

Chlorpyrifos Induced Alterations in Glycogen Content in Some Organs of Fresh Water Fish, *Channa gachua* (F. Hamilton)

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ABSTRACT: Highly effective pesticides are used tremendously which enters into aquatic system and brings multiple changes in the organism by altering the growth rate, nutritional value, behavioral pattern and biochemical composition. From aquatic ecosystem fish is used as food by the major part of the world. So, it is essential to secure the health of fish. The main objective of the present investigation is to evaluate the impact of organophosphorus insecticide, chlorpyrifos on glycogen content in *Channa gachua*, an economically important fresh water fish. The lethal concentration was found to be 0.05 ml/L for 96 hrs. Under the laboratory condition the fishes were exposed to sub lethal concentration (i.e. 1/4th conc. of LC 50) for 24, 48, 72 and 96 hrs and investigated significant changes in glycogen content in some organs. The glycogen content in muscle, gill, intestine, stomach and brain was found to be declined after pesticide exposure. The rapidly decreased total glycogen content might be due to the influence of pesticide that disturbs the metabolism due to the toxicity stress. It is concluded that chlorpyrifos either in low or high concentration is very toxic to non target organism.

KEYWORDS: Chlorpyrifos, glycogen, *Channa gachua*, sub lethal concentration, insecticide.

I. INTRODUCTION

The fresh water is harvested for drinking purposes in all the areas and also it is used for agriculture and fisheries (Binukumari and Vasanthi, 2014). Environmental contamination by different pesticide is one of the major cause of pollution. Pesticides are frequently used to control pests in agricultural and domestic fields, but they also enter in to an aquatic system through intentional application, runoff from farms, accidental or illegal release and affect to non-target organism like fish. The fatal effect of the pesticides on fish, particularly on its physiology and biochemical parameters is witnessing in the toxicological studies (Chaudhary and Yadav, 2013). Chlorpyrifos is an organophosphate pesticide widely used in agriculture, aquaculture and fishery pest control. Its excessive use creates numerous environmental issues. Chlorpyrifos is directly toxic to the nervous system and is also transformed inside animal to chlorpyrifos oxon. Due to its lipophilic nature, fishes are able to absorb and bio concentrate chlorpyrifos to moderate or high levels (Ogueji, 2013).

Fish serves as a bioindicator species as it sensitively responds to change in the aquatic environment so it plays an important role in the monitoring of water pollution (Srivastava et.al., 2010). Fish accumulates pollutants directly from contaminated water or via food chain and those pollutants adversely affect the physiology and metabolism of fish. The nutritional value of fish depends on their biochemical composition which is affected by pesticidal stress (Saradhamani and Binukumari, 2011). Some workers reported that the organophosphate pesticides are more toxic as they produce significant reduction in biochemical component in different tissues. The mode of action of the pesticide varies from species to species, and animals. Keeping this fact in view the present study is framed to evaluate the toxic effect of chlorpyrifos on glycogen content in some organs of fish *C. gachua*.

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II. MATERIAL AND METHODS

A) MATERIAL:

- 1) Selection of fish -In the present work the fresh water fish, *C.gachua* was selected keeping in mind their availability in local river Krishna around Karad city and its suitability for toxicity testing. Healthy specimens of size 15 ± 1 cms and weight 50 ± 5 gms. were collected from the river with the help of a fisherman. The collected fish were brought to the laboratory, they were treated with 0.1% $KMnO_4$ solution to prevent dermal infection and acclimatized to the laboratory condition for a period of 10 days. During this period, they were fed once a day on pelleted fish feed and were kept in aerated aquarium. After 10 days fish with normal behavioral activity and good health condition were selected for further experimental purpose.
- 2) Pesticide - Commercially available organophosphorus pesticide, chlorpyrifos brought from local Agro- chemist shop was used for present research work.

B) Methods:

- 1) Determination of LC_{50} - To understand the influence of time over the toxic effect of chlorpyrifos at lethal concentration on *C. gachua* at different period of exposure, the fish were grouped into six groups and exposed to different concentrations viz. 0.04, 0.045, 0.05, 0.055, 0.06 and 0.065 ml/L for 96 hrs and recorded the mortality of fishes.
- 2) Experimental set up - Under the experiment healthy fish were divided in to three groups, each group contained ten fishes. Group 1st was considered as control and group 2nd and 3rd as the experimental groups. Fish in the experimental groups were exposed to sub lethal concentration, i.e., 1/4th conc. of LC_{50} of chlorpyrifos for 24, 48, 72 and 96 hrs. At the end of each exposure period the fish were sacrificed and desired organs were quickly excised to study biochemical parameter by employing following method.
- 3) Biochemical method - Anthrone method was used to determine total glycogen content in tissues, (Roe, 1955). To make a test sample, freshly weighed (100 mg.) tissue was homogenized in 1 ml of 30 % KOH and 2 ml of 96 % ethanol, the homogenate was centrifuged at 3000 rpm for 15 min and the supernatant was obtained. In 0.5ml of supernatant, 0.5 ml of distilled water and 5 ml. of anthrone reagent was added. A standard glucose was also run along with the test sample. The samples were boiled in a water bath for 8 min. for color development. The optical density was measured in a UV spectrophotometer at 620nm. The glycogen content of the tissue was calculated using the OD of the unknown sample and that of the standard glucose solution.

