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Original Research Paper

# Impact of Quinalphos on Acid Phosphatase and Alkaline Phosphatase Activity in the Tissues of Freshwater Fish, *Labeo rohita*

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## ABSTRACT

The present study deals with the effects of insecticide, Quinalphos on the acid phosphatase and alkaline phosphatase activity of various tissues like gill, liver, kidney and muscle in the freshwater fish, *Labeo rohita*. Fishes were treated with Quinalphos for 1 day, 2 days, 3 days, 10 days and 20 days in water containing sublethal concentrations of Quinalphos (6.06 ppm). The observations indicate that there was decline in acid phosphatase and alkaline phosphatase activity of Quinalphos treated tissues in the fish.

## INTRODUCTION

Indiscriminate use of pesticides in agriculture to prevent the crops from peril has been increased over the years, especially in the developing countries. These pesticides even when applied in restricted areas are washed and carried away by rains and floods to large water bodies like ponds and rivers and alter the physico-chemical properties of water (Balachandra et al. 2001). These are proving to be highly toxic not only to fishes but also to other organisms, which form food of the fishes (Madhab Prasad et al. 2002).

The enzyme acetyl cholinesterase (AchE) is responsible for hydrolysing the neurotransmitters in to chlorine and acetic acid. The enzyme controls extra hepatic neuromuscular junction. The inhibition of the AchE is linked directly with mechanism of toxic action of organophosphate pesticides. Irreversible binding to the catalytic size of enzyme and protection of cholinergic effect as an indicator of exposure to these compounds, acetyl cholinesterase and non specific cholinesterase activities in blood and tissues have emerged as a diagnostic tool in the biomedical area. The quantification of this enzyme has been applied in the laboratory and field study with vertebrates to asses exposure to organophosphorus insecticides, which are dependent on their binding capacity to the enzyme active site and by their rate of phosphorylation in relation to the behaviour and age (Dutta & Arends 2003).

The persistence of toxic chemicals in aquatic environment becomes dangerous for the survival of fish and their food organisms. Therefore, it is essential to study the toxic effects of pesticides on living organisms.

## MATERIALS AND METHODS

The freshwater fish, *Labeo rohita* (body length 5-7 cm, body weight 5-6 g) were collected from Aliyar Dam and acclimatized to laboratory condition for 2 weeks in a large cement tank at 24±3°C. The fish were fed regularly with conventional diet rice bran and oil cake in 1:1 ratio. Feeding was stopped one day prior to the start of the experiment. Technical grade of Quinalphos insecticide was used in the investigation. Batches of 10 healthy fish were exposed to different concentration of the insecticide. $LC_{50}$ value for 72 hrs was calculated by using probity analysis.

Five groups of fish were exposed to 6.06 ppm (sublethal concentration of 72 hrs LC50 value) concentration of the Quinalphos for 1, 2, 3, 10 and 20 days. Another group was maintained as control. At the end of each exposure period, fishes were sacrificed and tissues such as liver, kidney, muscle and gill were dissected and removed. The tissues were homogenized with 80% methanol, centrifuged at 3500 rpm for 15 minutes, and the clear supernatant was used for analysis of different parameters. The results were expressed as mg/g wet weight of the tissue. Different enzymological parameters like acid phosphatase (ACP) and alkaline phosphatase activity (ALP) were analysed.

## RESULTS AND DISCUSSION

The results of ACP and ALP activity in the gill, kidney and muscle are presented in Tables 1 and 2. ACP showed a varied trend in its level in different tissues of *Labeo rohita*.

