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Histopathological study of vital organs of *Clarias batrachus* (Linn.) Induced to deltamethrin

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Abstract

Deltamethrin is an insecticide belongs to a group synthetic pyrethroids. The current investigation includes histopathological alteration in the liver, kidney, testis and ovary of *Clarias batrachus* exposed to a sublethal concentrations (0.015 ppm conc.) of deltamethrin for 30 days. The induced fish showed significant histopathological changes viz. in liver histology showed degeneration, necrosis and hyperemia, testes histology showed spermatogonia cell's number reduce and condensed, inflammation of cells, tubules vacuolization, and ovary showed follicular cells disruption, epithelial cells degeneration resulted vacuolation, disrupted oogonia with breakdown of germinal vesicle. The present study revealed that deltamethrin affects the fish, *Clarias batrachus* (Bloch.) at very low concentration.

Keywords: Deltamethrin, *Clarias batrachus*, toxicity, insecticide and histopathological

Introduction

Water pollution from agriculture wastes based pollutant like fertilizer, pesticide, etc. has erupted as worldwide problem from beginning of the century. The indiscriminate and injudicious use of the pesticides in agriculture field causes contamination in water bodies like ponds, lakes, and river and wetland regions through surface runoff. For controlling the vectors in public health programme direct spray of pesticides affected the non-target organisms such as fishes, mammals and birds (Lawson, 2011) [12].

Insecticides are the synthesized chemical compounds applied to control a broad spectrum of herbaceous pests which cause destroy the crops and insectivores. Beside advantages, pesticides uses have significant drawbacks. Persistence of insecticides in environment threatens in long-term disorder in ecosystem relation causes the loss of biodiversity. The insecticides that are usually applied belong to chemical groups of Organophosphate, Carbamate, Chlorinated Hydrocarbons, Pyrethroids and Nicotinoids (Suvetha, 2015) [22]. Deltamethrin belongs to type-II pyrethroid compound and popular insecticide having toxic effect on non-target organism especially fish.

Hence, present investigation to illustrate the effects of insecticide, deltamethrin on histopathology of the vital three organs of the experimental fish, *Clarias batrachus*, locally known as "Mangur", is a freshwater air breathing fish.

Materials and Methods

The live healthy walking catfish, *Clarias batrachus* was procured from the local fish market and brought to laboratory in open container, 0.1% KMnO₄ solution bath given to protect from external infection there after acclimatized for 15 days to laboratory conditions. Fish measure average length (9-10 cm) and weight (30-35 g). The fish were fed with artificial pellet floating feed (Growel Pvt. Ltd.) contain 32% crude protein. Daily ration is given at rate 3% of body weight feeding once a day.

To determine LC₅₀ acute values of deltamethrin insecticide for 24, 48, 72 and 96 hours following the methods of APHA (2005) [3]. The LC₅₀ values for these periods were resulted as 1.5 mg, 0.85 mg, 0.45 mg and 0.15 mg respectively. For the sub-lethal concentration followed Hart *et al.* (1945) [8] method.

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10 acclimated fish were exposed to a sub-lethal concentration (0.015 ppm) of deltamethrin (Calculated by Finney 1978) [7], and side by side 10 fish as that of experimental one was maintained as the control group for 30 days. On 30th day at the end of exposure period the fish were sacrificed liver, testis and ovary were quickly dissected out. Small pieces organs were fixed in ten percent Neutral Buffered Formalin for 18-24 hours. The standard histological method (Luna, 1968) [13] was followed to prepare paraffin embedded tissue blocks. Rotary microtome was used to cut tissue blocks into serial sections (5-7 μ). Haematoxylin-Eosin (HE) stain was used to stain the histological sections. Photomicrographs of fish tissue samples were taken.

Results

Tissue samples liver, testes and ovary of *C. batrachus* were treated with sublethal deltamethrin concentration 0.015 mg/l at 30 days. Histopathological observation of three vital organs of treated fish as well as control one have described below.

