

## Fish as Bioindicators of Pesticide-Induced Histological and Biochemical Toxicity

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### SUMMARY

Fish is considered an important aquatic organism, but it is facing multiple threats from anthropogenic activities, environmental factors, and climate conditions. As a result, these threats not only decline fish species, but they also lead to instability economically, nutritionally, and environmentally. The main objective of this review is to explore the threats to the aquatic biodiversity of the world, including Pakistan. The main threats that are critically emerging and posing significant damage are the addition of agrochemicals in rivers and lakes, unchecked discharge from industrial zones that are mostly sources of heavy metals, unintentional release of sewage that is a source of eutrophication, and changes related to the water medium. These are leading sources of habitat destruction and loss of spawning spots for fish species, deterioration of water quality, and making fish species vulnerable to disease. Additionally, Pakistan has a diverse variety of aquatic ecosystems that contain freshwater and marine fish species. These species have high commercial and ecological importance. It is of utmost importance to protect aquatic biodiversity by identifying key roles and ensuring environmental stability. The findings of the literature emphasize biodiversity conservation, toxicological assessment, and environmental sustainability approaches. This review provides future research directions related to biodiversity protection and stakeholder engagement for policy development and implementation with proper assessment.

**Keywords;** Pollution, Aquatic contamination, Pesticides, Ecotoxicological biomarkers

### INTRODUCTION

Pesticides are substances used to control or combat pests (Abd et al., 2025), which are helpful in production. These pesticides are classified based on their target species (Spanoghe et al., 2025) such as insecticides, fungicides, and herbicides (Faiz, 2022). The proper use of pesticides is beneficial for crop productivity by controlling diseases (Ye et al., 2025) caused by pests. However, the unnecessary and improper use of pesticides leads to extensive damage to the environment (Parven et al., 2025) including aquatic and terrestrial ecosystems (Altaf et al., 2015; Altaf, 2016). They can accumulate in the bodies of organisms, water, air, and soil, leading to bioaccumulation and biomagnification in the environment. When these pesticides reach water bodies, they effect non-target aquatic organisms particularly fish causing hemato-biochemical, histopathological, behavioral, oxidative stress, and genotoxic effects. Furthermore, these pesticides can cause extensive damage to particular organs and inhibit or lower hormones production, leading to endocrine disruption (Liu et al., 2025), uncontrolled cell division leading to cancer, complications in gestation leading

to reproductive toxicities or impairment in aquatic and terrestrial organisms. The growing alarm over contamination due to toxicants, particularly pesticides, necessitates urgent focus on mitigation strategies or alternative source to reduce pesticide-induced toxicity in wildlife.

Pesticides induced contamination in the aquatic ecosystems includes multiple direct and indirect sources (Gul et al., 2025), such as runoff, atmospheric deposition, leaching and volatilization (Iqbal et al., 2025), resulting into loss of biodiversity. The persistence and dispersion of pesticides in aquatic environment depends on the chemical nature of pesticide. Mainly, pesticides persistence is more on the surface water as compare to column or sediment. Along this, these harmful chemicals are discarded in the terrestrial environment, which are more potent to human (Tabassum et al., 2025). The mortality and survival of fish depends on the concentration of pesticide (Alvim et al., 2025). Pesticides can be reached into fish body through ingestion of food content (Asad et al., 2024). In the water, fish is considered important biomarker of pesticide pollution (Oudah, 2024). Histological and biochemical toxicities are direct indicator of pesticides induced toxicity in the fish body (Rohani, 2023).

The increasing use of pesticides in farming has led to significant pollution of water bodies, which adversely affects sensitive organisms, such as fish. This exposure results in severe damage at the tissue and cellular levels, making fish important bioindicators of environmental pollution. However, comprehensive studies on the impact of various pesticides on fish health are scarce in Pakistan, a nation with rich yet vulnerable aquatic biodiversity. Therefore, this research aims to examine the histological and biochemical changes in fish tissues due to pesticide exposure and to highlight their potential as biomarkers for evaluating the health status of aquatic environments.

