ASSESSING TOXIC THREAT OF COPPER CHLORIDE ON OVARY OF FISH, *Clarias batrachus* (Bloch)

A. M. Avinashe
P.G. Dept. of Zoology, S.S.S.K.R. Innani Mahavidyalaya Karanja (Lad), Dist-Washim (M.S.), India
ajayavinashe@gmail.com

ABSTRACT

The main purpose of historic Rishi Lake of Karanja (Lad) Dist. Washim is fish culture, irrigation, animal activity and domestic purpose. The water body of lake is polluted by effluent / agriculture run-off containing numerous chemicals, Insecticide, pesticides constantly used in agriculture sector as well as human- animals' activity. Water pollution is a serious issue in the world and in developing countries like India on account of huge population. Aquatic pollution has its own importance and directly concern with human life, as water is the main component of living organism. One of the aquatic animal, fresh water fish, directly affects by polluted water on its external and internal, vital organs too.This piece of work includes toxic effect of copper chloride on the histopathology of ovary of the fresh water fish, Clarias batrachus (Bloch). Arranged two sets of aquarium for the experimental purpose and administered the sub lethal doses of copper chloride for exposer period of 7, 14, 21 and 28 days. Remove ovary of experimental fish, and carry forward for microtechnique stages, and made observation for the point of investigation findings.

Keywords: *Copper chloride, Ovary histopathology, Clarias batrachus*

Introduction

Water pollution is a major problem of the century and addition of pollutants changes the natural qualities of water. So that it is easily reflected that the water is to be clean and free from pollution, for safe food and drinking water. The harmful effect of any toxicant which was moved to water bodies can see assessed by investigation health of aquatic fauna. The contaminant are entered in the body of organism via absorption and other routes, goes deep in the tissue and affects physiology, biochemistry and metabolism of the organism.

Among the environmental pollutants, metals, pesticides, insecticides, inorganic and organic chemicals are mostly concerned to their potential toxic effect and ability to bio accumulates in ecosystems. Many species of fresh water fishes, especially catfishes have been found to bio accumulate most of these heavy metals in their tissues (Cu, Cd, Zn, Pb, Fe, etc.) Heavy metals are most injurious to fish. This pollution has not only depleted the fish stock but also has threatened the human health by incorporating into food chain (Pip, 1995).

Metal pollution from multifarious sources like effluents from run-off and untreated sewage system has adverse effects on aquatic ecosystem. The metal contamination in aquatic ecosystem is yet considered to be unsafe not only for only the human beings, but also for the other aquatic organisms. Fish living in the metal contaminated water may accumulate toxic trace metals via their food (Mendel and Uluozlu, 2007).Overspray and runoff of pesticide from agriculture field may easily find their way into the natural water surfaces and adversely affect the quality of water surface and creates hazards for aquatic life resulting in serious damage to non target species including fish (Bondarenko et al., 2004).

The chemicals have entered into the aquatic system and create pollution, which pose a great threat to aquatic organisms including fish. There are several reports regarding the effect of pesticides on physiology o fish (Girija, 1984). Copper is among the most toxic of the heavy metals in the fresh water and marine biota and often accumulates and causes irreversible harm to some species at concentrations just above levels required for growth and reproduction.

Materials and Methods

Ten live female species of fresh water fish, *Clarias batrachus* were selected for the present experiment work having weight between 70 -76 gms and length between 19-22 cms. The fish were collected from the Adhan Dam, Injuri Tq. Karanja. The fish were brought to the laboratory

in well oxygenated plastic bags without any injury. Fish were washed by taking 1% KMnO4 solution for 5 minutes for removal of dermal infections. Then fish were allowed to acclimatize to the laboratory condition for a period of fortnight prior to the conducting the experiment. Food provides to the fish, small pieces of boiled eggs particularly at morning hours. The fishes were maintained in separate aquaria containing aged tap water.

Water used-Aged tap water used during entire experimental work. As per the standard method the physicochemical parameters of used aged tap water were determined periodically (APHA, 1998).

