

# EFFECTS OF METHYL PARATHION ON HISTOLOGY OF GILL ON FRESHWATER FISH, OREOCHROMIS MOSSAMBICUS

Roopavathy J and Nandana P.K

Assistant Professor, PG and Research Department of Zoology, Nirmala college for women, Coimbatore -18

Master of Science, PG and Research Department of Zoology, Nirmala college for women, Coimbatore -18

Jroopavathy27@gmail.com

**ABSTRACT :** Parathion is an organophosphate insecticide and extensively used to control harmful insects of agriculture. The present study is aimed to study the toxicological of Methyl parathion on histology of gill on freshwater fish, *Oreochromis mossambicus*. This is done to determine the median lethal concentration ( $LC_{50}$ ) of methyl parathion on freshwater fish, *Oreochromis mossambicus* and to investigate the histological changes in the gill freshwater fish treated with methyl parathion. At the end of 15th day the fishes were dissected and the liver were removed from exposed and control group of fishes. The tissues were processed and sectioned at 4 $\mu$ m and then were stained with Haematoxylin- Eosin. The observation of the slides was done under light microscope at 40x magnification and photographed. The exposed group showed histopathological changes in the tissues. The present study reveal that the gills are susceptible to high degree of damage even at a very low amount of toxicant present in water. The histological changes observed in fish used in this study show that pollution is affecting fresh water fish living in these aquatic bodies and that this is seriously threatening an agricultural region. Urgent measures must be taken to correct this situation before it becomes a critical issue for the region.

**Key words:** *Oreochromis mossambicus*, Methyl parathion, secondary gill lamellae

## I.INTRODUCTION

India is primarily an agro-based country with more than 60-70% of its population dependent on agriculture. However, 30% of its agricultural produce is lost owing to pest infestation. In the absence of a better alternative, deployment of pesticides become inevitable despite their known hazardous effects. Utilization of pesticides in India is about 3% of the total world consumption and is increasing at the rate of 2-5% per annum (Bhadbhade *et al.*, 2002). With the increasing industrialization human beings are continuously disturbing the delicate ecological balance in aquatic ecosystems. Pesticides are mainly synthetic organic compounds that are deliberately introduced into the environment to control selected organisms (Roopavathy *et al.*, 2013)

Water is one of the precious liquid of the natural resource. A plentiful supply of clean water is essential for survival of human being, plants and animals. The disposal of industrial and agricultural waste directly into aquatic medium burdens the ecosystem (Nagarathamma and Ramamurthy, 1983).

Methyl parathion is one of the OP pesticides developed to substitute of organochlorides. OP are less persistent in the atmosphere, being easily linked to organic matter, being absorbed to sediments and particle material in suspension (EPA, 1986). Methyl parathion is extensively applied as an insecticide in agriculture, food storage shelters and pest control programs due to its high activity against a broad spectrum of insect pest. It is also widely used in fish culture systems to eliminate aquatic larvae of predatory insects that threaten fish larvae (Silva *et al.*, 1993). The acute toxicity as "stimulus severe enough to bring about response speedily usually within four days for fish". Though the acute toxicity tests, many workers emphasize more no sublethal toxicity than on acute toxicity (Roopavathy, 2016).

Acute toxicity tests are designed to measure the effects of toxic agents on aquatic species during a short period of their life span (Ebrahimpour *et al.*, 2010) and allow us to assess the effects of various pollutants on the biology of fish (Javed and Saeed, 2010) to find out their abilities to adapt under toxicity levels and to forecast possible effects of toxicity on them (Rauf *et al.*, 2009 and Azmat *et al.*, 2012).

However, the reports on the effects of methyl parathion in freshwater fishes are still scanty. In the light of above information and ideas, present investigation is aimed to study the effect of methyl parathion on histology of gill on freshwater fish *Oreochromis mossambicus*.

## II. MATERIALS AND METHODS

The present study was undertaken to investigate the acute toxicity and the effect of pesticide (*Methyl parathion*) on freshwater fish, *Oreochromis mossambicus*.

### 2.1 Selection Of Fish: *Oreochromis mossambicus*

The *Oreochromis mossambicus* (Mozambique tilapia) is a tilapia cichlid fish native to Southern Africa. It is an economically and commercially important fish and is easily adaptable to lab conditions. It is an ideal animal for toxicity studies in aquatic biology. It is a popular fish for aquaculture and it does not need much effort for maintenance. Due to human introduction it is now found in many tropical and subtropical habitats around the globe, where it can become an invasive species because of its robust.