## **PESTICIDES POLLUTION IN AQUATIC ECOSYSTEM**

Aquatic ecosystem is fundamental area of aquatic organism to survive (de Oliveira Sousa et al., 2025). Mostly, pesticides are found in the aquatic ecosystem due to agricultural activities, particularly in developing nations such as Pakistan. But the presence of pesticides in any site of the aquatic body is vulnerable mark of toxicity for aquatic organism including fish (Boro et al., 2025). The presence of single pesticide in water can induce toxicity in fish but when these pesticides form mixture then they cause diverse damage. So, both minute or large dose can broad significant threat for fish.

## **FISH AS BIOINDICATORS OF AQUATIC POLLUTION**

The toxicity of pesticides in the aquatic environment is mostly evaluate through fish as model organism (Muhammad et al., 2019a; Muhammad et al., 2019b; Altaf et al., 2021; Altaf, 2021; Muhammad and Abideen, 2022; Mansour et al., 2025), due to its physiology, ecology and sensitivity to pesticides and other eco-contaminants. Fish exposed directly to toxicants in aquatic bodies. Due to wide range biomarkers in fish body, it has hematological, behavioral, oxidative stress and histopathological examination which helpful in the toxicity evaluation of contaminants particularly pesticides and heavy metals. In fish species, the most widely used fishes are carps,

tilapias and catfishes with speedy reproduction rate, maintenance and genetic variations (Kwikiriza et al., 2025). These characteristics make fish as suitable model organism to assess the health of aquatic ecosystem and also helpful for indication of environmental disruption.

### HISTOLOGICAL EFFECTS OF PESTICIDES ON FISH

Fish exhibited wide range of histopathological alterations due to exposure to pesticides (Maurya et al., 2019), which lead to damages in the particular organs. These particular organs include gills, brain, liver, kidney, muscles and intestine (Mokhtar, 2021). Gills are the primary organ in the fish which direct expose to pesticides and changes induced such as uplifting, fusion, hyperplasia and necrosis. Liver is detoxifying organ in organisms including fish, which also showed damages due to pesticides exposure, that include degeneration, necrosis, vacuolization and dilation in hepatocytes. The osmoregulatory organ kidney is also experienced disruption such as degeneration, shrinkage and inflammation. These organ damages are the early detection of contamination with potential toxicity of pesticides in fish. So, we can predict the ecotoxicological damages and risk assessment through the use of histopathological approach.

**Table 1: Histological Alterations in Fish Due to Pesticide Exposure**

Fish Species	Pesticide Exposure	Histological changes	Tissue/Organ Affected	References
<i>O. niloticus</i>	Thiamethoxam, Chlorpyrifos, and glyphosate	Lamellar disruptions, epithelial uplifting, degeneration and necrosis	Gills, Liver, testes, ovary and muscles	(Barb, 2025; Mansour et al., 2025; Nanthanawat et al., 2025)
<i>L. rohita</i>	Emamectin benzoate	Necrosis, degeneration, and lamellar disruptions	Liver, kidney and gills	(Kumar et al., 2025b)
<i>C. catla</i>	Cypermethrin	Necrosis, vacuolization, hepato-degenerative cells	Liver	(Sharma and Jindal, 2020)
<i>C. carpio</i>	Afidopyropen	Hemorrhages, necrosis, degeneration and vacuolization	Liver and Kidney	(Dodamani et al., 2025)
<i>C. idella</i>	Lufenuron	Congestion, dilation, vacuolation, degeneration and necrosis	Kidney and liver	(Khan et al.)
<i>C. punctatus</i>	Cypermethrin and malathion	Degeneration, pyknosis, splitting and vacuolization	Liver, kidney, muscles and gills	(Kumar et al., 2025a)
<i>D. rerio</i>				
<i>C. gariepinus</i>	Cypermethrin, Butachlor	Steatosis, pyknosis, degeneration, and Karyolysis	Kidney, gills, and liver	(Ibrahim et al.; Opute and Mbajorgu, 2025)

A study (Maurya et al., 2019) reported the extensive damage such as necrosis, hepatocytes degeneration and gills structural damage in *Heteropneustes fossilis*. Fish species exposed to pesticides in riverine system showed a wide range toxicity including liver, gills, and kidney (Tahir et al., 2021). Carbofuran exposure to *O. niloticus* showed necrosis, degeneration, epithelial uplifting, apoptosis and nuclear damages (Américo-Pinheiro et al., 2020). *O. mossambicus* exposed to heptachlor caused dilation, hypertrophy, degeneration and lamellar breakdown (Manimekalai et al., 2022). A pesticide triflupyrzimid exposure to *Labeo rohita* caused damages in gills such as uplifting, lamellar degeneration, hypertrophy and necrosis while apoptosis and degeneration were also observed in hepatocytes (Nayak et al., 2023). Table 1 emphasizes the various histological alteration due to pesticides exposure.

