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# A review on liver enzymes as useful biomarker to evaluate the effects of pesticides on freshwater fish

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### **Abstract**

Contaminated freshwater through the enormous use of toxicants like pesticides is unsafe for consumption, affecting public health, fishing, tourism, and the environment. Pesticide toxicity is a global issue, with most poisoning occurring in developing countries. Pesticides disturbed ecological equilibrium and have an adverse effect on a variety of creatures, which has major effects on aquatic ecosystem. They have the ability to directly poison fish and other aquatic invertebrates, impairing their ability to breathe. Pesticides can enter aquatic environments through runoff from agricultural fields, posing significant risks to aquatic life. Chemicals from pesticides, such as lead or copper, contaminate water bodies, and affect fish and other non-targeted species including human health too. Because of cumulative qualities of different synthetic pesticides they circulate in ecosystems and may be accumulated by various living organisms and even migrate through food chains and alter the functioning of different organs of living organism. Fish covered a major part of worlds food supply hence it is essential to secure the health of fish. Biomarker studies can inform regulatory decisions regarding pesticide use and environmental protection.

The liver, the most important organ of essential metabolic pathways, is negatively affected by toxic chemicals. Liver enzymes like ALP, ACP, AST, and ALT enzymes serve as vital biomarkers for assessing pesticide impacts on fish liver and blood. Monitoring changes in these enzyme activities can provide valuable insights into fish populations' health status and the overall environmental impact of pesticide use. A comprehensive understanding of how liver enzymes in freshwater fish can be used as biomarkers to assess pesticide impacts is needed to protect aquatic ecosystems from pesticide contamination. Changes in species composition, community organization, and diminished resilience to environmental stressors are examples of long-term ecological consequences. Integrated strategies, such as better pesticide management techniques, are needed to mitigate these effects.

**Keywords:** Pesticides; Freshwater Fish; Liver Enzymes; Aquatic Ecosystem

## **1. Introduction**

The use of pesticides in agricultural farming has led to environmental problems and the development of resistant pests. Insects are major pests in food and fiber production, acting as vectors for various disease organisms and infecting plants, animals, and humans. Pest control relies on large-spectrum poisonous pesticides, which pollute quality of water, air, and soil by discharging excessive formulations, extending crops to the water's edge, accidental spillage of agrochemical formulations, run-off and erosion, and fallout from air pollution. Untreated industrial discharge and domestic wastes fortify with contaminants like toxic pesticides, heavy metals, and organic compounds, which are routinely applied to protect crops and animals from pests, insects, weeds, and other diseases. Contaminated water is unsafe and not suitable for aquatic animals, including fish, as they are oversensitive to toxicants in water and can be used as bio-indicators in the investigation of aquatic ecosystems. Toxic chemicals and pesticides may have also an impact on algae and planktonic

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species, upsetting the food chain and influencing higher trophic levels. Through bioaccumulation, pesticides can built up in the tissues of aquatic species, increasing the poisons present in top predators. They can modify aquatic environment, interfere with the growth and reproduction of non-target species, and cause indirect harm to them. Fish, being the top aquatic trophic, represent a strong image of an aquatic ecosystem and pose a threat to human health when consumed as a food source. Fish have the ability to accumulate toxic materials from the environment in various body parts, making them a crucial bio-indicator in understanding aquatic ecosystems.

Routes of pesticides exposure in fish –

- By Dermally- Direct absorption through the skin.
- By Breathing- uptake of pesticides through the gills
- By Orally- drinking pesticide–contaminated water or consuming on pesticide- contaminated prey. It is also called 'secondary poisoning' when consumer consuming another animal that has been contaminated by a pesticide [1].

Fish can bio-concentrate pollutants in their body tissues, leading to behavioural disorders, histo-pathological changes in liver, gills, blood, hematopoietic tissue, and brain. Fish can pass these toxicants on to other animals, including humans, through bioaccumulation. Bioaccumulation and bio-magnification can also have prolonged effects on fish liver enzymes. Continuous exposure to low levels of pesticides can lead to chronic effects on liver enzyme activities and overall metabolic homeostasis (Figure-1).