### 2.2 Collection Of Fish

Fishes were procured from Chambath Farm, Palakkad with an average weight of 15-20 gram and 10cm length irrespective of sex. They were brought to the laboratory in well packed polythene bags containing oxygenated water.

### 2.3 Acclimatization Of Fish

Fishes were stocked in large plastic tubs, which were washed thoroughly prior introduction of fish to prevent functional infection. Fishes were acclimatized to laboratory condition for about 15 days before the commencement of experiment. Water was changed frequently to avoid fungal growth and contamination by fungal growth and contamination by metabolites.

### 2.4 Methyl Parathion (Test Chemical)

Methyl parathion is an organophosphorus pesticide and it is an active substance, extensively used as a pesticide in agriculture, food storage shelter, pest control programs and fish culture tanks to kill the aquatic larval stages of predator insects that threaten fish larvae. Methyl parathion is a highly toxic insecticide ranked by USEPA as a class 1 toxicant.

### 2.5 Estimation Of Lethal Concentration And Experimental Set Up

1gm of methyl parathion is mixed with 1000 ml of distilled water for preparing stock solution (0.1%). Appropriate narrow range of concentration (2.0, 2.2, 2.4, 2.6 and 2.8) was used to find the median lethal concentration using a minimum of 10 fishes for 96 hours. It was found as 2.4 ppm for 96 hours using probit analysis method (Finney 1964). Two groups of fishes were exposed to 10% of  $LC_{50}$  value and 30% of  $LC_{50}$  (0.24 ppm and 0.72ppm) concentration of pesticide methyl parathion for 15 days. Another group was maintained as control. For each experimental study tissue sample were collected from the fish for analysis of histology of gill.

### 2.6 Histology Analysis Of Tissue Sample

The histological sections of the gill of the control and experimental fish were taken by adopting the procedure as described by Humason (1972). The tissues were isolated from the control and phorate treated fish and rinsed with physiological saline solution (0.9% NaCl) to remove, blood, mucus and debris adhering to the tissues. They were fixed in Bouin's fluid for 24 hours and the fixative was removed by washing through running tap water overnight. The tissues were processed for dehydration using ethyl alcohol as the dehydrating agent and were passed through a graded series of alcohols, cleaned with methyl benzoate and embedded in paraffin wax. Sections were cut at 5 $\mu$  thickness and stained with hematoxylin and counter stained with eosin (dissolved in 95% alcohol). Then the sections were mounted in Canada balsam after dehydration and cleaning and photomicrographs were taken using Magnus photomicrographing equipment.

## III. RESULT AND DISCUSSION

In the present study *Oreochromis mossambicus* were exposed to methyl parathion at different concentration for a period of 15 days. The treated fish groups were compared with control group for the histological changes in gill and marked changes were observed. Teleost fishes have five pairs of gill arches. In the front, four pairs the slender gill filaments from two lines facing towards the back and these two lines are joined to each other at the base by a gill septum. The last pair of gill arches generally transforms into the pharyngeal bone and does not play a role in respiration, numerous semi-circular secondary gill lamellae are lined up along both

sides of the gill filament.

In the present study the *Oreochromis mossambicus* were exposed to sublethal concentration 0.24ppm and 0.72ppm for a period of 15 days. Shandha sathyanarayan *et al.*, (2012) studied the histopathological effects of pesticide on *Cyprinus carpio* which showed severe damage with shrinkage of the tips of secondary lamellae. He also observed lesions such as epithelial hyperplasia and curling of secondary lamellae on the gills, swelling and thrombosis of the tips of several secondary lamellae and club shaped secondary lamellae, the similar result we have observed.

