



**RESEARCH ARTICLE**

**Studies on The Toxicity of Pesticide Methylamine on Few Catfish Species**

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**ABSTRACT**

The Present study was carried out in the Department of Zoology, N.R.E.C. College, Khurja from 2008 to 2010. In the present study, the toxicity of methylamine on freshwater teleosts i.e. *Clarias batrachus* and *Heteropneustes fossilis* have been noticed. Lethal and sublethal levels of the pesticide methylamine have been considered for histomorphology and histopathology in various organs viz. gills. The results revealed that the pesticides are the primary source of pollution in pond water and threatened too many diseases in aquatic living organisms.

**KEYWORDS**

Methylamine, *Clarias*, *Heteropneustes*, Sublethal, Pesticides.

**INTRODUCTION**

Fish is one of the most important foods for human health and a rich source of protein, fat, minerals, and vitamins. Fish liver oil is rich in many vitamins like A, D, and B-complexes. The fish therefore can solve the problem of malnutrition in poor and developing countries. The pesticidal pollution is a global burning problem due to its extensive use. Pesticides are the chemicals used to control pests. Their residue reaches plants and animals through air, water, and soil. In the environment, human beings and domestic animals (cows, pigs, and pets) ingest these pesticides through food and fodder respectively, by consuming milk and meat products exposed to pesticides animals, further endanger human health. Pollution through the air, also deposits pesticide residue on crops. Pesticides may find their way into the over groundwater reservoirs, streams thus producing

an adverse impact on the aquatic biota including fishes. The concentration of various pesticides is increasing day by day in the ponds, streams, and rivers since they are used for insect control. These synthetic chemical compounds have altered the chemical nature of our environment. These chemicals reach in freshwater ecosystem due to spraying by public authorities, forest management and in agricultural fields. Thus day by day increased use of pesticides is poisoning significant threat to freshwater ecosystem especially fish. Fishes are highly susceptible to pesticides. Freshwater animals like fishes are one of the primary sources of food for rapidly increasing human populations, but the discharge of agricultural, municipal and industrial wastes into the aquatic environment causes high mortality of fishes. American public health association (APHA-1995), American water works association (AWWA) and water Environment Federation (WEF) in their 19th, 20th and 21st editions gave standard methods for the examination of water and wastewater. The environmental stress may be chemical, physical or radioactive in nature. Balasubramani and

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Pandian (2008) found endosulfan suppresses growth and reproduction in zebrafish. Chemical pollution caused by different types of chemical substances viz., elements and their compounds, various organic substances such as insecticides, pesticides, solvents, fertilizers and variety of drugs to which we are frequently exposed. Even if present in minute quantities, their range, toxicity, and persistence hurt ecological systems of land and water, which cannot separate from the animal life, it's health and environment. Today about 1,000 or even more chemical formulations are used as pesticides around the world, in which about 250 are commonly used in agriculture, including about 100 insecticides, 50 herbicides, 50 fungicides, 20 pesticides and 30 other chemicals.

## **MATERIAL AND METHOD**

The present study was carried out in the department of zoology, N.R.E.C. College, Khurja for the period 2008 to 2010. The present study was designed to observe the toxicity of a carbamate pesticide methylamine on two freshwater food fishes *Clarias batrachus* (Mangur) and *Heteropneustes fossilis* (Singhi) concerning histopathological aspects. Different types of insecticide carbamate pesticides were commonly used in agriculture operations and in controlling insect pests in the area of western Uttar Pradesh, including the vicinity of Khurja, District Bulandshahr. The carbamate pesticide has been selected for present investigation. Its chemical name is methylamine with molecular formula  $CH_3NH_2$ , molecular weight 31.06, soluble in water, boiling point 630c and melting point: 93.50c. Methylamine is used to control the broad spectrum of chewing and sucking insects and mites in a large variety of crops, especially rice, cotton, and wheat, etc. It is toxic to the body and central nervous system of the pests. It is an inhibitor of acetyl choline stress. The reason for selecting these *Clarias batrachus* and *Heteropneustes fossilis* fishes is that they are well known for their food value and easily available in fish market at a much lesser price as compared to the carps. External characteristics of *Clarias batrachus* are Dorsal fin, anal fins but separate from caudal, pectoral fins with a pungent poisons