III. RESULT

LC 50

Median lethal concentration (LC_{50}) is the most widely accepted basis for acute toxicity test and it is the concentration of a test chemical which kill 50% of the test organisms after a particular period of exposure, usually 96 hrs. Generally in toxicity tests, death is a decisive criterion because it is easy to determine and has obvious biological and ecological significance. The mortality of fishes at different concentrations viz. 0.04, 0.045, 0.05, 0.055, 0.06 and 0.065 ml/L of chlorpyrifos for 24, 48, 72 and 96 hrs. is shown in table 1. It is clearly indicates that the LC_{50} value for chlorpyrifos at 96 hrs. was 0.05ml/L.

Total glycogen-

The glycogen content obtained in different tissues of control fish and chlorpyrifos treated fish at different time intervals is shown in table 2 and represented graphically in figure 1. The result showed significant decrease of glycogen content in all the tissues under present investigation. The graphical representation showed the mean values obtained for total glycogen

in different tissues of fish over the control along with standard deviation. In control fish the glycogen content was in order of stomach > intestine > brain > muscle > gill. At the exposure of 96 hrs. sub lethal concentration of chlorpyrifos, mean values of total glycogen depleted in the order of stomach > intestine > muscle > brain > gill. At 96 hrs. sub lethal exposure of pesticide the mean value of glycogen linearly decreased in stomach as 1.1525, 0.9795, 0.8867 and 0.6959 mg/gm wet wt. of tissue for 24,48,72 and 96 hrs. respectively, when compared with the control (1.4820) at the end of 96 hrs. The mean value of intestine glycogen content (0.7224 mg/g wet wt. of tissue) had shown a significant decrease

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when compared to control group (1.4538mg/g wet wt. of tissue). The mean value of muscle glycogen content in the experimental groups were 0.6676 , 0.5393, 0.4813, and 0.3470 mg/g wet wt. of tissue and that of control group was 0.8196.

Table 1. Mortality of *C.gachua* exposed to chlorpyrifos.

No. of fishes	Concentration ml/L	Exposure period (hrs.)			
		24	48	72	96
10	0.04	N	N	N	1
10	0.045	N	N	1	2
10	0.05	N	1	2	2
10	0.055	1	1	2	3
10	0.06	1	2	3	3
10	0.065	6	4	0	0

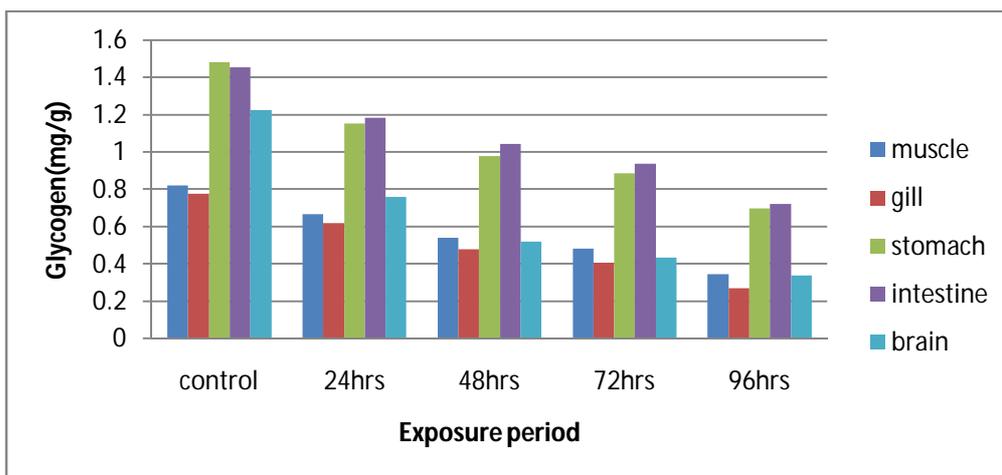
Note: N- No mortality

Table 2. Glycogen content (mg/g wet wt. of tissue) in different tissues of fish, *C. gachua* on exposure to sub lethal concentration of chlorpyrifos for 24, 48, 72 and 96 hrs.

