocytometer (Shah and Altindag, 2004). To determine RBC Blood was diluted 1: 200 with haym's diluting fluid. The erythrocytes and leucocytes i.e total R.B.C were reported as 10^6 1mm^3 (Wintrobe, 1974).

Estimation of hemoglobin

The hemoglobin (Hb) measurement was determined by the cyanmethaemoglobin method (Wintrobe, 1974).

Determination of PCV

PCV was determined by Wintrobe's method (2000 rpm/hr).

Total WBC

Total WBC cells are counted using an improved neubaur haemocytometer (Shah and Altindag, 2004). To count total WBC of Blood was diluted to 1:20 with “turksdiluting” fluid and placed in haemocytometer. The total no. of WBC count is reported as $10^3/\text{mm}^3$ (Wintrobe, 1974).

Sample collection for histopathological studies

The fish *Catla catla*, 25±5g weight, were exposed to 1/5th sub lethal concentration of heptachlor, an organochlorine insecticide for 3,7,15,30 and 45 days were sacrificed and liver

and anterior kidney tissue was quickly isolated and washed in fish ringer solution, of which pH is about to equal. Histopathology of the tissues was studied by the method of Clayden (1962).

The physiological saline solution (0.75% NaCl) was used to rinse and clean the tissues. They were fixed in aqueous Bouins fluid for 48 hours, processed through different series of alcohol, cleared in xylene and embedded in paraffin wax. Sections were cut at μ thick, stained with Ehrlich hematoxylin eosin, dissolved in 70% alcohol and were mounted in Canada balsam. Tissue damage at cellular level caused by the heptachlor is examined and the change in the individual cells are visualized to ultimately arrive at a conclusive diagnosis by employing microscopic examination of tissue, in which the tissue is sectioned to single cell thickness and stained to differentiate the individual tissue elements. The tissues are then transferred to the block marker. The tissue are embedded in paraffin was (58-60°C) blocks. Sections were cut of 6-8 μ thickness stained with Haematoxylin – Eosin (dissolved in 70% alcohol) (Humason, 1972). The sections of tissues were observed under microscope and the conditions in different tissues were photographed at lower and higher power of magnification using Nikon micro photographic equipment.

Results and discussion

The hematological parameters constitute a good indicator of physiological response (Blakhall, 1972). A significant decrease in RBC, Hb concentration and packed cell value (PCV) has been observed earlier in fishes exposed to different pesticides. The hematological parameters are also considered as potential bio markers of exposure to agrochemicals due to their sensitivity to certain toxic agents (Heath, 1995).

Table 1. Effect of different concentrations of Heptachlor on the mortality of the fresh water fish *Catla catla*.

Concentration of Heptachlor (mg/l)	Log Concentration	No. of animals exposed	No. of animals died	Percent mortality	Probit mortality
0.075	-1.125	20	02	10	3.96
0.162	-0.791	20	05	15	4.55
0.222	-0.654	20	10	30	5.13
0.270	-0.569	20	12	50	5.30
0.411	-0.386	20	14	70	5.67
0.501	-0.300	20	16	80	6.04
0.512	-0.291	20	20	100	7.59
0.535	-0.272	20	20	100	7.59

Ralio et al. (1985) reported that the blood parameters of diagnostic importance are erythrocyte and leucocytes counts, haemoglobin and leucocyte differential counts. Haematological study is important for toxicological research, environmental monitoring of fish and their health conditions during culture because fish generally are so intimately associated with the aquatic environment. Fish in close association with their aquatic environment and any changes in this environment would be reflected in alterations in their haematological studies (Suzana Golemi et al., 2012).

The findings of the present investigation also reveal a similar decreasing trend in all the parameters such as RBC, Hb content and PCV% suggesting that the heptachlor induce changes which give evidence for decrease haematopoiesis followed by anemia induction in test fishes (Park et al., 2004). The decreased erythrocyte

count and haemoglobin content observed in this study may be due to the disruptive action on the erythropoietic tissue, which in turn affected the cell viability.

The haematological parameter of *Catla catla* fed with heptachlor for the period of 45 days was shown in Table 1. The blood samples were collected at 0 (Control), 3,7,15, 30 and 45 day's intervals during the experimental period. The Red blood cells count was significantly lower at for 45 days (2.67 ± 0.21) when compared to control (4.32 ± 0.14) and other treated groups. The maximum decrease in Hb% was recorded at 45 days (7.11 ± 0.17) and higher value is observed in control group (11.86 ± 0.24). The PCV % was recorded the maximum decrease was observed on day 45 (16.11 ± 0.11) compared to control group (27.48 ± 1.18) (Table 2).