spine, scales absent, four pairs of barbels present, dendritic accessory respiratory organs dorsal to gills. External characteristics of *Heteropneustes fossilis* fish, skin without scales, body elongated and laterally compressed, barbels long and four pairs, dorsal fin is short without a spine, pectoral fins long with a sharp poisonous spine, ventral fin situated at the level of the dorsal fin, accessory breathing organs present, and air bladder also present. This fish is highly nourishing and esteemed as food.

## **METHODOLOGY**

For the experimentation following methods were applied for the studied.

### **Histopathological Studies:-**

Fresh and healthy specimens of *Clarias batrachus* and *Heteropneustes fossilis* were procured from local fish dealers, which were approximately same in size and weight, used for bioassay experiments. Arora et al., (1971a) showed bioassay studies of some commercial organic insecticides on exotic carp. After disinfecting them with 0.1% potassium permanganate ( $KMnO_4$ ) solution, they were accumulitized for the duration of 15 days in the laboratory conditions. During this period fishes were provided no food. The series of experiments were set up to determine the effect of the pesticide methylamine on the tissue under study i.e. gill, liver and kidney. Median tolerance limit for 96 hours (in which 50% of the experimental fishes died within 96 hours) was treated as the lethal concentration which was 0.027 ml/liter. Five fishes were kept in the first series of lethal concentration. Sublethal concentration (chronic treatment) i.e. in which no mortality occurred for a longer period was taken in the second set of experiment. The sublethal concentrations selected was 0.009 ml/liter which was 1/3 of the median tolerance limit for 96 hours. Five fishes each were exposed in this concentration i.e., 0.009 ml/ liter for 30 and 60 days. In chronic exposure, the solution was changed regularly after 24 hours interval. Control experiments were also conducted side by side for comparison. In control experiments, the fishes approximately of the same size and equal in number were kept, and

both experimental and controlled fishes were not fed with diets. The temperature was maintained at  $21 \pm 300c$  for these experiments. After a definite period of exposure fishes of both series, control and treated (acute and chronic) were sacrificed and the required tissues i.e. gills. Anees (1978) studied sublethal and chronic levels of three organophosphorus insecticides on the hepatic pathology of the freshwater teleost. *Channa punctatus* (Bloch). Anthony et al., (1986) also showed the effect of sublethal levels of diazinon on histopathology of the liver. Arechon and Plumb (1990) demonstrated the sublethal effect of malathion on channel catfish *Ictalurus punctatus*. Bana (1992) showed toxicity of pesticides of extensive agricultural use in Meerut district to a freshwater teleost *Clarias batrachus* (Linn) and their effects on individual tissues. Bana, Bhatnagar, and Tyagi (1993) studied histological change due to malathion poisoning on *Clarias batrachus*. Banerjee and Paul (1993) showed an estimation of acute toxicity of ammonium phosphate to the fresh water catfish *Heteropneustes fossilis*. Aguiwo (2002) studied the effect of cympush pesticides on growth and survival of African catfish, *Clarias garipinus*. Balasubramani and Pandian (2008) found endosulfan suppresses growth and reproduction in zebrafish.