Bioaccumulation = Food chain transfer + Bio-concentration – Elimination + Growth dilution

#### **Figure 1**Routes of pesticides exposure in fish

Research on the response of different fish species to synthetic pesticides is of particular significance for numerous reasons. First of all, it may consider the group's material in terms of economic activities, including human health. Secondly, the fact that the pesticides are known to bio-accumulate and even bio-magnify along food chains in progressively greater concentrations as they are incorporated into the diet of aquatic organisms [2], and as fish have higher trophic levels, may reflect not only the hazardous impacts of pesticides and the reactions of non-target species, such as those of economic importance, but also allow inferences relating to the state of whole food webs. The level of accumulated pesticides may pose severe health risks when assessing the intake of hazardous pollutants by humans.

## **2. The specific liver enzymes**

Liver enzymes are protein which producing bile and substances that helpful in blood clotting, breaking down food and toxins, and also fighting infection. ALP, ACP, AST and ALT is the most significant liver enzymes and their relationship with pesticide impacts on fish blood and liver detailed below:

#### **2.1. ALP (Alkaline Phosphatase)**

**Normal Function:** ALP is involved in the hydrolysis of phosphate esters in alkaline conditions. It plays a role in several metabolic processes, including bone mineralization and liver function.

**Impact of Pesticides:** Exposure to pesticides can lead to alterations in ALP activity in fish. Increased ALP activity is often observed as a response to liver damage or stress caused by pesticide toxicity. This enzyme's elevation can indicate ongoing tissue repair processes in the liver.

#### **2.2. ACP (Acid Phosphatase)**

**Normal Function:** ACP catalyzes the hydrolysis of phosphate esters in acidic conditions and is involved in cellular metabolism.

**Impact of Pesticides:** Similar to ALP, changes in ACP activity can indicate cellular damage in the liver due to pesticide exposure. Increased ACP levels can suggest cellular leakage and damage, reflecting the stress on liver tissues.

#### **2.3. AST (Aspartate Aminotransferase) and ALT (Alanine Aminotransferase)**

**Normal Function:** AST and ALT are key enzymes involved in amino acid metabolism and are primarily found in the liver and other tissues.

**Impact of Pesticides:** Elevated levels of AST and ALT in fish blood serum often indicate liver cell damage or necrosis caused by pesticides. These enzymes leak into the bloodstream when liver cells are damaged, making those useful indicators of liver health.

Liver enzymes adopt different modes of action, influenced by dose and duration of exposure. Enzyme assays are suitable biomarkers for assessing liver damage or stress. Field studies and lab studies are used to understand the real-world implications of pesticide exposure on fish populations. Changes in liver enzymes may impact fish health at the population level, affecting reproductive success, growth rates, and overall aquatic ecosystem fitness. Pesticides can impact fish liver enzymes through various mechanisms due to their toxicological properties. Fish liver enzymes are crucial in metabolizing foreign substances and can activate or detoxify pesticides, leading to the formation of reactive intermediates that can damage cellular components. Detoxification pathways involve enzymes conjugating pesticides to facilitate their excretion. Direct induction of specific liver enzymes can disrupt normal physiological functions, while oxidative stress can damage liver cells and impair enzyme function, affecting overall liver health and metabolic capacity. Endocrine disruption can occur due to pesticides affecting hormone synthesis and regulation, affecting fish reproductive and metabolic health.

