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TOXIC EFFECT OF CHLORPYRIFOS ON TOTAL LIPID CONTENT IN THE ADDUCTOR MUSCLES OF THE BLACK CLAM *VILLORITA CYPRINOIDES* (GRAY)

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Abstract: The agrochemical poisoning in Kuttanad, the Rice Bowl of Kerala, cause major threat to Vembanad Lake ecosystem. The knowledge of its effects on non-target aquatic species needs to be investigated. The present study was carried out to investigate the effect of the largest market-selling organophosphorous insecticide chlorpyrifos on lipid content in adductor muscle of the bivalve *Villorita cyprinoides* from Vembanad Lake. On the basis of LC_{50} value (23ppb) from acute toxic bioassay, the bivalves were exposed to four different groups *ie*, two sub-lethal concentrations of chlorpyrifos (1/10 and 1/20 of LC_{50} value), one solvent (acetone control), and a solvent-free (tap water) control for 10 and 20 days. After exposure to chlorpyrifos, the variation in lipid content in adductor muscle was estimated by Barnes and Blackstock Method. The result showed a progressive decrease in the lipid content with the exposure period. The depletion in lipid content suggests high lipolytic activity that could be due to pesticidal stress. In particular, the present study indicate that chlorpyrifos toxicity suppress biochemical constituents like lipid in bivalves and also suggests that *Villorita cyprinoides* may be employed as a reliable biological indicator for assessing pesticide contamination or other aquatic environmental stressors. Thus it is understood that monitoring the lipid content of various tissues of bivalves exposed to the toxicants, provides an early warning system for aquatic pollution whereby stressors can be detected at an early stage.

Key words: Lipid alteration, agrochemical, *Villorita cyprinoides.* Clam, Lethal oxicity.Orgaonophosphate

INTRODUCTION

Vembanad is a major Ramsar site, the second largest wetland in the country, which is very close to Kuttanad, the rice bowl of Kerala. The lake plays key role in agricultural development of Kuttanad agrosystem. Now it is under the risk of chemical contamination, and pesticide poisoning, which alter the entire water quality of the wetland ecosystem and threaten the survival of several aquatic species. The presence of organophosphate pesticides in the environment has been increased in the last decades as a consequence of an extensive use of such compounds in the agricultural activities .It disturbs the entire metabolism after entry in the body of aquatic organisms. Exposure to pesticides evokes several behavioral, physiological, and biochemical changes that appear to be closely related. (Lomte *et al.,* 1982) To counteract any stress, energy reserves, which might otherwise be utilized for growth, and reproduction will have to be diverted towards enhanced synthesis of detoxifying ligands or expended in order to maintain an elevated efflux of pollutants (Satyaparmeswara *et al.,* 2006).

Chlorpyrifos (*O*,*O*-diethyl *O*-3,5,6-trichloro-2-pyridyl phosphorothiolate, CPF), is a broad spectrum organophosphate insecticide used in agriculture. In a recent rodent study it is reported that oral administration of CPF led to the production of ROS and lipid peroxidation in brain and liver tissues (Lemus *et al.,* 2000).

The present study was undertaken to evaluate the toxicity of the insecticide chlorpyrifos, which is widely used in paddy fields of Kuttanad. Fresh water bivalves amongst the molluscs are economically important, hence, an attempt is made to investigate the effect of chlorpyrifos on the total lipid content in adductor muscles of freshwater bivalve clam *Villorita cyprinoides* which is commonly seen in the freshwater end of the Vembanad lake.