Tissue	Control	24 hrs	48 hrs	72 hrs	96 hrs
Muscle	0.8196 ± 0.0247	0.6676 ± 0.002	0.5393 ± 0.010	0.4813 ± 0.004	0.3470 ± 0.005
Gill	0.7754 ± 0.0371	0.6183 ± 0.004	0.4795 ± 0.008	0.4062 ± 0.008	0.2702 ± 0.005
Stomach	1.4820 ± 0.026	1.1525 ± 0.011	0.9794 ± 0.010	0.8867 ± 0.004	0.6959 ± 0.006
Intestine	1.4537 ± 0.008	1.1826 ± 0.010	1.0439 ± 0.017	0.9353 ± 0.009	0.7224 ± 0.010
Brain	1.2242 ± 0.026	0.7586 ± 0.010	0.5193 ± 0.004	0.4336 ± 0.006	0.3373 ± 0.010

Each value is the mean of 5 individual determinations ± indicates SD.

Figure 1. Changes in glycogen content (mg/g wet wt. of tissue) in different tissues of fish, *C. gachua* on exposure to sub lethal concentration of chlorpyrifos for 24, 48, 72 and 96 hrs.



The mean value of glycogen was decreased in the brain as 0.7586, 0.5193, 0.4336 and 0.3373 mg/gm wet wt. of tissue for 24,48,72 and 96 hrs. respectively, where as the glycogen content in the brain of control group was 1.2242. Decreased amount of glycogen content was noticed in the gill during chlorpyrifos treatment, so the mean value of control group was 0.7754 whereas the mean values of the experimental groups were 0.6183 , 0.4795 , 0.4062 and 0.2740 mg/gm wet wt of tissue for 96 hrs.

IV. DISCUSSION

In the present study when the fish were exposed to chlorpyrifos, glycogen was significantly declined. This declination might be due to the toxic action of the pesticide on the tissues and utilization of stored glycogen to overcome the pesticide stress. Similar results have been reported by Chaudhary and Yadav (2013) in fish *Nemachelus botia* on exposure to meothrin. Padma Priya (2013) reported that the tissue glycogen content decreased on exposure of fish *Channa punctatus* to imidacloprid. Decreased liver glycogen has been observed in fish *O. mossambicus* due to toxic impact of dichlorvos (Lakshmanan et al., 2013). Satyavardhan (2013) reported that the glycogen in different tissues of fish *Ctenopharyngodon idella* was significantly reduced by the fenvalerate and malathion intoxication. Nagaraju and Venkata Rathnamma (2013) showed that the glycogen content decreased in the different tissues of *Labeo rohita* on exposure to sub lethal and lethal concentrations of profenofos and carbosulphan. Level of glycogen content in the liver of *Cyprinus carpio* was decreased when exposed to distillery effluent at different periods (Saroja et al., 2013). In fish *Lepidocephalus thermalis* a sudden decrease in glycogen content was reported in liver, kidney and muscle tissue on exposure to sugar factory effluent (Hyalij., 2013). Magar and Dube (2013), observed that the fresh water fish *C. punctatus* exposed to sub lethal concentration of malathion showed decreasing trend in cardiac muscle glycogen. The glycogen content in the brain, gill, intestine and liver of fish *Cirrhina mrigala* was decreased significantly on exposure of fenthion (Israel Stalin and Sam Manohar, 2012). The significant decrease was observed in the gills, kidney, and brain of fish *Punctius ticto* due to the toxic action of dimethoate (Ganeshwade et al.; 2011). Palanistry et. al., (2011) studied and reported decline in the glycogen content of liver, brain, muscle and gill tissue of *Mystus cavasius* after short term exposure to electroplating industrial effluent chromium. Thenmozhi (2011), suggests that depletion in biochemical parameter like glycogen in liver, gill and muscle of *Labeo rohita* during exposure period of malathion. The serum total glucose content has been observed to be decreased after phorate treatment in fish *C. punctatus* (Singh, et.al., 2010). Decrease in the glycogen content in gill, liver, muscle and kidney of *Labeo rohita* when exposed to tannery effluent after acute exposure for 96 hrs, has been reported by Muley et. al., (2007). Ghosh (1987) indicates gradual depletion in muscle and liver glycogen of *C. batracus* when exposed to sub lethal concentration of three organophosphates.

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The tissue specific decline in glycogen may be due to its speedy consumption to assemble the energy demands (Padma Priya; 2013). Where as the glycogen is advantageous for immediate energy release on demand in any animal. Under the toxic stress to mitigate energy demand, glycogen level in tissue falls down. In present study glycogen content in different tissues was found depleted due to the toxins which damage the structure of tissue and results in the weakened metabolism process. So the results of present investigations are similar to the findings of various earlier workers.

V. CONCLUSION

In view of above results and discussion in the present study the glycogen content in different tissues of *C. gachua* was decreased when exposed to sub lethal concentration of chlorpyrifos. Thus, from the present study it is concluded that insecticide intoxication disturbs the functional activity of cells with consequential changes in biochemical composition of fish, that affect the nutritional value of fish. It may be dangerous to human beings due to the consumption of those fish in which insecticides have accumulated in their tissues.

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