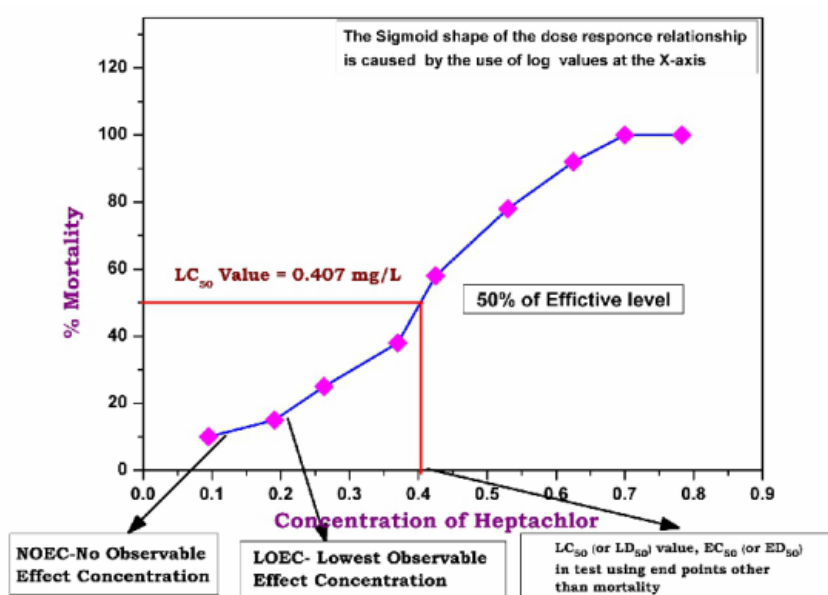


Figure 1. Representing percent mortality Vs log concentration of Heptachlor

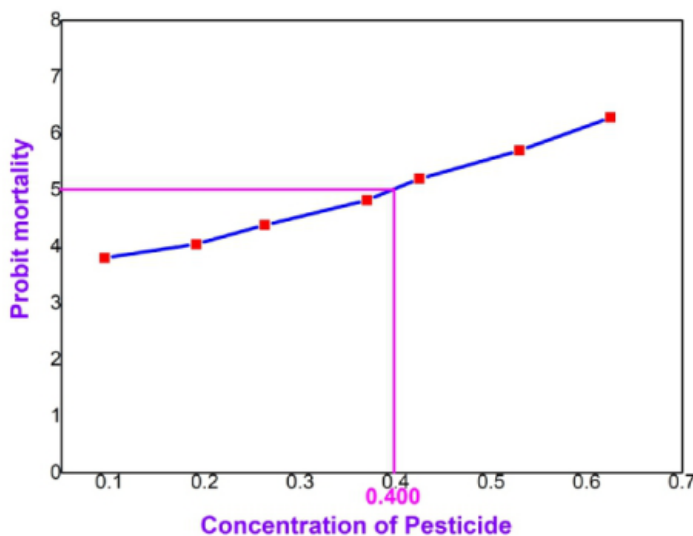
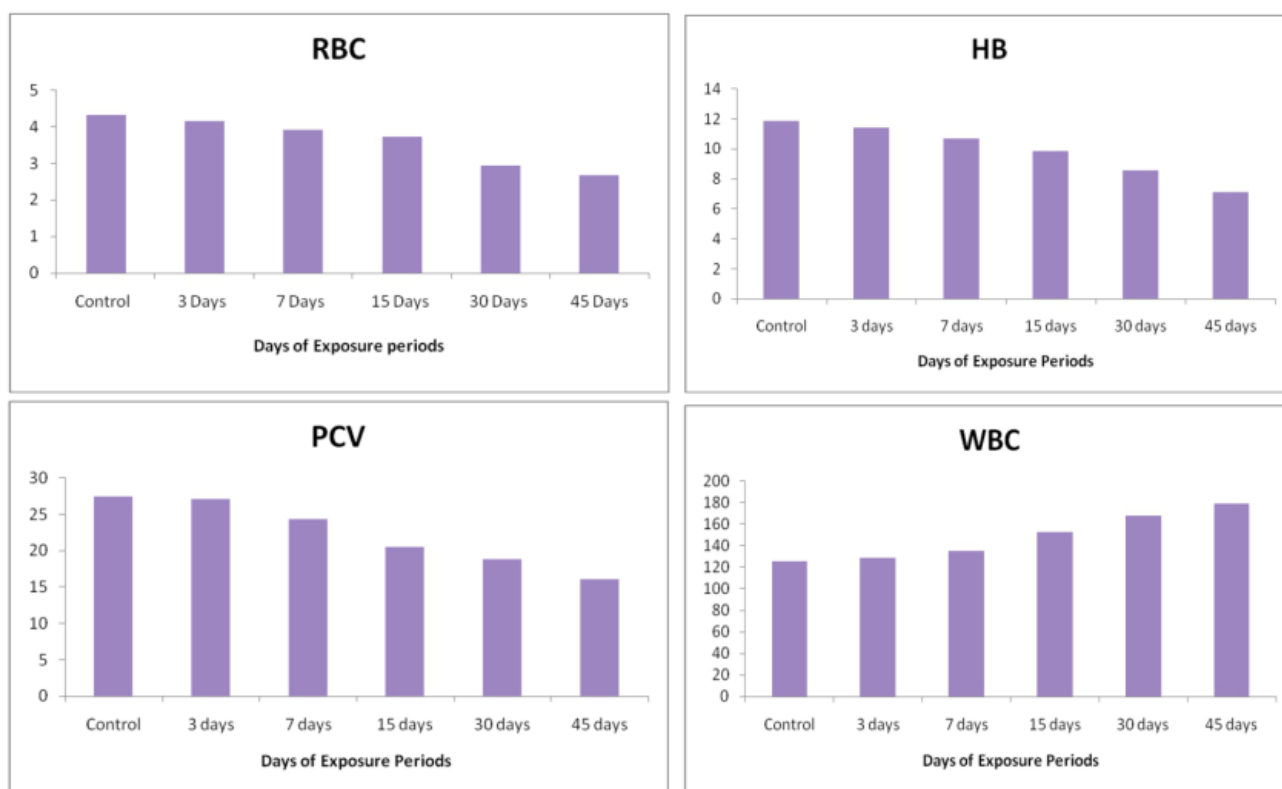