## RESULT AND DISCUSSION

### (1) Histomorphology of Gills:-

Fishes are the first vertebrate where gills evolved for gaseous exchange and are mostly composed of a highly complex vasculature, surrounded by a large surface area epithelium that provides a thin barrier between fishes blood and aquatic environment. The gill of *Clarias batrachus* and *Heteropneustes fossilis* are located near the head region and composed of five paired gill arches on both lateral sides of the pharynx. Anchored to the gill arches is a complex arrangement of epithelium, circulatory, and neural tissues. Gill filaments are the basic functional unit of gill tissues, long and narrow projections lateral to the gill arch that taper at their distal end. Each filament is supplied with an afferent filament artery that extends along with the filament. Blood

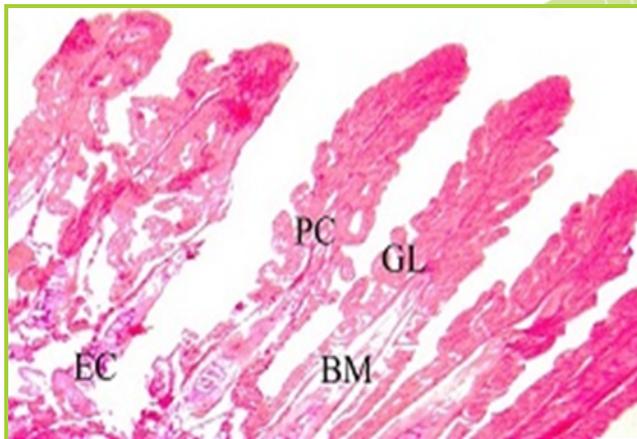
in this vessel also travels across the filaments breadth through numerous folds on the dorsal and ventral surfaces of the filament-termed as lamellae, lying perpendicular to the filaments long axis. Blood that crosses the lamellae drains into an afferent filament artery that runs along the length of the filament and carries blood in the opposite direction to that in the afferent filament artery. The region of the filament that contains the afferent blood supply is commonly referred to as the afferent edge, whereas the region that collects efferent blood is known as the efferent edge. These two terms are synonymous with trailing edge and leading edge, respectively, relative to water flow across the filament. Gill filaments contain three distinct vascular systems: (1) the respiratory circulation which receives the entire cardiac output and perfuses the secondary lamellae; (2) a nutrient system that arises from the post lamellar circulation and perfuse filament tissues; (3) a network consisting of subepithelial sinusoids surrounding afferent and efferent margins of the filament and traversing the filament beneath the interlamellar epithelium. Lamellae are evenly distributed along a filaments length, and the spaces between lamellae are channels through which water flows. Each lamella reveals that it is primarily composed of two epithelial sheets, held apart by a series of individual cells, termed as pillar cells. The spaces around the support cells and between the two epithelial layers are perfused with blood, flowing as a sheet, not through vessels. Lamellae dramatically increase the surface area of the gill filament epithelium and results in a small diffusion distance between the blood that perfused with each lamella and the respiratory water. Moreover, blood flow through the lamellae is countercurrent to the water which flows between them.

### (2) Histopathological stress in Gill due to Methylamine

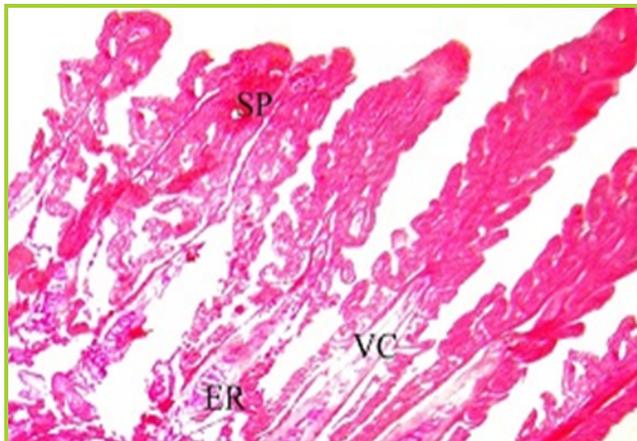
Sub lethal treatment:-

Gill shows severe erosion and fusion of secondary gill lamella and space in between basement membrane and primary lamellae