Assessing the impact of pesticides on freshwater fish using liver enzymes as biomarkers is a crucial area of study. When exposed to pesticides, these enzymes can undergo changes in activity levels or expression patterns, which can serve as indicators of stress or damage to the liver and overall health of the fish. Additionally, these toxic compounds are related to the environmental pollution and accidental poisoning [3]. Pesticides, including organophosphates, organochlorides, carbamates, pyrethroids, and pyrethrin, can affect fish liver enzymes, affecting their metabolism and liver function. These effects are classified as physical, protoplasmic, respiratory, or nerve poisons, which interact with fish metabolism and liver function.

## **3. Pesticides and Alterations in liver enzymes in fish –**

Many researchers evaluated the effects of pesticides on fish and noted various alterations in liver enzymes. Jyothi and Narayan [4] suggested that liver enzymes are the most commonly used biochemical markers of hepatocellular necrosis. Similarly, elevation in ACP and ALP activities in plasma, kidney, and gill, while a significant inhibition in both activities was also observed in liver tissues by Rao [5] in his work due to the exposure of monocrotophos in *Oreochromis mossambicus*. Koul *et al.* [6] and Mastan and Ramayya [7] recorded biochemical alterations in *Channa gachua* after exposure to dichlorvos and noted an increase in alkaline phosphatase (ALP) in plasma, SGPT (ALT), and SGOT (AST) in

both acute and chronic studies. The increase in ALT and AST activities noticed in the study of Sivaperumal [8] supports tissue damage [9]. The elevation in alkaline phosphatase suggested an elevation in lysosomal mobilization and cell necrosis due to pesticide toxicity, which was also noted, indicating an increase in the activities of these enzymes in serum as a result of impairment of hepatic tissue and the liberation of these enzymes into circulation from the damaged tissues, also investigated by Sivaperumal [8]. Same as Kumar *et al.* [10] and Okonkwo *et al.* [11] reported elevations in liver enzymes after exposure to cypermethrin. These investigators had suggested that the increased liver enzyme activities were due to the stepped-up transamination, where amino acids are used to create intermediates for the tricarboxylic acid cycle, in an effort to manage the energy crisis during stress caused by the pollutants.

Karami-Mohajeri and Abdollahi [12] also found liver damage exposed to phorate and carbaryl in *Clarias batrachus,* which was thought to be a consequence of the production of lipolytic mitochondrial enzymes that dissolve lysosomal membranes, cell membranes, and hepatocellular organelles, hence setting free liver enzymes into the blood. However, some of these pollutants are long-term, causing notable alterations in enzymatic activities [13]. Fish convert and adapt to their metabolic activities during acute stress situations [14]. Such transformations have been realized through changes in biochemical markers assessed as indicators of tissue destruction. Similarly, Bhavika [15] reported a significant increase in alkaline phosphatase levels in the liver, kidney, gills, and muscle of *Oreochromis mossambicus* and *Labeo rohita* after exposure to imidacloprid and curzate. An increase in alkaline phosphatase levels may indicate liver and renal damage [16, 17]. Harabawy *et al.* [18] also concluded that the sub-lethal effects of synthetic pesticides in fish can be indicated by a remarkable elevation or decline in enzymatic activities. Elevation in the level of hepatic enzymes is due to oxidative stress caused by pesticide exposure, which results in the cell membrane destruction of hepatic cells and the outflow of these enzymes to the blood [19]. Rahman *et al.* [20] testified to a significant elevation in serum ALT, AST, and ALP levels in *C. carpio* treated with profenofos. Similar findings were also noted in the serum of *Oreochromis niloticus* exposed to endosulfan, *Oreochromis niloticus* exposed to deltamethrin, and *C. carpio* exposed to lufenuron [21, 22, 23]. Shah and Parveen [24] reviewed sub lethal concentrations of pesticide causes completely changes in the behavior, histology, physiology, hematology, defense mechanism etc. that becomes toxic to the aquatic animals including fish. Foreign compounds like pesticides are predominantly bio-transformed in the liver by the action of metabolizing enzymes along with aminotransferases, microsomal enzymes, and oxygenases, as suggested by Stickel *et al.* [25]. Hence, the liver has a prominent metabolic flexibility in retort to enzyme induction to a variety of metabolites, along with the capacity to manage the expression of genes coding for the biosynthesis of such enzymes. The disturbance of the purity of biochemical and physiological processes in fish has been observed by determining the alterations in the activities of enzymes in serum, plasma, and tissues of the liver, kidney, gill, brain, and muscle of the fish [26].