During the study period, the ACP activity in the fish was

Table 1: Acid phosphatase activity (IU/L) in the tissues of the fish, *Labeo rohita* on exposure to insecticide, Quinalphos.

| Name of the Tissues | | Exposure Periods | | | | |
|---|---|---|---|---|---|---|
| | | 1 day | 2 days | 3 days | 10 days | 20 days |
| Liver | Control | 4.31 ± 0.02 | 4.31 ± 0.02 | 4.31 ± 0.02 | 4.31 ± 0.02 | 4.31 ± 0.02 |
| | Experimental | 3.11 ± 0.06*** | 2.17 ± 0.05*** | 1.99 ± 0.00*** | 1.51 ± 0.05*** | 0.91 ± 0.02*** |
| | 't' value | 8.35 | 8.22 | 1.37 | 1.33 | 2.52 |
| | % change | -27.88 | -49.68 | -53.85 | -64.98 | -78.90 |
| Gill | Control | 7.95 ± 0.02 | 7.95 ± 0.02 | 7.95 ± 0.02 | 7.95 ± 0.02 | 7.95 ± 0.02 |
| | Experimental | 4.31 ± 0.03*** | 3.94 ± 0.05*** | 1.99 ± 0.06*** | 2.55 ± 0.08*** | 1.21 ± 0.02*** |
| | 't' value | 1.18 | 5.43 | 2.28 | 5.02 | 8.53 |
| | % change | -45.80 | -50.50 | -74.97 | -67.93 | -84.78 |
| Kidney | Control | 6.03 ± 0.01 | 6.03 ± 0.01 | 6.03 ± 0.01 | 6.03 ± 0.01 | 6.03 ± 0.01 |
| | Experimental | 1.49 ± 0.03*** | 0.98 ± 0.06*** | 0.01 ± 0.01*** | 1.86 ± 0.08*** | 0.41 ± 0.02*** |
| | 't' value | 2.01 | 8.58 | 3.25 | 3.98 | 3.65 |
| | % change | -75.30 | -83.75 | -99.78 | -69.16 | -93.20 |
| Muscle | Control | 5.64 ± 0.02 | 5.64 ± 0.02 | 5.64 ± 0.02 | 5.64 ± 0.02 | 5.64 ± 0.02 |
| | Experimental | 2.01 ± 0.02*** | 1.87 ± 0.3*** | 0.94 ± 0.05*** | 2.94 ± 0.08*** | 0.97 ± 0.06*** |
| | 't' value | 1.67 | 1.65 | 1.51 | 1.82 | 1.52 |
| | % change | -64.36 | -66.84 | -83.33 | -47.87 | -82.80 |

Results are mean (±SD) of 6 observations; % change = percent decrease (-) over control
*** = Significant at 0.001 level.

decreased up to 20 days showing maximum percentage decrease of 78.90, 84.78, 93.20 and 82.80 in liver, gill, kidney and muscle respectively on 20th day.

The ALP activity was also decreased from 1 to 20 days exposure period. The maximum percent decrease was observed in kidney (95.45 on 2nd day), gill (92.93 on 3rd day), liver (92.92 on 20th day) and muscle (89.45 on 3rd day).

The enzyme alkaline phosphatase is associated with glycogen and is linked with transportation of intermediate compounds in glycogenesis or glycogenolysis but it has also been linked with DNA in the nucleus, secretion and formation of proteins (Bradfield 1950) and tissue growth. The decrease in the tissue ALP activities in this study may be attributed to the toxins in the fish tissue which affects the synthesis of enzyme protein directly or indirectly and increased metabolism due to an increase of toxic substances and the production of toxic metabolic products destructive to enzymes.

Acid phosphatases are hydrolytic lysosomes for the hydrolysis of foreign material, hence they have a role in certain detoxification functions. It is known as inducible enzyme whose activity in animal tissue goes up when there is a toxic impact and the enzyme begins to counteract. The pronounced increase in protease activity may be due to the damage caused to lysosomal membrane, thus permitting the leakage of lysosomal enzyme into the cytosol as suggested by Sherekar & Kulkurni (1987). ALP is a hydrolyzing enzyme. It catalyses the hydrolysis of phosphomonoester to the inorganic phosphorus alteration in ACP activity in fish due to the effect of pesticide.