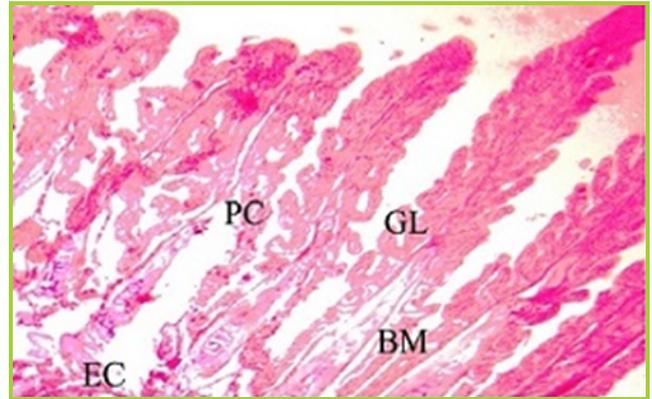
increased gradually. Vacuolization followed with lacunae formation was observed in secondary lamellae. Clumping of blood cells was of common occurrence. The general appearance of gill becomes altogether changed due to pesticide exposure within 30 days. Most prominent and severe changes were observed after 60 days exposure to methylamine. Secondary gill lamellae have been thoroughly lost. Basement membrane separated completely from primary gill lamella forming a continuous space. Further erosion of basement membrane was observed at different places. The shape of gills appeared as if they had been hypertrophied. The cells of the cartilaginous axis were found to be enlarged at places. Hyperemia was also observed. Fig:-1, 2, 3, 4,5,6,7 and 8 showing the status of gills of *Clarias batrachus* and *Heteropneustes fossilis* fishes.



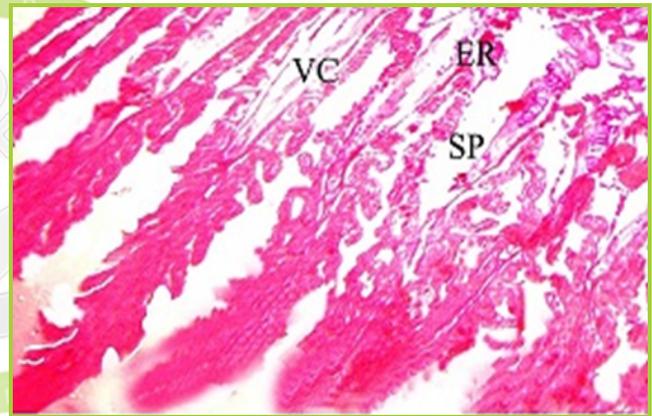
**Fig:-1 Microphotograph of Gill of *Clarias batrachus* as control for 30 days (T.S.) X - 150**



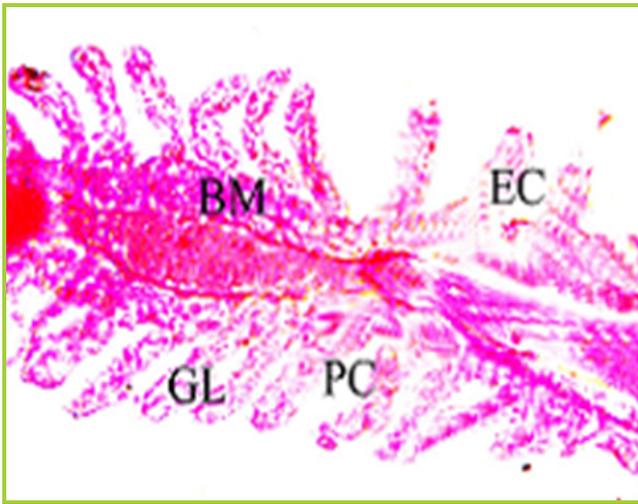
**Fig:-2 Microphotograph of Gill of *Clarias batrachus* at sublethal treatment for 30 days (T.S.) X - 400**



