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Effects of Quinalphos on Glycogen and Protein Content in Different Tissues of the Freshwater Fish, Channa Gachua (Hamilton, 1822)

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ABSTRACT

This study was conducted to investigate changes in the glycogen and protein content of the gill, liver, muscle and kidney of the air-breathing fish *Channa gachua* exposed to chronic exposure of quinalphos for 10, 20 and 30 days exposure at 1/10th of 96 hr toxicity (0.147 ppm). The estimated glycogen in the tissue gill, liver, muscle and kidney were found to reduce during the exposure period, the liver was most affected than the other tissues in compare to control. The disruption of enzyme associated with carbohydrate metabolism resulted in glycogen depletion. The estimated protein in the tissue gill, liver, kidney and muscle were found to significantly reduce during the exposure period. Protein degradation in the tissues of test fish suggests that the quinalphos disturbing the process of protein metabolism by declining the protein level due to the disturbance in the proteolysis.

Keywords : *Channa gachua*, Gill, Glycogen, Kidney, Liver, Muscle, Protein, Quinalphos

I. INTRODUCTION

Pesticide is defined by United Nations Environment Programme (UNEP, 2005) as any substance or mixture of substances intended for preventing, destroying, repelling or mitigating any pest. Chemical pesticides are generally designed for controlling pests and increase the economy of agriculture industry and to meet the world's need for abundant, safe and affordable food and fiber, at the same time such pesticides are became highly toxic to the other organisms in the environment. The use of such chemical pesticides in agriculture is a problem to ecosystem it is resulting in the environmental pollution (Barbieri, 2008) and toxicity risk to the non-target organisms (Venkateswara Rao, 2006) including human health (Calvert *et al.*, 2001; Snodgrass 2001) and cause a major public health problem. Pesticides are a very important group of environmental pollutants used in intensive agriculture for protection against diseases and pests. Quinalphos is an organophosphate pesticide which is extensively used in agriculture for pest eradication (Das and Mukherjee, 2000) and it is hard insecticide having potential and hazardous effect (Muttappa *et al.*, 2014). It is highly toxic to non-target organisms. Quinalphos (O, O-diethyl O-quinoxalin-2-yl phosphorothiate) is a synthetic organophosphate, non-systemic, broad spectrum insecticide and acaricide, acting as a cholinesterase inhibitor with contact, stomach and respiratory action, it is extensively used in Indian agricultural field owing to its low bioaccumulation and high degradation (Mishra and Devi, 2014). Contamination of aquatic ecosystem by Quinalphos is fairly common and had serious impact on fish (Chebbi and David, 2009). Fish is the best understood aquatic

species and can be a front line indicator of suspected aquatic pollutants because of that fish used as test organism. Ever since aquatic ecosystems have been considered potential sources of contamination, fishes can be served as bio-indicators used for the quality assessment of the aquatic system (Lopes *et al.*, 2001; Dautremepuits *et al.*, 2004) and can used as bio-monitor organisms for the monitoring of environment.

Glycogen is the main storage polysaccharide of animal cells and serves as a primary energy source for metabolic processes under the stressful condition. The disturbance in the glycogen profile is an indicator of stressful condition due to pesticide intoxication. The stored glycogen may be utilized for their survival in the polluted environment and no further glycogen synthesis, so this could be the reason of glycogen depletion (Satyavardhan, 2013).

Protein is one of the main targets for elucidation of effects by the pesticides (Marigoudar, 2012). The protein content in the tissues of animal plays a role in metabolism of animals (Palanivelu *et al.*, 2005). Proteins occupy a unique position in the metabolism of cell because of the proteinaceous nature of all the enzymes which mediate at various metabolic pathways (Harper, 2006; Lehninger, 2008). Estimation of total protein contents of vital organs are considered as important factor for toxicological studies (Mary Chandravathy and Reddy, 1996).