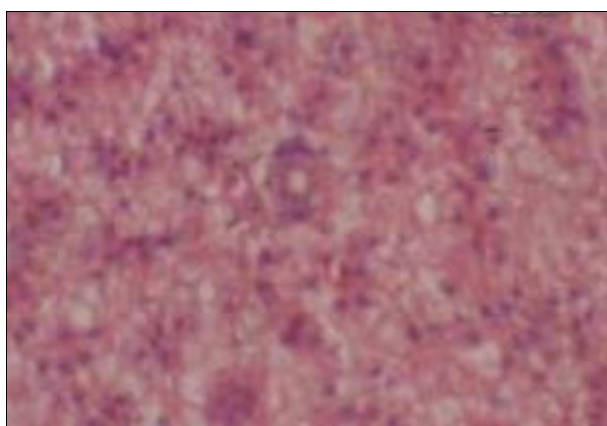


Fig 1: a) Section of normal liver tissue of *C. batrachus*. 100X H.E.

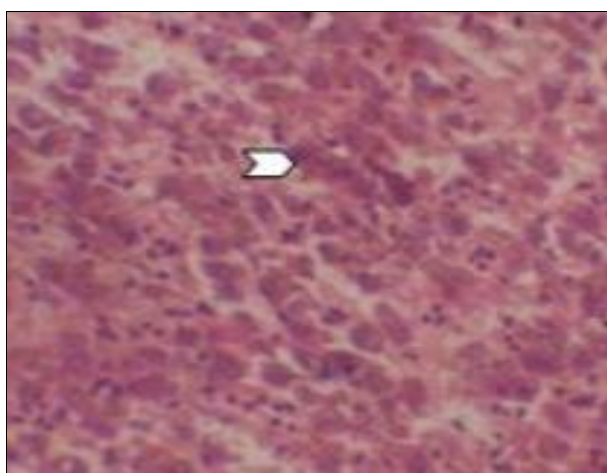


Fig 1: b) Section of the liver of Deltamethrin 0.015 mg for 30 days treated *C. batrachus* showing necrotic hepatocytes, hemorrhage (\rightarrow) and blood congestion. 100X H & E.

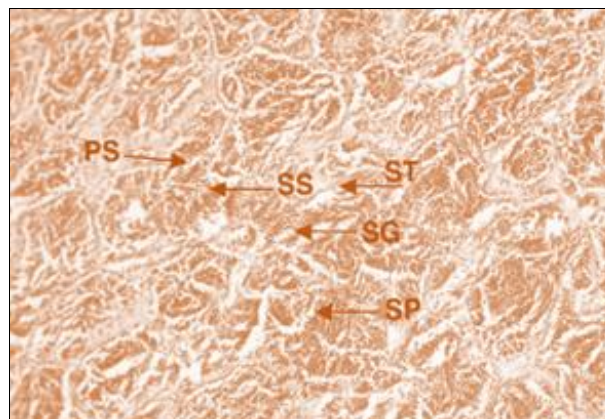


Fig 2: a) – Section of the testes of control *Clarias batrachus* showing spermatide (ST), sperm (SP), secondary spermatocyte (SS), spermatogonia (SG), primary spermatocytes (PS). 200X H & E.

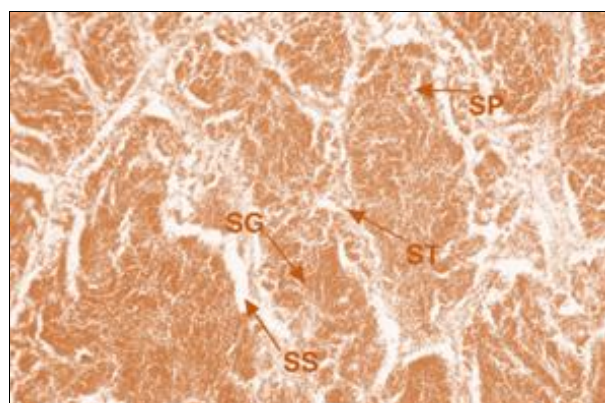


Fig 2: b) Section of testes of the 0.015 mg/L deltamethrin for 30 days exposed *Clarias batrachus* showing spermatide (ST), sperm (SP), spermatogonia condensation (SG), and secondary spermatocyte vacuolation (SS). 200X H&E.