### BIOCHEMICAL EFFECTS OF PESTICIDES ON FISH

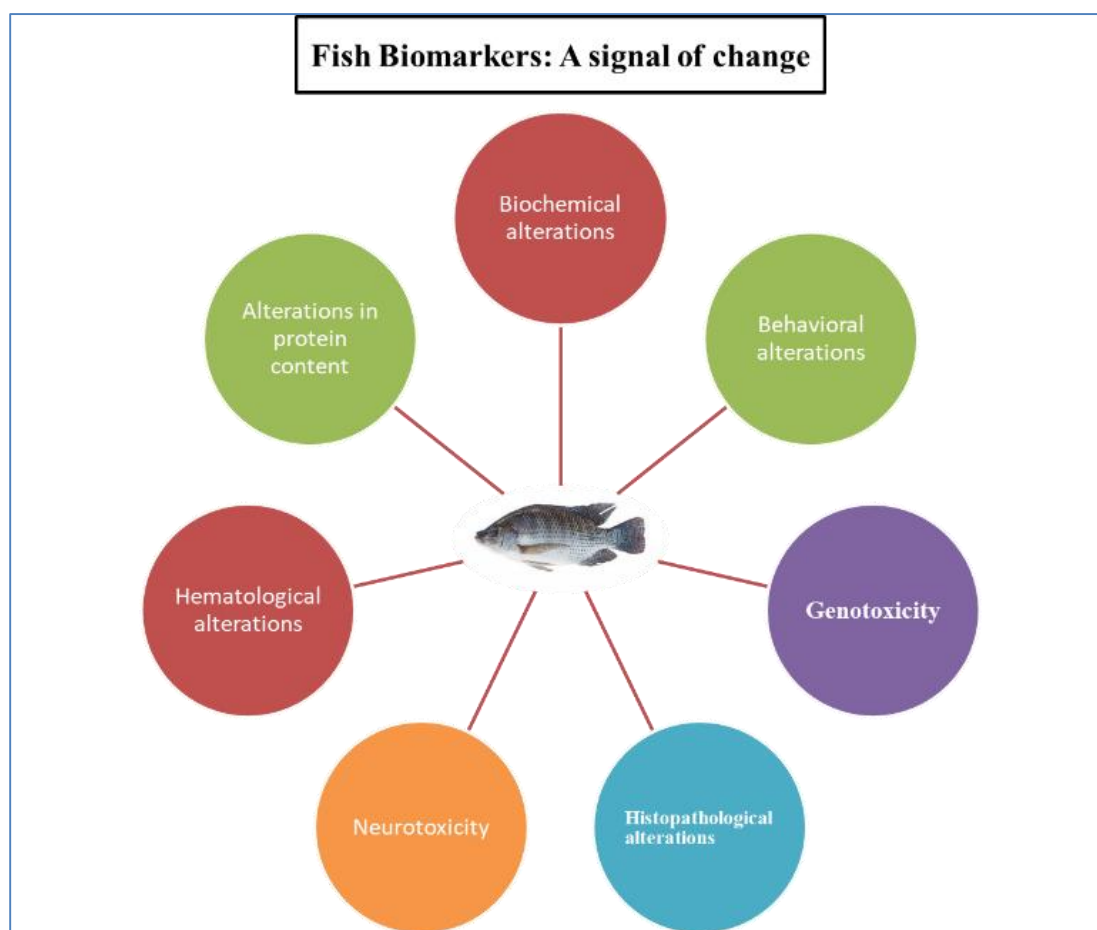
Pesticides cause disparity between the antioxidant enzymes of the organism and reactive oxygen species (ROS) (Rajak et al., 2025), resulted oxidative stress. The accumulated pesticides in the fish lead to biotransformation, which ultimately cause induction or generation of reactive oxygen species. The generated stress of ROS lead to enzymatic inhibition, mutation and membrane rupturing. Pesticides also cause oxidative stress in fish, which can measure to lipid peroxidation level in the fish body (Barb, 2025). During this stress, antioxidant enzymes such as CAT, SOD, POD and GPx showed decrement. Oxidative stress cause destruction of cellular tissue level damages that is more important biomarker for assessment of the long-term exposure of pesticides. These studies reported the oxidative stress in fish species such as chlorpyrifos and atrazine caused oxidative stress in common carp (Xing et al., 2012), deltamethrin in *Clarius gariepinus* (Amin and Hashem, 2012), chlorpyrifos in silver barb (Hossain et al., 2025), malathion and cypermethrin in *Channa punctatus* (Kumar et al., 2025a), roundup in goldfish (Noor and Rahman, 2025) and thiamethoxam in *O. niloticus* (Mansour et al., 2025). Table 2 depicts the important biomarkers that selected in toxicological studies.