Hence, the present study was conducted to investigate the chronic effect of quinalphos 25% EC on glycogen and protein content in the gill, liver, muscle and kidney tissues of freshwater fish *Channa gachua*.

II. MATERIALS AND METHODS

Test Organism and Acclimatization

The freshwater fish *Channa gachua* (length 18 $\pm$ 1.00 cm and weight 40 $\pm$ 5.00 gm) was procured from Godavari River, Kaygaon Toka near Aurangabad. These fishes were acclimatized to the laboratory conditions with de chlorinated tap water and fed with dried prawn for 10-15 days at a room temperature 27 $\pm$ 2^0C prior to the experimental condition.

Toxicity assay:

To the study of chronic toxicity of quinalphos, $1/10^{th}$ of 96 hr LC_{50} concentration (0.147 ppm) was used for 10, 20 and 30 days exposure period in experimental glass aquaria of 40 l capacity. Ten fish equal size were used in 20 l of water.

Glycogen estimation

To the study of the glycogen levels in the gills, liver, muscle and kidney, fishes were divided into two groups as control and experimental. After exposure, both control and experimental fishes were sacrificed. The fishes were dissected and gills, liver, muscle and kidney were processed for Glycogen estimation, it was done by Anthrone reagent method (Dezwaan and Zandee, 1972) the optical density was measured at 620 µm. The data was subjected to one-way analysis of variance (ANOVA) and the significance difference was set up at $p < 0.05$.

Protein estimation:

The total protein content of the pesticide exposed tissue samples were estimated by the modified method of Lowry *et al.*, (1951). The optical density was measured at 540 µm against blank. The standard graph was plotted with bovine serum albumin (BSA) as standard, supplied by Sigma Chemical Company, U. S. A. The values were expressed as mg/100 mg wet weight of the tissue. The data was subjected to one-way analysis of variance (ANOVA) and the significance difference was set up at $p < 0.05$.

III. RESULTS AND DISCUSSION

Glycogen content:

During the glycogen estimation in gill, liver, muscle and kidney tissue of *Channa gachua* was found to be decreased in response to chronic exposure of quinalphos. The glycogen content in gill, liver, muscle and kidney tissue was found significantly decreased. The results shows glycogen content in liver tissue was drastically affected in chronic exposure for 20 days exposure (9.17 ± 0.26) mg/100mg and for 30 days exposure period viz. 9.05 ± 0.31 mg/100mg in compare to control (9.57± 0.23) mg/100mg, where 10 days exposure (9.25 ± 0.21) mg/100mg shows non-significant reduction in compare to control (9.57± 0.23) mg/100mg. This indicates that, the effect of quinalphos on glycogen content of liver tissue significantly affected than other tissues by percentage change 5.43 % for chronic exposure. The observed values of glycogen content tabulated in table no. 1 and presented in graph viz. fig. I. respectively. The fall in glycogen level may be due to utilization of stored carbohydrates in liver for energy production as a result of pesticide-induced hypoxia. Shobha *et al.*, (2007) stated that the decrease in glycogen content maybe due to the inhibition of hormones which contribute to glycogen synthesis. The glycogen stored in liver, kidney and intestine can be made accessible to other organs by virtue of their possession of an enzyme glucose-6-phosphatase (Vornanen *et al.*, 2011). The depletion in glycogen synthesis is also attributed to the inhibition of the enzyme glucose-6-phosphatase or glycogen synthetase which mediates glycogen synthesis. The decrease in glycogen content may also be either due to decreased synthesis as a consequence of toxic stress (Dezwaan and Zandee, 1972). Swarna Kumari *et al.*, (2008) observed the depletion in glycogen content in gill, liver, muscle, brain and kidney of *Ctenophayngodon idella* exposed to organophosphorus pesticide dichlorvos. Similar significant reduction was reported in glycogen content was found in the liver, kidney and muscle tissues of *Oreochromis mossambicus* after exposed to dichlorvos (Lakshmanan *et al.*, 2013). The decrease in glycogen level in the vital tissues may be due to the decreased rate of glycogenesis. Similar glycogen content depletion was observed in *Cirrhina mrigala* exposed to fenthion (Stalin and Das, 2012), in *Ctenopharyngodon idella* exposed to fenvalarate and malathion (Satyavardhan, 2013), in *Labeo rohita* exposed to dimethoate (Binukumari and Vasanthi, 2013), in *Labeo rohita* exposed to phenthoate (Somaiah *et al.*, 2014), in *Channa gachua* exposed to quinalphos (Pakhare *et al.*, 2016).