Figure 2. Representing probit mortality Vs log concentration of Heptachlor

Table 2. Changes of Hematological parameters of *Catla catla* after exposed to sub lethal concentration of Heptachlor for 45 days

Hematological parameters	Control	Days of exposure Periods				
		3	7	15	30	45
RBC (m/cu mm)	4.32±0.14	4.16±0.18	3.93±0.21	3.74±0.25	2.95±0.18	2.67±0.21
% Change	---	(-3.70)	(-9.02)	(-13.42)	(-31.71)	(-38.19)
HB (g/dl)	11.86±0.24	11.43±0.05	10.71±0.09	9.86±0.15	8.55±0.12	7.11±0.17
% Change	---	(-3.62)	(-9.69)	(-16.86)	(-27.90)	(-40.05)
PCV %	27.48±1.18	27.11±0.03	24.38±0.07	20.51±0.09	18.88±0.15	16.11±0.11
% Change	---	(-1.34)	(-11.28)	(-25.36)	(-31.29)	(-41.13)
WBC (1000/cu mm)	125±2.58	129.11±2.15	135.23±1.87	153.15±1.15	168.29±1.11	179.15±1.38
% Change	---	(2.99)	(7.87)	(22.16)	(34.24)	(42.90)

**Figure 3.** Variations in Hematological parameters of *Catla catla* after exposed to sublethal concentration of Heptachlor for 45 days

Haematological parameters are potential biomarkers of exposure to agrochemicals due to their sensitivity to certain toxic agents (Heath, 1995). Haematology is an important factor that could be considered for the fish diet quality assessment. Ologhobo (1992) reported that the most common blood variables consistently influenced by diet are the haematocrit (Ht) and haemoglobin (Hb) levels. Probiotics have been used in tilapia (Abd El-Rhman et al., 2009), which reported positive effects on hematological parameters. On the other hand, *O. niloticus* fed diet supplemented with *B. subtilis* (Soltan and El-Laithy, 2008) or supplemented

with *Pediococcus acidilactici* (Ferguson et al., 2010) showed insignificant variation in Hb and PCV contents among the control and fish that were fish groups fed diet enriched with probiotics. Fish fed the diet supplemented with probiotics showed the highest values of Hb, RBCs and WBCs (Marzouk et al., 2008). Reported that both fish groups fed the diet supplemented with dead *Saccharomyces cerevisiae* yeast and both of live *B. subtilis* and *S. cerevisiae* showed significant ($P < 0.05$) variation in the PCV level when compared to fish fed the control diet (Firouzbakhsh et al., 2012).