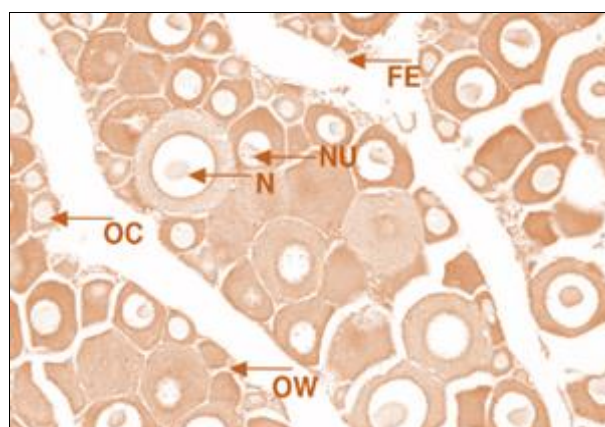


Fig 3: a) Section of the ovary of control *Clarias batrachus* showing (NU) Nucleolus, (OC) Oocyte, (OW) Ovarian wall, (FE) Follicular epithelium, (N) Nucleus. 200XH&E.

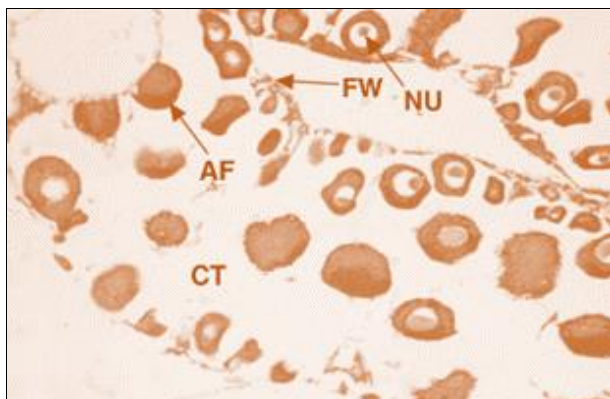


Fig 3: b) Section of the ovary of deltamethrin- 0.015 mg/L for 30 days exposed *Clarias batrachus* showing (NU) Nucleolus condensed, (FW) Follicular wall disrupted and (CT) Connective tissue degenerate (AF) Atretic follicle. 200XH&E.

Liver

The liver of the control *C. batrachus* composed of a mass of polyhedral cells. The polyhedral cells remain arranged in groups enclosing bile passage. Each hepatocyte has a granular cytoplasm taking deep stain and centrally placed spherical nucleus containing prominent nuclei and chromatin. The liver is richly supplied with blood vessels. The parenchymal cells made hepatic tubules which carried blood sinusoid. The hepatocytes are arranged around the blood sinusoid in cord-like structure known as hepatic cell cord. There are bile ductile in between the cord of hepatic cells, the terminal biliary system is disposed between the cell plates and the parenchymatous ground structure, exhibited moderate vacuolization (Figure:-1.A.). The histology of the treated group of *C. batrachus* exposed to sub-lethal concentrations of deltamethrin- 0.015 mg/L for 30 days showed hepatic cirrhosis in form of vacuolization, space formation and resulting haemorrhage, vacuolar degeneration, necrosis, hyperemia and mononuclear cells filtration in portal regions (Figure:-1.B.).

Testes

The testis of normal fish group of *C. batrachus* are paired, elongated whitish structure situated on either side, ventral to the kidney, in abdominal cavity at posterior region. They are attached to the body wall through mesovarium. The testes are formed of a large number of seminiferous tubules of different sizes. The tubular walls are internally lined by germinal epithelium. Cells of various spermatogenic stages are present in each testicular lobule. The spermatogonium or sperm mother cell's shape is spherical or rounded and centrally placed nucleus. The primary spermatocytes are smaller in size than spermatogonia and are having darkly stained nuclei. Mostly lobules consist sperm nests (Figure:-2.A.). The treated fish group sub-lethal concentrations of deltamethrin- 0.015 mg/L for 30 days revealed remarkable alteration in the histology of testes like the seminiferous tubules are normally of varying shapes and sizes, each tubule has a definite thin fibrous wall which is not distinguished after spawning. It shows reduction in the number and condensation of spermatogenic cells as well as inflammation of cells, contraction and vacuolation of tubules (Figure:-2.B.).

Ovaries

The control fish group of *C. batrachus* has a normal histology

have thick ovarian wall with increased vascular supply and conspicuous blood capillaries. The connective tissue in the stromal was evident in good volume. The germ cells become associated with small epithelial cells more into cortex. The associated epithelial cells multiply and surround the germ cell which is now called oocyte developing into the stage I, stage II, stage III etc. and they will develop into the mature ovum which is nourished by the surrounding follicular cell (Figure:-3.A.). The *C. batrachus* after exposure to sublethal dose of Deltamethrin- 0.015 mg/L showed disrupted follicular cells. Nucleolus shows condensation of crescent shaped dark granules at one side. Degeneration of epithelial cells causes vacuolation, breakdown of germinal vesical, many disrupted oogonia are the changes caused due to the exposure of ovary of *C. batrachus* to sublethal dose of Deltamethrin showing in (Figure:-3.B.).