The activities of hepatic function enzymes (ALP, AST, and ALT) were significantly elevated on exposure to chlorfenapyr, dimethoate, and acetamiprid in the investigation of Ghayyur *et al.* [27]. They clearly demonstrated the negative impacts of chlorfenapyr, dimethoate, and acetamiprid pesticides on selected hormonal, histopathological, and hematological biomarkers of a freshwater fish, *C. mrigala*. The aftereffects of pesticides on liver cells may result in body metabolic changes as well [28]. In the study of Esenowo *et al.* [26], there was a significant increase in the AST, ALT, and ALP levels of African catfish, *Clarias gariepinus,* exposed to chlorfenapyr in comparison with the control, and it was indicated that acute exposure to chlorfenapyr can change liver enzyme activities and serum lipid profile, initiating a jump in the energy requirement of the exposed organism. The existence of specific enzymes in the blood may cause organ dysfunction because they are present in the tissue cells of the liver, kidney, heart, gills, muscles, and other organs. Ajibare *et al*., [29] also suggested that an increase in liver enzymes might be related to cell damage in liver stress conditions and cell degradation in the liver, heart, or muscles caused by synthetic pesticides. Research on the use of liver enzymes as bioindicators to assess pesticide exposure in freshwater fish is limited. However, studies suggest that liver enzymes can be a useful marker of liver injury in organisms exposed to synthetic pesticides, providing insight into their effects on different tissues and metabolic processes.

## **3.1. Best Alternatives to Pesticides**

Bio-pesticides, derived from plants (e.g., neem oil), microorganisms, or minerals, can effectively control pests with less environmental impact. Biological control involves introducing natural predators, parasites, or pathogens to control pest populations. Crop rotation and diversification disrupt pest life cycles, while physical barriers like nets, screens, or row covers prevent pests from reaching crops. Trap cropping and pheromones lure pests away from main crops, while cultural practices like proper irrigation management, planting timing, and sanitation can reduce pest pressure and minimize pesticide use. Potential harm to birds, beneficiary insects, and fish can be minimizing by using pesticides only when necessary.

#### **4. Future prospects**

The indiscriminate use of pesticides poses a threat to non-target organisms, ecosystems, and human health. The future of pesticide use depends on innovative technologies, sustainable farming practices, and consumer awareness. Governments, researchers, and farmers are focusing on developing alternatives to minimize environmental impacts. Monitoring fish liver enzymes provides insights into fish populations in pesticide-contaminated environments. Parameters studied can be used as potential biomarkers for pesticide toxicity in environmental bio-monitoring. Chemical pollutants from industry, urbanization, and agricultural practices are entering aquatic systems, posing a significant threat to fish and aquatic species. Environmental stressors cause tissue damage, growth retardation, genotoxicity, reproductive disturbance, and tissue bioaccumulation. Future pesticide use is focusing on reducing environmental impact and improving efficacy through sustainable practices and alternative technologies. Key trends include Integrated Pest Management, biological pesticides, Precision Agriculture, gene editing, and organic farming practices. This monitoring can help in assessing the effectiveness of regulations and conservation efforts aimed at reducing pesticide runoff into water bodies.

#### **5. Conclusion**

In conclusion, the use of liver enzymes as biomarkers for pesticidal toxicity in fish not only provides insights into current environmental health but also lays the foundation for proactive environmental management and conservation strategies in the future. By leveraging biomarker data, stakeholders can work towards sustainable pesticide management practices that minimize negative impacts on aquatic ecosystems and human well-being.

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