



# **Toxic Effects of Profenofos Based Insecticide on Freshwater Fish Mozambique Tilapia (*Oreochromis mossambicus*)**

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## **Authors' contributions**

*This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.*

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## **ABSTRACT**

This study was performed to investigate the toxic effects that associated with experimental exposure of mossambique tilapia to Profenofos based insecticide containing Profenofos 40% + Cypermethrin 4%. 96 hours exposures to 2.0068  $\mu\text{L/L}$  and 2.80952  $\mu\text{L/L}$  doses were conducted to evaluate the toxic effects, and various toxicological endpoints were assessed. Acute toxic stress was observed with fish displaying behavioral toxicity. Histopathological alterations in the liver, kidney, and brain occurred exclusively after treatment. The Histopathological effects of Profenofos based pesticide on the liver, kidney and brain tissues in *Oreochromis mossambicus* were determined by light microscopy. The fish were exposed to different concentrations of Profenofos

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based insecticide for 96 hours with parallel untreated control. No Histopathological effects were observed in control group. *dilation of Bowman's space, G: deterioration of glomerulus, RT: increase in diameter of renal tubule, CD: cellular degeneration*. Cellular degenerative processes can result in tissue necrosis in very severe scenarios. Collapse of glomeruli and blood hemorrhage and can lead to rise in the level of edematous fluid within interstitial substance were observed in kidney tissues exposed to Profenofos based insecticide. Hepatic lesions in the liver tissues of fish exposed to profenofos based insecticide were characterized by cytoplasmic vacuolation, cellular degeneration, damage of nuclei, bile stagnation as well as clogging in the blood sinusoids. Pesticides have the potentials of causing an increase or decrease of enzymatic processes and as such result in histological alterations. Noticeable alterations in Stratum opticum, Stratum fibrosum and griseum superficial, Stratum griseum central, Stratum album central and Stratum griseum periventricular. This study has shown that increase in concentration of pesticide with treated produced water resulted in degeneration and damage brain cells as well as clogging in the blood sinusoids. Pesticides have the potentials of causing an increase or decrease of enzymatic processes and as such result in histological alterations. From the obtained results, it is assumed that Profenofos induced general toxic impacts.

**Keywords:** Mozambique tilapia; profenofos; insecticide; tissues alterations; toxicology.

## 1. INTRODUCTION

"Profenofos (organophosphate) is among the major toxicant polluting freshwater bodies, producing a significant effect on fish health. Toxicity testing is critical for determining the impact and fate of toxicants in aquatic habitats. The acute toxicity trials on zebra fish (*Brachydanio rerio*), tilapia (*Oreochromis mossambicus*), and common carp (*Cyprinus carpio L*) have been estimated earlier [1,2,3]. The impacts of contaminants on fish survival rates and health, as well as human health, have generated cause of concern due to the proliferation of contamination in water. Contamination of water with large amounts of pesticides leads to fish mortality or starvation by destruction of food organisms. Moreover, many toxicants have been shown affecting the growth parameters and reproduction, with evidence of tissue damage.

Therefore, given the ecological impact of this pesticide, this study was carried out to explore the effect of Profenofos on the health status of Mozambique tilapia. For achieving the objective, the fish were exposed to LD<sub>50</sub> of Profenofos based insecticide for 96 hours. Moreover, the toxic effects were assessed on histopathological findings in certain organs in Mozambique tilapia after the exposure.

Selected insecticide protrin (PROFENOFOS 40%+ CYPERMETHRIN 4% E C) is a Broad Spectrum Insecticide widely used locally to control sucking pests as well as Bollworm, Thrips, Aphid, Jassid, Mealy bug etc. It is a strong contact and stomach insecticide having a

quick knockdown effect. It is also having acaricidal action. It can also control grown-up larvae, if resistance is not an issue.

Profenofos exposed biochemical alterations and histomorphological changes in Gills, Liver and Muscles have been reported earlier [4]. The present study was conducted to investigate the effects of *Oreochromis mossambicus* to 96-h LD<sub>50</sub> of protrin. Histological alterations in the liver, kidney and brain were used to assess the toxic effects of this pesticide.

Histology is the microscopic study of tissues through sectioning, staining and examining those sections under a microscope.