The physicochemical characteristic of water used –

- D.O. = 6 + 0.3 mg/L
- Temperature = $29^0C \pm 1^0C$
- Total hardness = 228 ± 2mg/L
- Total alkalinity =243 ± 3mg/L

Test Toxicant: The Copper chloride was selected as a toxicant for the present work. The physiochemical properties of Copper chloride are as follows,

- Properties of heavy metal : Copper chloride
- Mole. Weight :170.47
- Melting point : 200^0C ($-2H_2O$)
- Boiling point : Decompose
- Solubility : Soluble in water
- Appearance : Blue green
- pH after solubility :Acidic

Preparation of Experimental aquarium:

96h LC_{50} and sub lethal concentration of copper chloride for the fresh water fish, *Clarias batrachus* was made available from literature. This was 0.005mg/l as 96h LC_{50}value and approximately 5 times (or 20%) less concentration as sub lethal concentration i.e. 0.001mg/L, was used to exposed fish for 28 days to study the histological study of ovary. Aqueous solution of copper chloride was added to the glass aquaria of 60x30x30 cm size containing 25 litre of water, toxicant solution added drop by drop with constant stirring. Total 5 female *C. batrachus* were transferred in to the experimental aquaria contains toxic solution. The Control/ Normal set having 5 female species of *C. batrachus* also run along the experimental set in similar size of aquaria. fishes were provides feed as small pieces of boiled eggs, once in a morning hours.

After the experimental days i.e.7, 14, 21 and 28 days is over, the experimental fish and control fish sacrifices in laboratory and remove the ovary of female fishes. This ovary washed and treats as per usual methods for the microtechnique process.

Observation

After the experimental design of timing, the ovary removed after exposure to sub lethal concentration i .e.0.001mg/L of toxicant copper chloride. Removed ovary proceeds further as usual method of microtechnique including fixation, dehydration of tissue, block preparation, section cutting, and staining process. To select quality slides of control and experimental sets for the observation purpose.

Histological structure of Ovary

Control Set: The histological slides of ovary of fresh water fish, *Clarias batrachus* observed under the research compound microscope. In control stages of ovary shows number of ovarian lamellae which are in the form of finger shaped projections, protruding in to the ovarian lumen. The ovary of control fish is dominated by the immature oocytes. These are characterised by small sized, large nuclei and deeply stains cytoplasm. Maturing oocyte is occasionally present. They are large in size, with large nucleus and several nucleoli. Few degenerating oocytes are also noticed during observation (Fig 1), in all control stages of 7, 14, 21 and 28 days, closely similar observations were find out.

Experimental Set: Fresh water fish, *Clarias batrachus (Bloch)* were exposed to experimental toxicant copper chloride to the sub lethal concentration i.e. 0.001mg/L for the period of 7 days, 14 days, 21 days and 28 days noticed the following observations.

Extensive vacuolation found in the oocortex, in the young oocyte (Fig.2), After 14 days of exposure, the necrosis of oolemma and hypertrophy of follicle cells was observed (Fig. 3). Degeneration of vitellogenic and large previtellogenic oocytes was noticed after 21 days exposure to sub lethal conc. of experimental toxicant copper chloride (Fig.4). After the 28 days exposure of experimental toxicant (Sub lethal conc.) observed prominent changes in the oolemma which resulted in atresia of oocyte (Fig.5).

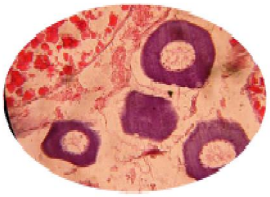

Fig.2

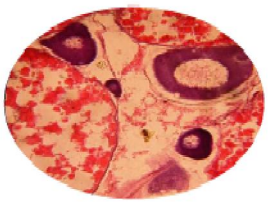

Fig.3

Fig.1

Fig.4

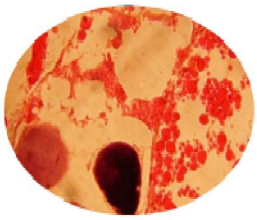

Fig.5

Fig.1-Transverse section through the ovary of fish, *Clarias batrachus* (Control). Haematoxylin-eosin, x 630.

Fig.2-T. S. through the ovary of fish, *Clarias batrachus*, exposed to sub lethal concentration of $CuCl_2$ for 7 days. Haematoxylin-eosin, x 630.

Fig.3-T. S. through the ovary of fish, *Clarias batrachus*, exposed to sub lethal concentration of $CuCl_2$ for 14 days. Haematoxylin-eosin, x 630.

Fig.4-T.S. through the ovary of fish, *Clarias batrachus*, exposed to sub lethal concentration of $CuCl_2$ for 21days. Haematoxylin-eosin, x 630.