The decrease in RBC number and haemoglobin content observed in this study might be due to the disruptive action of the pesticides on the erythropoietic tissue as a result of which the viability of the cells might be affected. Alterations in the haematological parameters were brought about by heptachlor as an anemic condition due to decreased synthesis of red blood cells and RBC in bone marrow equivalents (Morgan et al., 1980). The decrease in haemoglobin concentration may be due to either an increase in the rate at which haemoglobin is destroyed or decrease in the rate of haemoglobin synthesis (Moss and Hathway, 1964) (Figure 3).

The increase in WBC count can be correlated with an increase in antibody production, which helps in survival and recovery of the fishes exposed to the toxicant (Seth and Saxena, 2003). A significant increase in WBC count in the present study indicate a hypersensitivity of Leucocytes to heptachlor and these changes might be due to immunological reactions to produce antibodies to

cope up with stress induced by Heptachlor (Ramesh and Saravanan, 2008).

Histomorphological Studies

Histopathological studies have been conducted to help for establishment exposure and other various biological responses. These investigations have also been proved to be a sensitive tool to detect the direct effects of chemical compounds with in target organs of fish in laboratory experiments (Scwaiger et al., 1996; Machado and Fanta, 2003; Sakr and Jamal Allail, 2005). Such analysis appears to be very sensitive parameters and is crucial in determining cellular changes that may occur in target organs, such as the gills, liver and gonads (Dutta, 1996).

The Histopathological studies were provide information about the health and functionality of organs. Tissues injuries and damages in organs can result in the reduced