Decreased phosphates activity may be due to alteration in membrane permeability, distribution of normal functioning of cell organalles like lysosomes and mitochondria and different suppressor mechanisms associated with toxicity together resulted in significant changes in the level of enzymes in the tissues examined. The decreased activity of ALP in fish is linked to the increased catabolic breakdown in melanomacrophage centres.

Elevation in function of ALP activity might be due to an accelerated membrane transport for the function related to anion compounds. Another possibility in activity of ALP may be due to the destruction of the hepatic smooth endoplasmic reticulum membrane. The decreased ALP activity may be due to the destruction of lysosomal membrane (Etim et al. 2006).

The decreased activity of ALP in the tissue is due to the accumulation of metals in fishes (Humstoc et al. 2007) which affects the synthesis of enzyme protein directly or indirectly and or increased metabolism due to an increase of toxic substances and the production of toxic metabolic products destructive to enzymes. Further, more decrease in the ALP activity might be due to the direct action of pollutants on the enzymes and or the other toxic effects produced in tissues. Vorbrodt (1959) has reported that phosphatase has an important role in the transport of metab-olites across the membrane.

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Table 2: Alkaline phosphatase activity (IU/L) in the tissues of the fish, ***Labeo rohita*** on exposure to insecticide, Quinalphos.

| Name of the Tissues | | Exposure Periods | | | | |
|---|---|---|---|---|---|---|
| | | 1 day | 2 days | 3 days | 10 days | 20 days |
| Liver | Control | 70.61 ± 0.05 | 70.61 ± 0.05 | 70.61 ± 0.05 | 70.61 ± 0.05 | 70.61 ± 0.05 |
| | Experimental | 55.99 ± 0.02*** | 38.61 ± 0.05*** | 21.49±0.06*** | 11.25±0.06*** | 5.00 ± 0.03*** |
| | 't' value | 2.68 | 5.09 | 1.65 | 3.63 | 1.63 |
| | % change | -20.70 | -45.32 | -69.57 | -84.07 | -92.92 |
| Gill | Control | 101.25 ± 0.03 | 101.25 ± 0.03 | 101.25 ± 0.03 | 101.25 ± 0.03 | 101.25 ± 0.03 |
| | Experimental | 31.28 ± 0.06*** | 20.59 ± 0.05*** | 7.16 ± 0.09*** | 9.64 ± 0.03*** | 12.48±0.02*** |
| | 't' value | 9.75 | 3.13 | 9.12 | 1.13 | 1.45 |
| | % change | -69.11 | -79.66 | -92.93 | -90.48 | -87.67 |
| Kidney | Control | 51.65 ± 0.16 | 51.65 ± 0.16 | 51.65 ± 0.16 | 51.65 ± 0.16 | 51.65 ± 0.16 |
| | Experimental | 4.61 ± 0.03*** | 2.35 ± 0.02*** | 3.75 ± 0.08*** | 12.38±0.06*** | 4.61 ± 0.05*** |
| | 't' value | 2.34 | 1.60 | 2.02 | 9.90 | 2.34 |
| | % change | -91.0 | -95.45 | -92.74 | -76.03 | -91.07 |
| Muscle | Control | 89.25 ± 0.03 | 89.25 ± 0.03 | 89.25 ± 0.03 | 89.25 ± 0.03 | 89.25 ± 0.03 |
| | Experimental | 21.49 ± 0.05*** | 11.02 ± 0.08*** | 9.42 ± 0.06*** | 20.25±0.03*** | 13.28±0.02*** |
| | 't' value | 1.26 | 4.00 | 3.40 | 1.09 | 5.05 |
| | % change | -75.92 | -87.65 | -89.45 | -77.31 | -85.12 |

Results are mean (±SD) of 6 observations; % change = percent decrease (-) over control
*** = Significant at 0.001 level

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