The *Oreochromis mossambicus* is one of the highly cultured commercial fish species in Bhopal. It is considered as a promising tropical aquaculture species, especially for low-income groups and a good source of protein. This fish showed excellent capacity to grow well in a wide range of thermal and environmental states. As it can withstand physical and chemical changes, and toxins in water, it's been often employed as a bio indicator of water pollution. Tilapia is now one of the most refereed model animals in aquatic toxicology investigations [5]

## 2. MATERIALS AND METHODS

Fish species of tilapia were collected using traps and gill nets from Patra Fish Seed Farm, Bhopal of Madhya Pradesh and acclimatized in seven fresh glass aquarium having 1.5ft x 1ft x 2ft in size. Dose of insecticide Protrin (Profenofos 40%

+ Cypermethrin 4% EC) were done as per LD50 calculation in aquarium. Aquariums were divided as 1 for control, 3 for treatment A (equal to concentration of LD50) and 3 for treatment B (equal to concentration of LD70). Fish were exposed to different concentrations of pesticide.

Liver, kidney and brain of fish were extracted and dehydrated to remove water from the tissue further procedure involves the immersion of samples in ascending grades of alcohol. Then tissues were cleared by immersion in xylene. Then impregnation is done to completely remove the clearing agent and tissues were embedded and sectioning is done using microtome. Then samples were stained, mounted and Photomicrography is done.

### 3. RESULTS

This study was designed to investigate the effects of Profenofos based insecticide on the Mozambique tilapia (*Oreochromis mossambicus*). Histopathological studies of liver,

kidney and brain of control and treated fish samples were performed.

#### 3.1 Histopathology of the Liver

Histological study showed a normal tissue internal arrangement pattern of the liver in the control (untreated) fish (Plate : 4.8.3-A). Results showed that no pathological alterations were observed in untreated fish as against various degrees of histological alterations exhibited by the exposed (treated) group (Plate : 4.8.3-B). Observed noticeable alterations included *BD: Bile duct, CV: central vein, CBD: close to a bile duct, BC: blood congestion, ND: nuclear degeneration, CN: cellular necrosis, ECV: expanded central vein*. This study has shown that increase in concentration of pesticide with treated produced water resulted in cytoplasmic vacuolation, cellular degeneration, damage of nuclei, bile stagnation as well as clogging in the blood sinusoids. Pesticides have the potential of causing an increase or decrease of enzymatic processes and as such result in histological alterations.

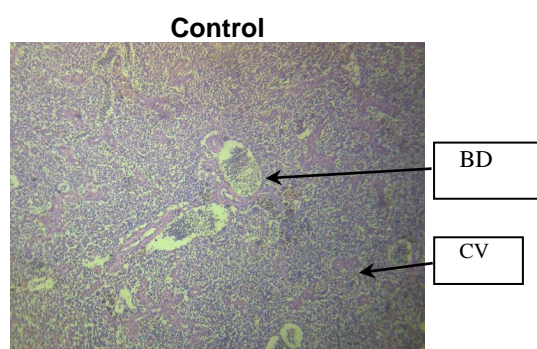


Plate: 4.8.3-A

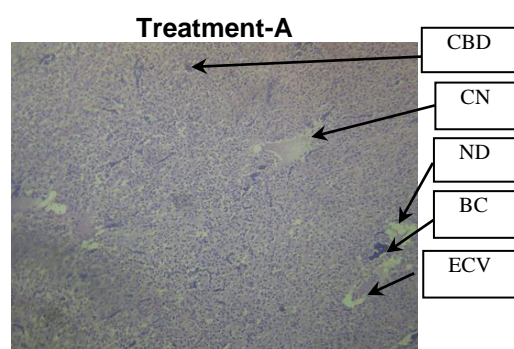


Plate: 4.8.3-B

**BD:** Bile duct, **CV:** central vein, **CBD:** closed bile duct, **BC:** blood congestion, **ND:** nuclear degeneration, **CN:** cellular necrosis, **ECV:** expanded central vein.