Table - 1: The amount of total glycogen (mg/100g) in different tissues of *C. gachua* exposed to chronic exposure (1/10th of 96 h) of Quinalphos (25 % EC).

Organs	Control	Experimental		
		10 Day (0.147 ppm)	20 Day (0.147 ppm)	30 Day (0.147 ppm)
Gill	5.50 ± 0.23	5.42 ± 0.28 (1.46)	5.38 ± 0.33 (2.19)	5.24 ± 0.23 (4.73)
Liver	9.57 ± 0.24	9.25 ± 0.21 (3.34)	9.17 ± 0.26 (4.18)	9.05 ± 0.31 (5.43)
Muscle	4.49 ± 0.35	4.46 ± 0.38 (0.67)	4.39 ± 0.35 (2.22)	4.32 ± 0.33 (3.79)
Kidney	6.15 ± 0.34	6.12 ± 0.16 (0.49)	6.09 ± 0.13 (0.98)	6.00 ± 0.17 (2.44)

Mg/g wet wt. of tissue. [Each value indicate the mean (X ± SD) of five estimations] [Values in the parenthesis indicate percent change over control] [Values are highly significant at p<0.05]

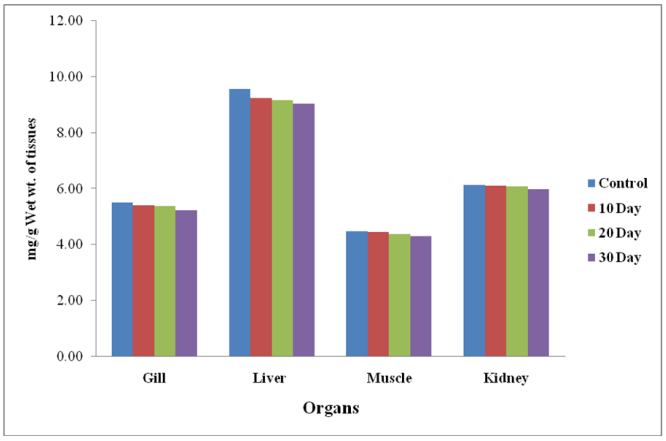

Fig. 1. The amount of total glycogen (mg/100g) in different tissues of *C. gachua* exposed to chronic exposure (1/10th of 96 h) of Quinalphos (25 % EC).

Protein content:

A significant gradual depletion in protein level was found as compared to control, in gill, liver, kidney and muscle tissues of *C. gachua* when exposed to chronic exposure period of quinalphos 25 % EC insecticide. The dose dependent and time dependent gradual decrease in protein level indicating the breakdown of these protein contents due to quinalphos intoxication. The observed values of protein content tabulated in table no. 2 and presented in graph viz. fig. II respectively. The data indicates that, muscle tissue was most affected followed by gills, liver and kidney and the protein content reduced gradually at 21.66 ± 0.08 mg/100mg, 21.28 ± 0.12 mg/100mg and 20.87 ± 0.14 mg/100mg for 10 days, 20 days and 30 days respectively as compared to control viz. 22.07 ± 0.09 mg/100mg. This reduction was highly significant (p < 0.05) of (1.86 %), (3.58 %) and (5.44 %) at 10 days, 20 days and 30 days exposure respectively. The other tissues viz. gill, liver and kidney was less affected than the muscle tissue. Depletion in muscle protein suggests stress in metabolism and impairment of protein synthesis machinery in fish; the catabolic process was initiated by increased proteolysis that led to rapid decline in protein concentration to meet the energy demand in extremely stressful condition or environment (Baruah *et al.*, 2004). Protein content significantly decreased in muscle tissue of freshwater fish *Labeo rohita* suggest that muscle was highly affected and gill and kidney are relatively less affected when exposed to indoxacarb (Ram, 2016). Sastry and Siddiqui (1984) reported the depletion in protein content in gill, liver, brain, kidney, intestine and muscle of *Channa punctatus* when treated with quinalphos. Schmidt (1975) stated that the depletion in protein content in the fish tissues may be due to metabolic utilization of the keto-acids to gluconeogenesis pathway for the synthesis of glucose; or due to directing free amino acid for the synthesis of necessary proteins or for the maintenance of osmotic and ionic regulations. Similar protein content depletion was observed in *Channa punctatus* when treated with malathion (Magar and Shaikh, 2012), in *Puntius ticto* when treated with dimethoate (Ganeshwade *et al.*, 2012), in *Oreochromis mossambicus* when treated with dichlorvos (Lakshmanan *et al.*, 2013), in *Channa gachua* when treated with methyl parathion (Jain, 2014), in *Channa gachua* when treated with quinalphos (25 % EC) (Pakhare and Reddy, 2017).

Table - 2: The amount of total protein (mg/100mg) in different tissues of *C. gachua* exposed to chronic exposure (1/10th of 96 h) of quinalphos (25 % EC).

Organs	Control	Experimental		
		10 Day (0.147 ppm)	20 Day (0.147 ppm)	30 Day (0.147 ppm)
Gill	17.42 ± 0.13	17.26 ± 0.15 (0.92%)	16.98 ± 0.23 (2.52%)	16.59 ± 0.26 (4.76%)
Liver	18.23 ± 0.16	18.11 ± 0.11 (0.66%)	17.98 ± 0.089 (1.37%)	17.38 ± 0.21 (4.66%)
Muscle	22.07 ± 0.092	21.66 ± 0.082 (1.86%)	21.28 ± 0.12 (3.58%)	20.87 ± 0.14 (5.44%)
Kidney	14.71 ± 0.027	14.56 ± 0.089 (1.02%)	14.41 ± 0.12 (2.04%)	14.22 ± 0.10 (3.33%)

Mg/g wet wt. of tissue. [Each value indicate the mean (X ± SD) of five estimations] [Values in the parenthesis indicate percent change over control] [Values are highly significant at p<0.05]

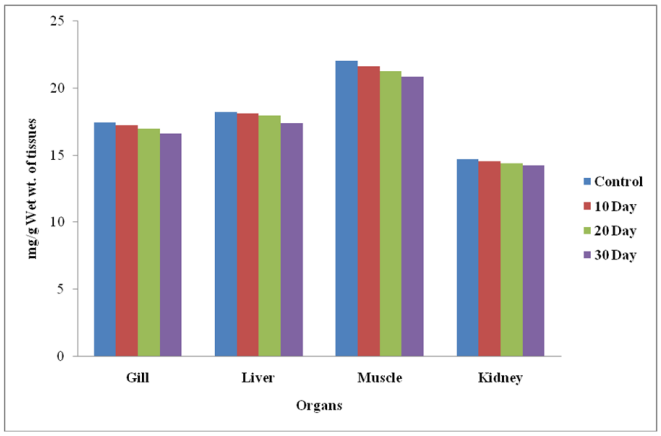

Fig. 2.: The amount of total protein (mg/100mg) in different tissues of *C. gachua* exposed to chronic exposure (1/10th of 96 h) of quinalphos (25 % EC).

IV. CONCLUSION

The contamination of quinalphos an organophosphate pesticide in environment is a serious threat to the non target organism. The depletion in glycogen and protein content indicates glycogen and protein was excessively utilized for metabolism in stress condition.

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