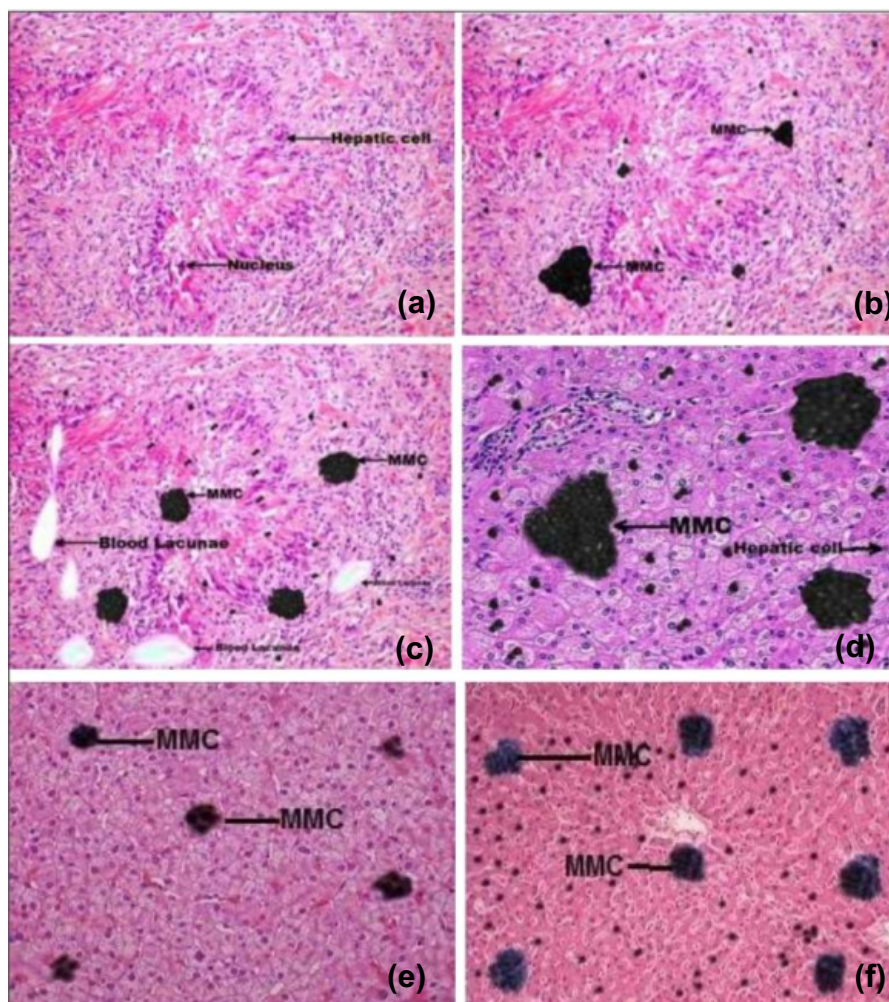


Figure 4. Section of the Liver of *Catla catla* (a) (Control) Showing the large number of hepatocytes and Nucleus: (b) Showing the appearance the appearance the appearance of Melano-macrophage centres exposed to Heptachlor: (C) Showing the appearance of Melano-macrophage centres and Blood lacunae exposed to Heptachlor: (d) Showing the appearance of Melano-Macrophage Centres and Hepatic cells: (e) Showing the aggregation of Melano-macrophage centres exposed to Heptachlor: (f) Showing the aggregation of Melano-macrophage centres exposed to Heptachlor.

survival, growth and fitness, the low reproductive success or increase of susceptibility to pathological agents. Frequency and intensity of tissue lesions depend on the concentrations of insecticides and the length of the period fish are exposed to toxins. Nevertheless, many, insecticides cause specific or non-specific Histopathological damage (Fanta et al., 2003).

Hence an attempt has been made to study the Histopathological modulations in the tissues of liver and anterior kidney of fresh water fish, *Catla catla* exposed to sub lethal (LC_{50}) concentration of heptachlor, for 45 days exposure period.

Histoarchitecture of liver and pathological modulations

The normal histology of liver of fish, showed the presence of:

- hepatocytes cells involved in the synthesis of proteins, cholesterol, bile salts and phospholipids.
- Sinusoids (the blood vessels similar to capillaries but with discontinuous epithelium) and
- Melanomacrophage centers (MMCs) (pigment containing cells and are normally located in the stroma of haemopoietic tissue of the liver).

The present work is the evidence for the result that the sub lethal concentration of heptachlor caused drastic histomorphological variations in Liver and Kidney tissues of *Catla catla*. The Melanomacrophage Centers in Liver and kidney tissues are also significantly varied. Melanomacrophage Centers (MMC) also play an important role in the fish response to foreign materials including infection causing agents.

The liver cells form more than 80 percent of the liver parenchyma and in fish liver they are set around the capillary space sinusoids. There is no basal membrane under epithelium of the liver sinusoids in fish. Hepatic cells play an important role in protein lipid and carbohydrate metabolism. Hepatic cells serve as storage site for some nutrients and also acting as detoxification center. It is obvious from the present results that the sub lethal concentration of heptachlor disrupted the structural integrity of liver tissue of fresh water fish *Catla catla*.

The hepatocytes are large in size and the nuclei are centrally situated. Heptachlor caused loss of basic architecture moderately vacuolated cells and mild shrinkage in liver tissue of fish on day 3.

Heptachlor also caused mild shrinkage and infection of hepatocytes in nucleus with condensation in the structure of chromatin and congestion in central veins in liver on day 7. Heptachlor also cause to increase oxidative stress that causes additional cytoplasmic esinophili nuclear density, necrotic foci are observed.

The liver also showed swelling and pyknosis of hepatocyte nuclei. The liver was severely damaged showing brilliant

proliferative hyperglycemia, peribilliary cirrhosis was manifested by fibrosis of hepatic tissue was observed on day 15. It is witnessed severe degree of vacuolations, degeneration of tissues. With pyknosis of the nuclei, increased the cell size and foamy cytoplasm filled with numerous spaces in liver tissue of *Catla catla* on day 30 (Figure 4).

The toxic effects of heptachlor in the liver of fish revealed a irreversible histopathological changes. On 30th day, diffuse congestion and haemorrhages of blood vessels, dilated and engorged sinusoids, micro to macrovacuolar degeneration of hepatocytes and bile duct hyperplasia were observed.

Heptachlor also induced necrotic changes such as cellular degeneration, Erythrotic cellular nucleus with condensed chromations, lack of nucleolus, mononuclear cell infiltrates, dense melanomacrophages and haemosiderin deposition well also noticed in hepatic tissue on day 45 in *Catla catla* treated with Heptachlor.