Fig. 1. Histopathological report of the Liver

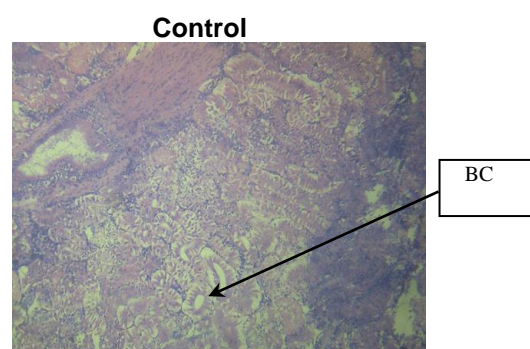


Plate: 4.8.2-A

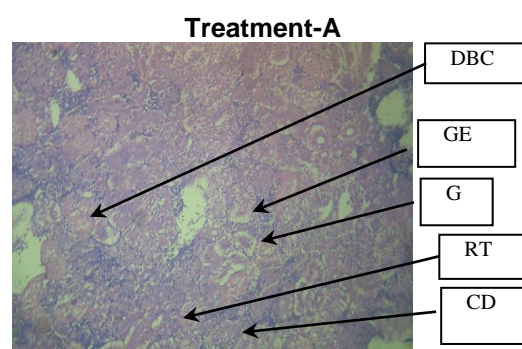
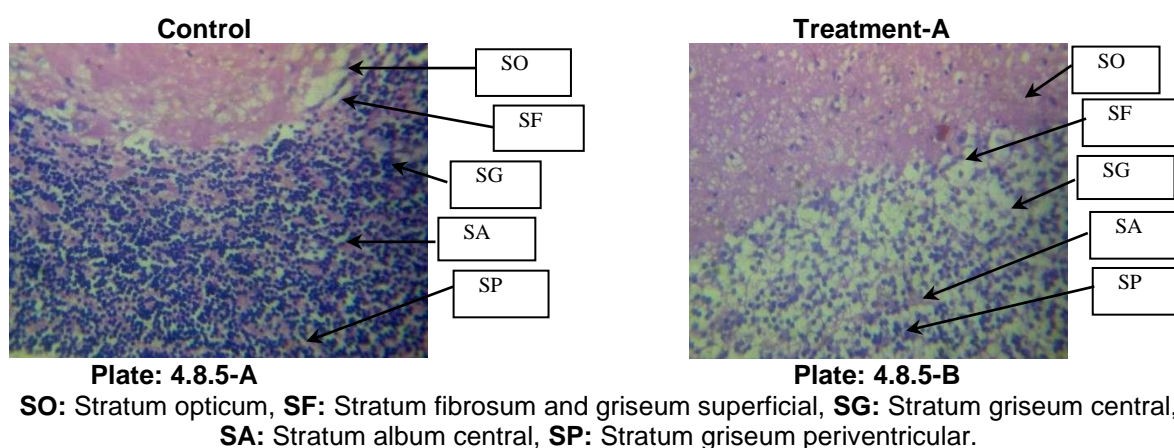


Plate: 4.8.2-B

**BC:** Bowman's space, **GE:** Glomerular expansion, **DBC:** dilation of Bowman's space, **G:** deterioration of glomerulus, **RT:** increase in diameter of renal tubule, **CD:** cellular degeneration

Fig. 2. Histopathological report of the kidney



**Fig. 3. Histopathological report of the Brain**

### 3.2 Histopathology of the Kidney

Histological examination of the kidney of *Oreochromis mossambicus* revealed a normal structural arrangement in control (untreated) fish (Plate: 4.8.2-A) as against various degrees of histological alterations exhibited by the exposed (treated) group (Plate: 4.8.2-B). The different histological changes observed were as follows: *BC:* Bowman's space, *GE:* Glomerular expansion, *DBC:* dilation of Bowman's space, *G:* deterioration of glomerulus, *RT:* increase in diameter of renal tubule, *CD:* cellular degeneration. Cellular degenerative processes can result in tissue necrosis in very severe scenarios. Collapse of glomeruli and blood hemorrhage can lead to rise in the level of edematous fluid within interstitial substance due to toxicities of pesticides in water.

### 3.3 Histopathology of the Brain

Histological study showed a normal tissue internal arrangement pattern of the brain in the control (untreated) fish (Plate: 4.8.5-A). Result showed that no pathological alterations were observed in untreated fish as against various degrees of histological alterations exhibited by the exposed (treated) group (Plate : 4.8.5-B). Observed slides had seen noticeable alterations in Stratum opticum, Stratum fibrosum and griseum superficial, Stratum griseum central, Stratum album central and Stratum griseum periventricular. This study has shown that increase in concentration of pesticide with treated produced water resulted in degeneration and damage to brain cells as well as clogging in the blood sinusoids. Pesticides have the potential of causing an increase or decrease of enzymatic processes and as such result in histological alterations.

Overall, these studies collectively emphasize the need for a multidimensional approach when investigating the Histopathological effects of pesticide exposure on tilapia, taking into accounts both structural changes in gill tissues and broader physiological responses.

## 4. DISCUSSION

The liver is the main organ for detoxification [6] that suffers serious morphological alterations in fish exposed to pesticides [7]. Alterations in the liver may be useful as markers that indicate prior exposure to environmental stressors.