Fig.5-T.S. through the ovary of fish, *Clarias batrachus*, exposed to sub lethal concentration of $CuCl_2$ for 28 days. Haematoxylin-eosin, x 630.

Discussion

In the present piece of work, the toxic effect of sub lethal concentration copper chloride (0.001mg/L)on the ovary of fresh water fish, *Clarias batrachus*(Bloch), indicate that the process of oocyte maturation seem to be arrested and various degeneration changes were noticed in the structure of ovary, effect due to experimental toxicity.

According Baile and Kadu (1992) observed impaired oocyte maturation in the ovary of air breathing fish, *Clarias batrachus* exposed to sub-lethal concentration of $CdCl_2$ for 40 days.

Lamely (1996) reported taratogenic deformities in sunfish as permanent pathological marker of selenium poisoning. They are congenital malformations that are due to excessive selenium in eggs. Excessive dietary selenium causes elevated concentrations of selenium to be deposited in developing eggs, particularly in yolk, both as an energy supply and as a source of protein for building new body tissue.Necrosis of oolemma and hypertrophy of cells was observed when the fresh water fish, *Channa punctatus* exposed to experimental toxicant, selenium dioxide for the 21days, reported by Avinashe and Patil (2013).

According to Wehbi and El-Greisy (2007) a 42 days direct exposure to diluted level of effluent did not have observable effects on the survival of fish, *Siganus rivulatus*, but lead to decrease in GSI in both sex and decreases in fecundity and fish fertility. GSI decreases have been reported in fish after exposure to breached kraft meal effluent or exposure to domestic effluent. Effects on gonads including reduction of gonad size and reduction in number of germ cells may bring the animal to sterility by complete voiding of germ cells.

Srivastav *et al*.(2008) noticed devicyprin induced gonadal impairment in Channa punctatus(Bloch) and they reported degenerative changes as fragmentation of nucleus in the chromatin network, displacement of yolk vesicle at the periphery, vacuolization in the nucleus, stromal haemorrhage and damage of germinal epithelium. According to them such histoanatomical abnormalities in ovaries may be caused by several factors, viz. ionizing, radiations, electric current, parasite infections, mechanical injuries, xenobiotic toxicants.

Thus the above result of present work of investigation as 'Assessing toxic threat of Copper chloride on ovary of Fish, *Clarias batrachus* (Bloch)' is almost similar to those of the above investigators.

References

APHA, (1998). Standard Methods for the experimentation of water and waste water. American Public Health Association, 874.

Avinashe, A.M. and Patil, G.P. (2013). Toxic effect of selenium dioxide on Histopathology of ovary of fresh water food fish, *Channa punctatus* (Bloch). Nat. Conf. on Advance in Life Sciences; Present and Future .P183-186.

Baile, V.V. and Kadu, M.R. (1992). Toxicity of Cadmium chloride on the ovary of Catfish, *Clarias batrachus* (Linn). Indian J. Comp. Anim. Physiol. 10(1); 7-11.

Bondarenko, S., J. Gan, D.L. Haver and Kabashima, J.N. (2004). Persistence of selected organophosphate and carbonate insecticide in water from a coastal watershed. Environmental Toxicology and chemistry 23 (11) : 2649-2654.

Girija M. (1984). Effect of Heptachlor and Metasystox on fish,*Tilapia mossumbica* (petero), M.Phil Dissertation Shri Venkateshwara university, Tirupati, India.

Lemly, A.D.(1996). Assessing the toxic threat of selenium to fish and aquatic birds, Environ.Monit. Assess. 43: 19-35.

Mendel D. and O.D. Uluozlu, (2007). Determination of trace metals in sediment and five fish species from lakes in Tokat, Turkey, Food Chemistry, 101, 739-245.

Pip E. (1995). Cadminium, lead and copper in freshwater mussel from Assiniboine river Monitoba, Canada J. Moll. Stud., 61:295-302.

Srivastava, R.K., Yadav K.K. and Trivedi, S.P. (2008): Devicyprin induced gonadal impairment in a fresh water food fish, Channa punctatus (Bloch) J. of Environ. Biol., 29 (2): 187-191.

Wehbi, O.M. and El-Greisy, Z.A. (2007). Comparative impact of different waste sources on the reproductive parameters and histology of gonads, liver and pituitary gland of *Siganus rivulaus*. J.of Applied Sci. Res. 3 (3): 236-244.