**Table 2: Biochemical Biomarkers Used in Fish Toxicity Studies.**

Biomarker	Type	Role/Function	Response to Pesticides	References
Catalase (CAT)	Antioxidant enzyme	Hydrogen peroxide breakdown	Increased or decreased	(Xu et al., 2022)
Glutathione	Antioxidant enzyme	Remove peroxidase	Increased or decreased	(Averill-Bates, 2023)
Acetylcholinesterase (AChE)	Neurotoxicity biomarker	Nerve signaling	Decreased	(Luque and Muñoz-Torrero, 2024)
Alanine Aminotransferase (ALT)	Liver enzyme	Liver damage indicator	Increased	(Valenti et al., 2021)
Aspartate Aminotransferase (AST)	Liver enzyme	Liver damage indicator	Increased	(Ndrepepa and Medicine, 2021)
Alkaline phosphatases (AP)	Liver enzyme	Liver or bile duct damage indicator	Increased	(Makris et al., 2023)
Lipid Peroxidation (LPO)	Oxidative stress biomarker	Oxidative damage indicator	Increased	(Valgimigli, 2023)
Superoxidase dismutase (SOD)	Antioxidant enzyme	Superoxide to hydrogen peroxide conversion	Increased or decreased	(Rosa et al., 2021)

### BIOMARKERS IN FISH FOR PESTICIDE TOXICITY

Biomarker is defined as the signal which indicate the unintentional body state of organism. In fish, a number of biomarkers are used to assess the potential toxicities of contaminants particularly pesticides. These can be used for toxicities caused due to short term or long-term exposure to contaminants (Zolghadri et al., 2023). Hematological, histopathological, Serum-biochemical, oxidative stress, genotoxicity, neurological and behavioral assessments are the biomarkers which assist to evaluate the potential damage due to toxicants, as described in figure 1. Hematological biomarkers are involved the investigation of blood profile of organism and observe changes in RBCS, Hb, MCH, MCHC, MCV, MPV, HCT and white blood cells. Serum biochemical biomarker used to assess the cholesterol, glucose, cortisol, triglycerides, albumin, globulin, bilirubin, ALT, AST, AP, HDL, LDL and VLDL. Histopathological biomarkers are investigated changes in organs such as brain, gills, liver, muscles and kidney. Behavioral biomarkers are indicator of apparent changes such as feeding, swimming patterns, stimulus response in fish due to exposure of toxicants. Oxidative stress biomarker includes changes in TBARS, ROS, SOD, CAT, GST, GSH, GR and AChE. In short, all of these biomarkers are detailed investigation of pesticides induced toxicities in organism including fish.



**Figure 1: Fish biomarkers for ecotoxicological studies.**

## **STATUS OF AQUATIC BIODIVERSITY IN PAKISTAN**

In Pakistan, alarming threat such as anthropogenic activities are increasing day by day (Abideen et al., 2023; Muhammad et al., 2024). These threats are due to our mismanagement and economic instability, which do not focus on the implementation of policies. Pakistan granted with diverse aquatic biodiversity in the aquatic bodies such as Indus, Jhelum, Chenab, Ravi and Sutlej, ponds, canals, coastal areas, lakes and streams (Ali et al., 2024; Muhammad et al., 2024; ALI and Sciences, 2025). These aquatic bodies are considered as hotspot zones for aquatic biodiversity including fish. This diverse aquatic ecosystem might be making possible to Pakistan, with 231 freshwater fish species while 522 marine fish species. Regrettably, fishes are facing alarming threat with decline in population (Prins et al., 2025). Particularly, juvenile fish population is under threat due to illegal netting and electrofishing. Freshwater aquatic bodies are the sites which are under threats due to eutrophication and sediment accumulation in aquatic bodies (Fujimoto et al., 2025). These changes need utmost attention to implement rules, monitoring and exploring the native diversity with careful strategies.

