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Zoology

TOXIC EFFECT OF COMMONLY USED FERTILIZERS ON THE GILLS OF FRESH WATER TELEOST *CLARIAS BATRACHUS*

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ABSTRACT The effects of commonly used fertilizers like diammonium phosphate, urea and calcium ammoniam nitrate on the histology of the gills of fresh water fish *Clarias batrachus* were studied. The toxic effect of diammonium phosphate was more pronounced than that of urea and calcium ammoniam nitrate. Diammonium phosphate is highly toxic fertilizer. Scattered necrosis of the lining cells of the lamellae with distortions where seen in gills of the fish. Pronounced congestion in the gill lumen and hyperchromatic nuclei of primary gill lamellae was seen in the fish. Calcium ammonia nitrate caused destruction of secondary gill lamellae but mucusal lining was well preserved in the fish *Clarias batrachus.*

KEYWORDS : Fertilizer, Diammonium phosphate, Gill, Teleost, *Clarias batrachus.*

INTRODUCTION

In the present day world of science and technology, there has been all-out effort for in increasing crop-production both in terms of quality and quantity. The chemical application in agriculture has thus become an essential part of increasing crop-production. This has given rise to manifold complex problems with regard to aerial, aquatic and terrestrial pollution. These fertilizers significantly decreased absolute erythrocyte count, haematocrit and haemoglobin of teleost fish *Clarias batrachus* and *Heteropneustes fossilis* when they were exposed to LC_{100} of the individual fertilizer for 24, 48, 72, 96, 120 and 144 hours of intervals (Singh, 1982 Naqvi, 1983). Simultaneously, total leucocytes gradually increased with increasing concentrations of the fertilizer giving an idea of infection to the fish. Remarkable histopathological changes in liver, kidney, gill and muscles were observed in the toxicity of above fertilizers, diammonium phosphate (nitrogen 18%, phosphoric acid 46%), urea (nitrogen 46.0%) and calcium ammonium nitrate (Nitrogen 25%, Calcium oxide 20%) (Naqvi, 1993). When these fishes were exposed to smaller concentrations of the fertilizers for prolonged intervals, i.e. 15-30 days, haemopoiesis was suppressed and hypercholesterolaemia with hyperproteinemia has occured.

MATERIALS AND METHODS

The fish were collected from the river Gomti of Lucknow with the help of local fishermen using hand nets. They were brought to the experimental laboratory in large plastic containers in natural water, avoiding injuries and stresses of all kinds. The fish were washed five times in tap water, then treated with 2.0% $KMn0_4$ to remove external infections. Normal, uninfected, and apparently healthy fish selected for the experiment were transferred to large glass aquaria and acclimated for 120 hr. Different concentrations of DAP, urea and calcium ammonium nitrate were prepared in aquaria and the effect on the fishes were observed. The body organs like gill, liver, kidney and gastrointestinal tract (Git) were quickly removed and fixed in 10% formal saline solution. Paraffin sections of 3 micron thickness were cut, stained with heamatoxylin and eosin for microscopic examinations.

OBSERVATIONS AND RESULTS

There are four pairs of gills and two rows of primary gill lamellae borne by the ceratobranchial and epibranchial segments of each gill arch. The interbranchial septum between the two rows of lamellae is short, so that the lamellae of two rows are free at their distal ends.

The gill lamellae are supported by gill rays which are partly bony and partly cartilaginous and are connected with the gill arch, and with each other by fibrous ligaments. Each gill ray is bifurcated at its proximal end to provide a passage for the efferent branchial vessel. Each primary lamella bears a large number of secondary lamellae on both of its sides. These flat leaf-like structures are the main seat of gaseous exchange. Secondary lamellae are free from each other. Each secondary lamella consists of a central vascular layer surrounded by a thin layer of connective tissue and epithelium. The vascular layer consists of a net work of capillaries supported by Pilaster cells. Only one afferent and on efferent branchial vessel is present in each gill arch in *Heteropneustes fossilis* but in *Clarias batrachus*, there are two efferent branchial vessels in each arch. Each efferent vessel runs through the entire length of the gill arch and gives off a series of primary afferent branches to the primary gill lamellae. Each primary afferent vessel divides laterally into a number of secondary vessels which run across gill rays dividing again into two to four tertiary branches to supply the secondary lamellae in the secondary lamella, each afferent vessel breaks up into a number of minute capillaries, interconnected with each other and forms the vascular central core of the secondary lamella. Exchange of gases takes place while the blood is circulating through these thin capillaries of the secondary lamella. These capillaries finally join to form a short vessel which carries the blood to the primary efferent vessel running along the margin of the primary gill lamella. The primary efferent vessel, thus, collects blood from the secondary lamellae of both sides and carries the oxygenated blood to the main efferent branchial vessel of the gill arch. The gill head is covered by a thick epithelium in which a large number of mucus glands are present. Besides the mucus glands, large eosinophilic cells and taste buds are also present in the epithelium covering the gill arch.