On 45th day, heptachlor induced multifocal necrosis, mononuclear cell infiltration, cytolytic changes of haemopoietic tissues and bile ductular hyperplasia with periportal fibrosis. Coagulative necrosis of hepatocytes were also observed.

Besides cellular damages Heptachlor caused the fraction of number of MMC's. The numbers of MMC's are gradually and significantly denser with increase in the exposure periods up to 45 days and the maximum number of MMC's are observed on day 45. The sub lethal concentration of Heptachlor induces also to increase the average size of MMC's in hepatic tissue. The higher average sizes of MMC's is increased in exposure periods up to 45 days compared to the control fishes, indicating the toxic intensity of Heptachlor in aquatic media.

Histoarchitecture of kidney and pathological modulations

Microscopic examination of head kidney of control group showed the presence of:

- haemopoietic tissue (blood forming tissue) and
- renal tubules (excretory in function).

It is obvious that the teleostean kidney and body which contains lymphoid tissue and many nephropores with intestinal lymphoid tissue respectively. The kidney is also well built with haemopoietic tissue, uriniferous tubules and glomerulus with Bowman's capsule made up of epithelial cells. The internal mass of kidney is usually divided into cortex and medulla.

The sub lethal concentration of heptachlor caused

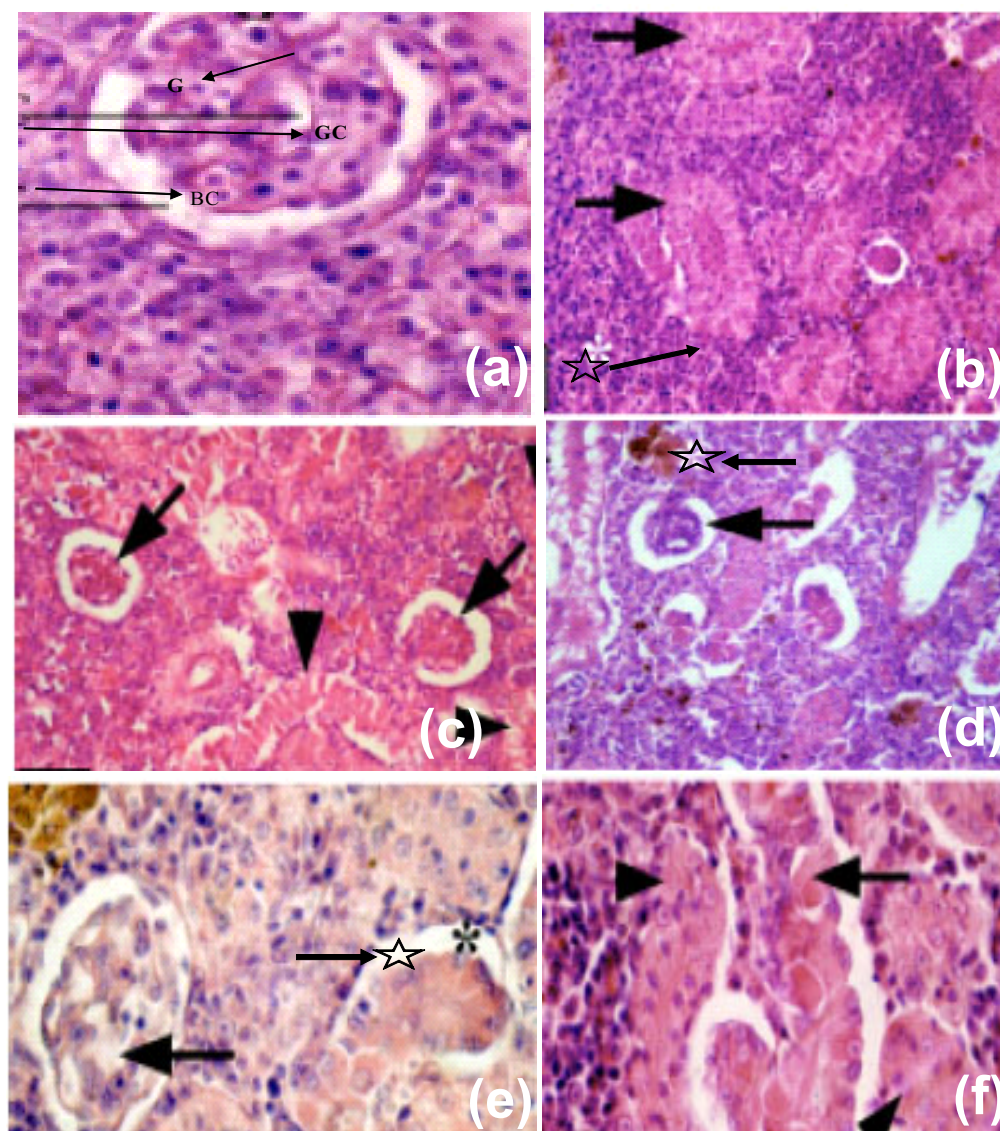


Figure 5. Section of the Kidney of *Catla catla* (a) (Control) glomerulus (G), glomerular capillaries (GC) and Bowman's capsule (BC) are appeared (b) After 3days of exposure: arrows point to renal tubules and (*) indicates lymphoid tissue (c) After 7days exposure to Heptachlor, arrows point to shrunken glomeruli and arrowheads indicate degenerated renal tubules (d) After 15days exposure to Heptachlor, arrow points to damaged glomerulus with increased mesangial matrix and arrowhead indicates severe vacuolation of tubularepithelium. Shrinkage of renal tubules can also be seen. (*) indicates hyaline droplets degeneration (e) After 30days exposure to Heptachlor, break down of glomerular capillaries (arrow) and lifting of tubular epithelium from its original position (*) (f) After 45days exposure to Heptachlor, note the great sloughing of renal tubular epithelium with coagulative necrosis (arrow) and syncytial tubules (arrowheads).

moderately degenerative changes in haemopoietic tissue mild loss of kidney architecture was observed on day 3. Heptachlor also caused moderate losses of kidney architecture hypertrophy ruptured tubular cells are observed on day7. Heptachlor also caused damage the kidney architecture, degenerative changes in haemopoietic tissue, hypertrophy and degeneration of epithelia of renal tubules, on day 15 (Figure 5).