## **EFFECTS OF PESTICIDES ON BIODIVERSITY**

Pesticide causes disruption in the environment due to targeting beneficial organism, which is leading threat of biodiversity damage worldwide. The extensive use of toxicants such as pesticides result decline in population, loss of species and habitat destruction. Insects are natural organism which helpful for the pollination of plants (Zariman et al., 2022), but improper use leading combat of beneficial insects due to toxicants particularly organophosphates and carbamates. Aquatic species particularly fish are facing threat of decline due to unintentional use and dumping of pesticides near to aquatic bodies. Soil microbes which are beneficial for maintenance ecological stability through cycling of nutrients (Srivastava et al., 2023), are also declining due to pesticides. The damage of beneficial organisms leading to disruption in ecological stability which mainly caused due to disruption of food chain and food web. A wide-range of pesticides are used depending on their purpose. Table 3 describe the pesticides in ecotoxicological studies.

## **MITIGATION STRATEGIES AND POLICY**

The toxicological condition induced by pesticides is hazardous condition for the survival of organism (Thakur et al., 2025). But it can be cope with the implementation of novel approaches of mitigating strategies such as integrated pest management, buffer zones, restoration techniques, less use of pesticides and disposal practices (Azeem et al., 2024). These strategies with regular monitoring of water might be helpful for the minimize toxicity in aquatic habitat. Furthermore, restoration approaches along these strategies also significant approach to ecosystem resilience. The cumulative stress of disease and pesticide exposure also a threat for fish health (Ashraf et al., 2024), which is emerging concern for biodiversity. By implementation of these strategies cannot helpful to only protection of fish but it can preserve the sustainability and ecosystem resilience.

**Table 3: Common Pesticides and Their Effects on Fish.**

Pesticides Type	Example Compound (s)	Targeted	Observed Effects in Fish	Affected Organs	References
Insecticide	Cypermethrin and Methomyl	Nervous system	Hepatotoxicity, reproductive toxicity, Hemato-biochemical toxicity, and Endocrine disruptions	Brain, Liver, gills, muscles, reproductive organs and Endocrine system	(Korkmaz et al., 2009; Meng et al., 2014)
Herbicide	Atrazine and glyphosate	Amino acids and photosynthesis of herbs	Histo-morphological, Oxidative stress and hemato-biochemical	Liver, gills and kidneys	(Xing et al., 2012; Liu et al., 2021; Khoshnood, 2024)
Fungicide	Thiram and Mancozeb	Cell membrane and sterol synthesis in fungi	Stress, hepatotoxicity and nephrotoxicity	Immune system, gills, kidney and liver	(Ibrahim et al., 2023; Alam et al., 2025)
Organochlorine	Endosulfan and DDT	Nervous system and sodium channel	Endocrine disruptions, hemato-biochemical toxicity, oxidative stress	Testes, brain, liver, kidney and endocrine organs	(Fadilah and Rahardja, 2023; Cossaboon et al., 2024; Kim et al., 2025)
Organophosphate	Chlorpyrifos and malathion	Nervous system	Neurotoxicity, Oxidative stress, hepatotoxicity	Brain, blood, nervous system, liver, muscles and gills	(Banaee et al., 2024; Varma and Vasudevan, 2025)

## CONCLUSION

Pesticides in the aquatic ecosystems are leading concern because they induced multi-organ damages with diverse modifications. These modifications pose tissue damages in vital organs of organism including fish, which ultimately lead to mortality. Histopathological and oxidative stress are true indicator of all types of the toxicities such as short term or long term in both aquatic and terrestrial environments. When a pesticide exposed in minute concentration to fish, it might be not indicated through other biomarkers but both of these will reveal induced toxicities. Mitigation strategies are good concern for the minimize pollution particularly pesticides, but proper management and strict regulation is necessary. In conclusion, aquatic biodiversity especially fish is under the alarming threat and need urgent concern for ecosystem sustainability and resilience.

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