"In another study, cloudy swelling, bile stagnation, focal necrosis, atrophy and vacuolization have been reported in the *Corydoras paleatus* exposed to methyl parathion" [8]). Hyperplasia, vacuolation, disintegrated blood vessels, disrupted hepatocytes, focal coagulative necrosis, disorganized hepatic canaliculi were observed earlier in *Labeo rohita* exposed to Cypermethrin [9]. Hepatic lesions in the liver tissues of fish *Gambusia affinis* exposed to deltamethrin were reported such as hypertrophy of hepatocytes, increase of Kupffer cells, circulatory disturbances, focal necrosis, fatty degeneration, nuclear pycnosis, narrowing of sinusoids [10,11,12]. "Tissue histology is regarded as a marker of exposure to pollutants and is an effective tool to evaluate the pollution level, particularly for sublethal impacts" [13]. "In the present study, Histopathological data revealed that Profenofos exhibited tissue alterations in fish liver, kidney, and brain. Several changes were produced, such as coagulative necrosis of hepatocytes with nuclear pyknosis, vacuolar degeneration, and accumulation of hemosiderin between hepatocytes. Changes in the liver could

be because the liver is the main site of detoxification, and it is expected that the toxicant would reach there abundantly for detoxification and disposal" [14]. "The inability of fish to regenerate new liver cells may also have led to necrosis of hepatic cells of sinusoids. The renal tissue showed narrowing of renal tubules, aggregation of inflammatory cells and hemolysis between renal tubules, degenerative and necrotic changes, severe depletion of hemopoietic tissue, and accumulation of hemosiderin between renal tubules. Lesions recorded in the kidney indicate nephrotoxicity caused by the tested compound and its metabolites since kidneys are the way to eliminate most of the organophosphorus compound. Vacuolar degeneration, severe atrophy and degeneration in muscle bundles with a focal area of necrosis were observed in muscles. Our results parallel findings on the histological changes of different fish species exposed to pesticides" [15]. Various liver and kidney alterations were revealed in PRO-exposed *Cyprinus carpio* [16] and in PRO-exposed *Egretta alba* [17].

Profenofos in the aquatic medium is a major factor responsible for drastic changes in the fish blood and tissues. Abnormal behavior and negative biochemical and Histopathological effects on the liver, muscles, and kidney were common features of fish health status due to Profenofos toxicity. So, this issue must be considered during toxicological analysis and control of the main aquatic pollutant.

## 5. CONCLUSION

We can conclude that the long term exposure of fish to protrin causes a continuous health hazard for the population and human population is also at high risk by consuming toxicated fish. The result of present study revealed that Profenofos based pesticide in the aquatic medium is a major factor responsible for drastic changes in the fish tissues. Abnormal behavior, and negative biochemical and Histopathological effects on the liver, brain and kidney were common features of fish health status due to pesticide toxicity. So, this issue must be considered during toxicological analysis and control of the main aquatic pollutant. Pesticides possess a high potential for endangering aquatic life because of its high toxicity. The rationalized use of pesticide considered the main factor in reducing aquatic pollution. Also must take the necessary precautions during the application of pesticides. If pesticides are selected wisely, used in combination with other pest control measures,

and applied safely, the pollution of aquatic resources can be avoided.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

## REFERENCES

1. Min KJ, Cha CG. Determination of the bioconcentration of phosphamidon and profenofos in Zebra fish (*Brachydanio rerio*) Bull Environ Contam Toxicol. 2000; 65:611–617. DOI: 10.1007/s0012800167. [PubMed] [CrossRef] [Google Scholar]
2. Rao RN, Madhavendra SS. Toxicity of chlorpyrifos to the fish *Oreochromis mossambicus*. Bull. Environ. Contam. Toxicol. 2003;70(5):985–992. Available:https://doi.org/10.1007/s00128-003-0079-0
3. Ismail M, Ali R, Ali T, Waheed U, Khan QM. Evaluation of the acute toxicity of profenofos and its effects on the behavioral pattern of fingerling common carp (*Cyprinus carpio* L., 1758) Bull Environ Contam Toxicol. 2009;82:569–573. DOI: 10.1007/s00128-009-9670-3. [PubMed] [CrossRef] [Google Scholar]
4. Parikh A, Miranda ER, Katoh-Kurasawa M, et al. Conserved developmental transcriptomes in evolutionarily divergent species. Genome Biol. 2010;11:R35. Available:http://doi.org/10.1186/gb-2010-11-3-r35
5. Yan W, Hamid N, Deng S, Jia PP, Pei DS. Individual and combined toxic genetic effects of micro plastics and heavy metals (Cd, Pb, and Zn) perturbgut micro biota homeostasis and gonadal development in marine medaka (*Oryzias melastigma*). J. Hazard. Mater. 2020;397:122795. Available:https://doi.org/10.1016/j.jhazmat.2020.122795
6. Dutta HM, Adhikari NK, Singh PK, Munshi JS. Histopathological changes induced by malathion in the liver of a freshwater catfish, *Heteropneustes fossilis* (Bloch); 1993.
7. Rodrigues EL, Fanta E. Liver histopathology of the fish *Brachydanio rerio* after acute exposure to sublethal levels of the organophosphate dimethoat 500; 1998.
8. Fanta E, Rios FS, Romão S, Vianna ACC, Freiburger S. Histopathology of the fish