Heptachlor caused ruptured tubular cells and nuclei showing the sign of shrinkage and clumping of blood cells, loss of haemopoietic tissue on day 30. The sub lethal concentration of Heptachlor caused severe damages and modulations are

intensified. The cells constituting the wall of uriniferous tubules have extensively shrunk and deshaped. On 30th day revealed severe congestion and hemorrhages of blood vessels, perivascular fibrosis, micronuclear cells infiltration, severe degeneration and necrosis of renal tubules and atrophy of glomeruli.

Heptachlor influenced on MMC's of kidney of *Catla catla*. The number and size of MMC'S are increased significantly with increase in the exposure periods. The maximum, variations in size and shape of MMC's are observed on day

45. Moreover the severity of lesions were highly pronounced with perivascular fibrosis, degeneration and necrosis of collecting tubules. The dead and desquamated epithelial cells were seen in the collecting tubules.

Conclusion

The present study was carried out to identifying the effect of sub lethal concentration of heptachlor on hematological and histopathological indices of fish *Catla catla*. It is obvious that the sub lethal concentration of heptachlor caused significant variations in haematological indices and irreversible modulations in histoarchitecture of liver and kidney of fish.

The findings of the present investigation revealed a decreasing trend in the haematological parameters such as RBC, Hb content and PCV suggesting that the heptachlor induced changes leads to decrease of haematopoiesis followed by anemic condition due to decreased synthesis of red blood cells. The decrease in haemoglobin concentration may be due to either an increase in the rate at which haemoglobin is destroyed or decrease in the rate of haemoglobin synthesis. A significant increase in WBC count in the present study indicate a hypersensitivity of Leucocytes to heptachlor and these changes might be due to immunological reactions to produce antibodies to cope up with stress induced by Heptachlor.

The Histopathological studies evidenced that the sub lethal concentration of heptachlor caused significant and identical variations in liver and kidney tissues. Heptachlor induced neurotic changes and haemosidrinde position was noticed on hepatic cells at 45 days of exposure period. Heptachlor also causes the fraction of number of MMC's. The MMC's number is dence paralled with increasing of exposure period.

The histopathological changes recorded in the liver and kidneys of *Catla catla* very clearly indicated that the pesticides strongly affect the health of food fish. It is thus suggested that care must be taken not to allow the entry of pesticides into the habitat of the fishes.

Conflict of Interest

We (Authors) hereby declare that we have no conflict of interest of any form pertaining to this research paper.

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