After 15 days of exposure to 0.24ppm methyl parathion lamellar clumping, hyperplasia of secondary lamellae, curling of secondary lamellae, shrinkage of secondary lamellae and destruction of epithelium were noted. The histological analysis in the control fish showed normal structure but in the treated fishes the gill exhibits the abnormalities like hyperplasia of secondary lamellae, disruption of structure of mucous layer, shrinkage of secondary lamellae, epithelial desquamation, blood lamellar congestion, aneurism, lamellar clumping, hyperplasia of secondary lamellae and destruction of epithelium. Hyperplasia of secondary lamellae, disruption of structure of mucous layer, shrinkage of secondary lamellae, epithelial desquamation, blood lamellar congestion and aneurism were observed in the gill of the fish. Pesticide contamination of surface water from agricultural use is a problem of worldwide importance. Once a pesticide is introduced into the environment, there is a reasonable chance that it will eventually find its way into waters. Thus aquatic environs probably present one of the most important complex environment as far as describing the fate and behaviour of pesticide (Tahir Anwar *et al.*, 2005). In this study, blood lamellae congestion, lamellar fusion, hyperplasia of secondary lamellae, disruption of structure of mucous and epithelial layer, indistinct filament layer and secondary lamellae and disruption of striated epithelium were noted at low concentration. And at high concentration, complete disruption of secondary lamellae and blood sinuses, necrosis of filament, blood accumulation in primary lamellae and distinct cell structure were observed. All the histopathological observation indicates that the exposure to sublethal concentration of methyl parathion caused destructive effects in gill tissues of *Oreochromis mossambicus*.

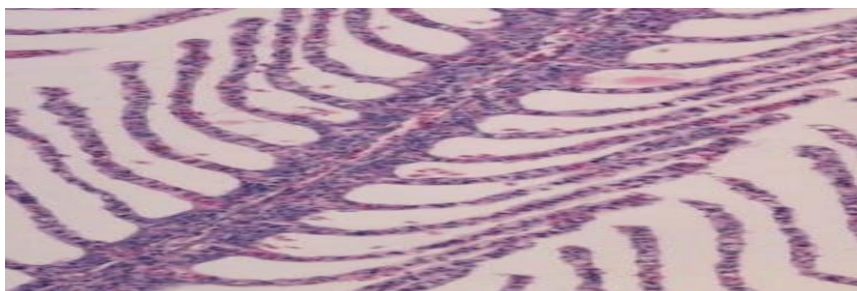


Fig.1. Control gill of *Labeo rohita* ; Primary gill lamellae (PGL), Secondary gill lamellae (SGL)

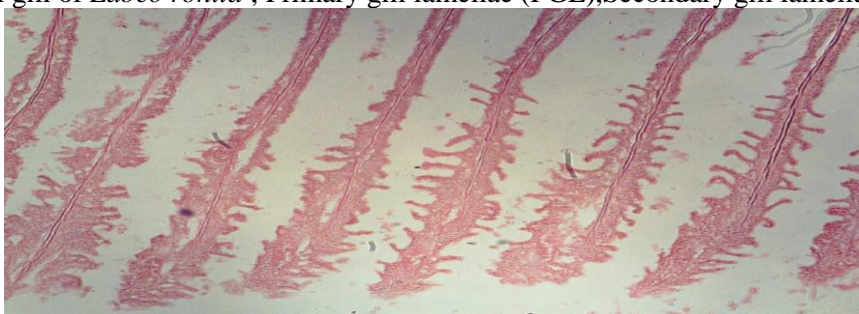


Fig.2: Gill of *Labeo rohita* after 15 days of exposure to 0.24 % leaf extract

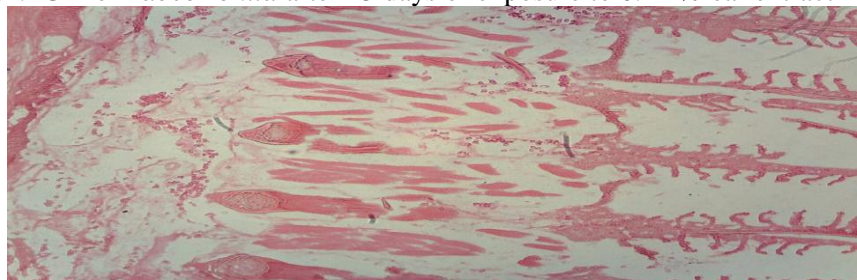


Fig.3. Gill of *Labeo rohita* after 15 days exposure of 0.72 % leaf extract

## CONCLUSION

The present study reveal that the gills are susceptible to high degree of damage even to a very low amount of toxicant present in the water. As the low concentration of the toxicants in the water may not be detectable by ordinary water analysis techniques and may be considered as within the safe limits, but in actual practise the vital organs especially the gills are adversely affected.

The histological changes observed especially in fish used in this study shows that pollution is affecting freshwater fish living in these aquatic bodies and that this is seriously threatening an agricultural region. Urgent measures must be taken to correct this situation before it becomes a critical issue for the region.

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