Discussion

The results as histopathological alteration of vital organs i.e. liver, testis and ovary of *Clarias batrachus* in response to sublethal dose of deltamethrin exposure have been discussed. The deltamethrin exposed *C. batrachus*'s liver in current investigation showed that hyperplastic hepatic and necrosis of hepatic cells. The findings of present study have similarity with earlier workers (Fricke 2012; Kalaiyarasi, 2017; Nordin, 2018) [6, 10, 15]. According to Mishra & Poddar (2016) [14] organ liver is considered as the major metabolic center of the body. Accidentally damages of liver would result so many physiological disturbances leading to subsequent mortality of fish. The fish exposed to cypermethrin become in stress and detoxification of the cypermethrin causes necrosis in hepatic cells (Ullah, *et al.* (2015) [23]. Similarly, *Labeo rohita* exposed to cypermethrin showed significant histopathological changes like hyperplastic hepatic and necrosis of hepatic cells (Jee *et al.*, 2005) [9]. Osman *et al.*, (2009) [16] recorded congestion and hemorrhage in the hepatic sinusoids with dilation of hepatic vessels, vacuolization and degeneration of hepatic cells, due to heavy metal salts exposure. Similarly other study Andem *et al.*, (2016) [2] the liver of *Clarias gariepinus* exposed to the cypermethrin showed hyperplasia, disintegration of hepatic mass, and focal coagulative necrosis. Recently, Sangeeta, *et al.*, (2020) [17] reported that application of ammonium chloride on liver of *Clarias batrachus* has similar changes in histology. The present experiment with Deltamethrin also showed similar results.

Sokal *et al.*, (1985) [21] postulated that testicular inflammation was commonly observed when environmental toxicants induce to aquatic and terrestrial animals, resulted inflammatory lesions, tubular cells vacuolated and along with distortion of seminiferous cells. Baronia and Sahai (1993) [4] reported albino rats exposed to pesticides showed that histological changes in degeneration of seminiferous tubules, interstitium getting enlarge and intertubular area haemorrhaged. Ahsan and Ahsan, (1974) [1] reported that *C. batrachus* injected to cadmium showed that an identical arrest of spermatogenesis. Recently, Shyni & Sreedhar (2014) [20] reported that sublethal dose of urea on testicular structure of the black clam resulted chronic effect on histology of testes. Similarly, Zutshi (2005) [25] reported the toxic effect of fenthion on the testes of *Glossogobius giuris*. Lata, *et al.*, (2008) [11] observed reduction in size with spermatids and sperms in degenerating condition. Recently, Sangeeta, *et al.*, (2020) [17] ammonium chloride exposed to fish *Clarias batrachus* showed same histological changes. The present

histopathological work reports are similar to the present findings.

Toxicity effect exhibited in histology of ovary in form of germinal vesical degeneration, epithelial cells necrosis resulted vacuolation and oogonia disruption. The ovaries damages were observed in present histological study of deltamethrin induced to fish, *Clarias batrachus*. The histopathological findings of present work also supported by Saxena and Saxena, (2008) [18]. The endosulfan induced zebrafish showed histological abnormalities in ovaries (Chow, 2013) [5]. Several effluents and aquatic pollutants toxic affect observed on ovary (Shukla *et al.*, 1984) [26]. The heavy metal exposed *Cyprinus carpio* showed histological changes in ovary reported by Vinodhini *et al.* (2009) [24]. Nitrogenous fertilizer, ammonium chloride exposed fish, *Clarias batrachus* showed histological changes in ovary like germinal vesical degeneration, epithelial cells necrosis and disrupted oogonia (Sangeeta, *et al.*, (2020) [17]. The above studies support the present experiment with Deltamethrin.

Conclusion

The sub lethal concentration of deltamethrin (0.015 ppm) for 30 days exposed experimental fish revealed significant histopathological changes in three vital organs, liver, testis and ovary. The present study concluded as such histopathological alteration would be due to dysfunction metabolism and energy demand in fish under stress caused by deltamethrin exposure.

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