Each gill arch has one set each of abductor and adductor muscles. The abductor muscles are present on the outerside of the gill arch connecting it with the proximal ends of the gill rays. The adductor muscles are present in the interbranchial septum and cross each other so as to become inserted on the opposite gill rays. Normal histology of gill of fishes *Clarias batrachus* is shown in figure -1. Scattered necrosis of the lining cells of the lamellae with distortions where seen in gills of the fish *Clarias batrachus* due to the toxic effect of fertilizer diammonium phosphate (figure -2). Gills are externally situated structure so, this structure shows pronounced congestion in the gill lumen and hyperchromatic nuclei of primary gill lamellae in *Clarias batrachus* (figure -3). Calcium ammonia nitrate caused destruction of secondary gill lamellae but mucosal lining was well preserved in the fish *Clarias batrachus* (figure -4).

DISCUSSION

Diammonium phosphate caused remarkable histopathological change in body organs – gill, liver, kidney and gastrointestinal tract of fish species *Clarias batrachus* and *H. fossilis* used in these studies at the LC_{50} within particular time exposures. Histological changes attributed to ammonia exposure causes swelling and diminished number of red blood cells, inflammation and degeneration of gills and kidneys, and lowered resistance to disease (Reichenbach-klinke, 1967; Flis, 1968; Smart, 1976 and Thurston et al., 1978). These histological changes were observed for a variety of fish species, tested under variety of water observed for a variety of fish species, tested under variety of water chemistry conditions. Solid residues, water effluents, and air emissions from phosphate fertilizer-producing industries cause the pollution of water, air, and land. A decrease in RBC number and hemoglobin was observed in the fish *Trichogaster fasciatus* after exposure to the fungicide RH-216 (Raizada and Gupta, 1982) with CCl_4, injections (Sharma and Gupta, 1982), while in *Heteropneustes fossilis* 7.6 ppm of malathion in the medium resulted in a fall in haemoglobin content from 12.35 to 11.42 g% (Mishra and Srivastava, 1983)., Inhibitory effects of ammonia and urea on gillcarbonic anthydrase enzyme activity of rainbow trout *Oncorhynchus mykiss* was also observed Hisar et al., (2004). Effect of sub lethal concentrations of Tobacco (Nicotana tobaccum) leaf dust on some biochemical parameters of Hybrid catfish, *Clarias gariepinus* and *Heterobranchus bidorsalis* was also reported (Kabir et al., 2011).

Ammonia toxicity in fish (Randall et al., 2002), and acute toxicity of inorganic fertilizers to African catfish, *Clarias gariepinus* was also reported (Ufodike et al., 2008).

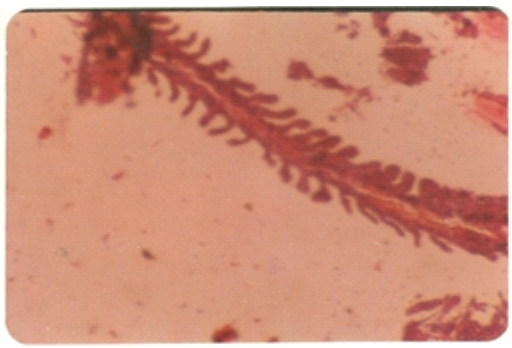

Fig. -1 Gill of Control fish *Clarias batrachus* H&E X 450.

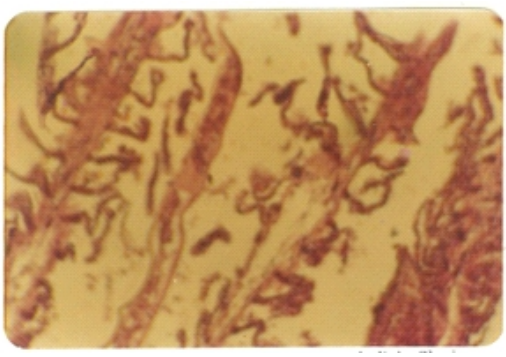

Fig. -2 Gill of Diammonium Phosphate treated fish fish *Clarias batrachus* showing distortion of the gill lamellae with scattered necrosis of the lining cells. H&E X 450.

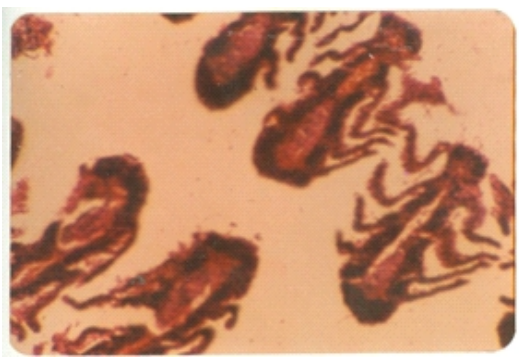

Fig. -3 Gill of urea treated fish *Clarias batrachus* showing hyperchromatic nuclei of primary lamellae with congestion in the lumen. H&E X 450.

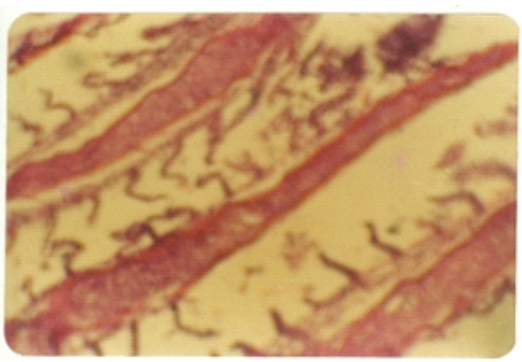

Fig. -4 Gill of Calcium Ammonium Nitrate treated fish *Clarias batrachus* showing destruction of secondary lamellae but lining of mucosa are well preserved. H&E X 450.

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