- Corydoras paleatus contaminated with sublethal levels of organophosphorus in water and food. *Ecotoxicol. Environ. Saf.* 2003;54(2):119.
9. Sarkar B, Chatterjee A, Adhikari S, Ayyappan S. Carbofuran- and cypermethrin-induced histopathological alterations in the liver of *Labeo rohita* (Hamilton) and its recovery. *J. Appl. Ichthyol.* 2005;21(2):131–135. Available: <https://doi.org/10.1111/j.1439-0426.2004.00590.x>.
  10. Cengiz EI Unlu E. Sublethal effects of commercial deltamethrin on the structure of the gill, liver and gut tissues of mosquito fish, *Gambusia affinis*: A microscopic study. *Environ. Toxicol. Phar.* 2006; 21:246-253.
  11. El-Houseiny W, Abd El-Hakim YM, Metwally MMM, Abdel Ghfar SS, Khalil AA. The single or combined Silybum marianum and co-enzyme Q10 role in alleviating fluoride-induced impaired growth, immune suppression, oxidative stress, histological alterations, and reduced resistance to *Aeromonas sobria* in African catfish (*Clarias gariepinus*) *Aquaculture.* 2022;548:737693. DOI: 10.1016/j.aquaculture.2021.737693. [CrossRef] [Google Scholar]
  12. El-Houseiny W, Khalil AA, Abd-Elhakim YM, Arisha AH, Moselhy AA, Dahshan H, Ahmed MM. Alleviative effects of dietary Silybum marianum and co-enzyme Q10 on waterborne nickel-induced impaired growth, immunosuppression, tissue damage, immune-related genes dysregulation, and reduced resistance to *Pseudomonas aeruginosa* in *Oreochromis niloticus*. *Aquaculture Reports.* 2022; 26:101308. DOI: 10.1016/j.aqrep.2022.101308. [CrossRef] [Google Scholar]
  13. El-Houseiny W, Algharib SA, Mohamed EAA, Metwally MMM, Mahmoud YK, Alghamdi YS, Soliman MM, Abd-Elhakim YM, El-Murr AE. Dietary parsley seed mitigates methomyl-induced impaired growth performance, hemato-immune suppression, oxidative stress, hepato-renal damage, and *Pseudomonas aeruginosa* Susceptibility in *Oreochromis niloticus*. *Antioxidants.* 2022;11:1185. DOI: 10.3390/antiox11061185. [PMC free article] [PubMed] [CrossRef] [Google Scholar]
  14. Mushigeri SB, David M. Fenvalerate induced changes in the Ach and associated AchE activity in different tissues of fish *Cirrhinus mrigala* (Hamilton) under lethal and sub-lethal exposure period. *Environ Toxicol Pharmacol.* 2005; 20(1):65–72. DOI: 10.1016/j.etap.2004.10.011. [PubMed] [CrossRef] [Google Scholar]
  15. Mohamed WA, El-Houseiny W, Ibrahim RE, Abd-Elhakim YM. Palliative effects of zinc sulfate against the immunosuppressive, hepato-and nephrotoxic impacts of nonylphenol in Nile tilapia (*Oreochromis niloticus*) *Aquaculture.* 2019;504:227–238. DOI: 10.1016/j.aquaculture.2019.02.004. [CrossRef] [Google Scholar]
  16. Rahman ANA, Mohamed AAR, Mohammed HH, Elseddawy NM, Salem GA, El-Ghareeb WR. The ameliorative role of geranium (*Pelargonium graveolens*) essential oil against hepato-renal toxicity, immunosuppression, and oxidative stress of profenofos in common carp, *Cyprinus carpio* (L.) *Aquaculture.* 2020;517: 734777. DOI: 10.1016/j.aquaculture.2019.734777. [CrossRef] [Google Scholar]
  17. Taha A. Assessment of non-target toxicity of profenofos insecticide on the aquatic bird; the white egret egretta alba. *Egyptian J Aquatic Biol Fish.* 2022;26(2):263–276
- Tietz NW. Clinical guide to laboratory tests, 2nd ed. W.B. Saunders Company, Philadelphia, USA. 1990;554–556.