**Fig:-3 Microphotograph of Gill of *Clarias batrachus* as control for 60 days (T.S.) X - 150**

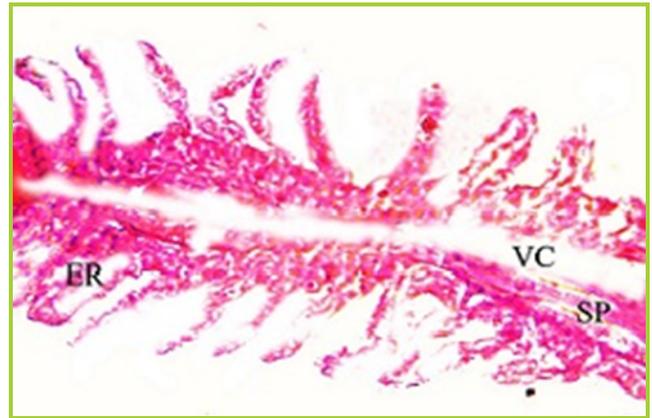


**Fig:-4 Microphotograph of Gill of *Clarias batrachus* at sublethal treatment for 60 days (T.S.) X - 400**

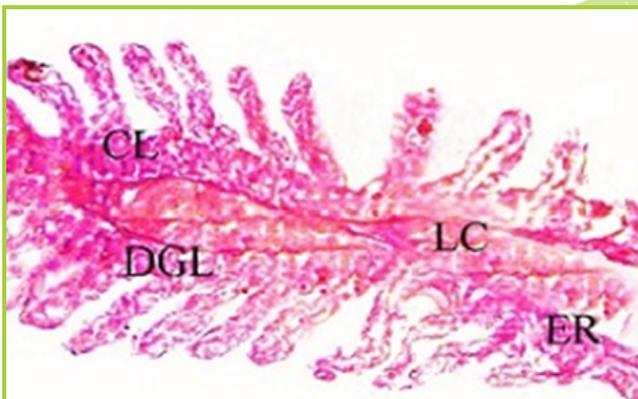


**Fig:-5 Microphotograph of Gill of *Heteropneustes fossils* as control for 30 days (T.S.) X - 150**

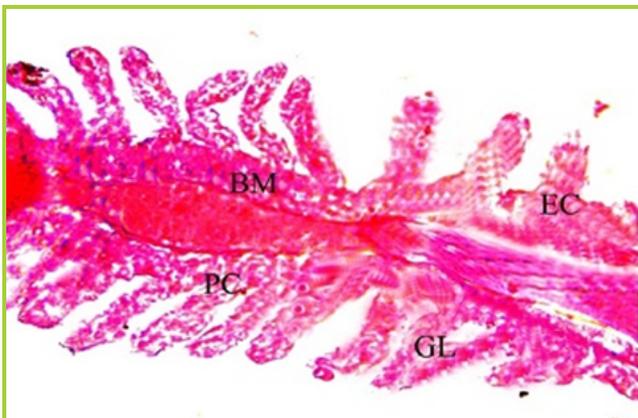
**Fig:-7 Microphotograph of Gill of *Heteropneustes fossils* for 60 days (T.S.) X - 150**



**Fig:-8 Microphotograph of Gill of *Heteropneustes fossils* at sub lethal treatment for 60 days (T.S.) X - 400**



**Fig:-6 Microphotograph of Gill of *Heteropneustes fossils* at sub lethal treatment for 30 days (T.S.) X - 400**



## CONCLUSION

This study demonstrated that day by day increasing the use of pesticides for insect control is posing a significant threat to freshwater organisms especially fishes. The discharged of agricultural waste by farmers, municipal and industrial wastes into the aquatic environment causes high mortality of fishes. Today about one thousand or even more chemical formulations are used as pesticides around the world, so pesticidal pollution is a global burning problem due to its extensive use. Protection of wildlife and water quality is not possible when using pesticides. If pesticides are selected wisely, used in combination with other pest control measures and applied safely, then the pollution of our surface waters and contamination of aquatic life can be avoided.

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