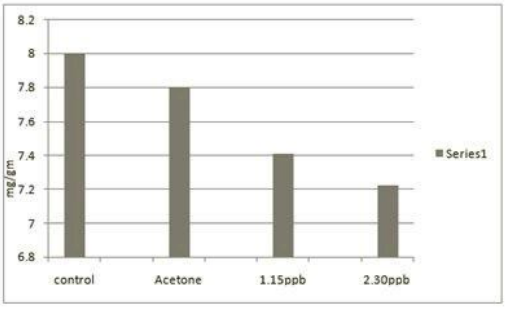
MATERIALS AND METHODS

Live and healthy samples of *Villorita cyprinoides* measuring about 24- 26 mm in length were collected from Vembanad lake near Muhamma Panchayath in Alappuzha district. Soon after the collection the bivalves were cleaned, transported to the laboratory in well aerated plastic containers and acclimatized in glass tanks for five days. Temperature, dissolved oxygen, and pH of the water were measured daily in each glass tanks. Commercially available chlorpyrifos 50% EC (Pyricon) was purchased from local market of Kuttanad. Chlorpyrifos stock solution was prepared by dissolving it in acetone solution and added to the experimental media to achieve the desired concentrations.The 96 hr LC_{50} value was calculated by Probit Analysis (Finney, 1971). After determining the LC_{50} value of chlorpyrifos (23 ppb) clams were divided into four groups, with ten clams in each group. Group I - control, group II - Acetone control and groups III - sub lethal concentrations of chlorpyrifos, viz., 1/10th (2.30 ppb) and 1/20th (1.15 ppb) of the 96 hr LC_{50} value. Tank water was renewed every 24 hr and fresh solution of chlorpyrifos was added to maintain pesticide concentration constant. Sampling was done after 10 and 20 days, excised the adductor muscle andLipid estimationwas doneby Barnes and Blackstock (1973) Method. One way ANOVA was used for comparing means which was followed further by LSD (Least significant difference) analysis (Zar, 1996).

RESULT AND DISCUSSION

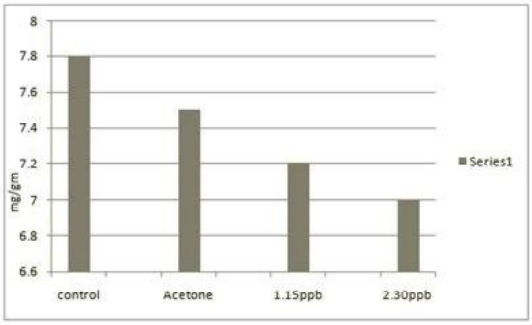
Lipid is one of the major organic constituent which acts as an energy reservoir. The liberated energy from the lipid can be used as and when required. In the present study the results demonstrated that after chronic exposure to pesticidechlorpyrifos, a marked depletion in lipid content in the adductor muscle of *Villorita cyprinoides*was observed as compared to control**.**. Lipid level in adductor muscle in pesticide treated group and control group on 10th and 20th day of exposure is represented in Figure 1and 2

After 10 and 20 days of exposure in two sub lethal concentrations, lipid content of adductor muscle decreased significantly. ANOVA revealed significant variation in total lipid content ($P<0.01$) between different group means of pesticide treated groups and control group after 10 and 20 days of exposure. LSD analysis also revealed that there was no significant difference in total lipid content in acetone control when compared to control. Depletion of lipid content in animal tissue after exposure to various pollutants was reported by several investigators .Kamble and Rao (2010), while studying the acute toxicity of Thiodon and Ekalux on the freshwater lamellibranch mollusc *Lamellidens corrianus,* observed decrease in lipid contents in different body parts. Kulkarni (2005) observed variations in the total lipid content from foot, hepatopancreas and gill of *L. corrianus* due to Haldane toxicity. . Lomte and Mule (1993) while studying the effect of copper sulphate on lipid



*Significantly different from control (P< 0.01)

Fig. 1. Lipid alteration in the adductor muscle of V. *cyprinoides* on 10th day of Chlorpyrifos exposure



*Significantly different from control (P< 0.01)

Fig. 2. Lipid alteration in the adductor muscle of V. *cyprinoides*on 20th day of Chlorpyrifos exposure.

content observed decreasing in trends from snail, *Thiara tuberculata* . Villalan *et al*. (1990) observed decreasing lipid content due to short term cadmium toxicity in the prawn, *Macrobrachium idea*. Rao *et al.* (1987) reported the lipid reduction in different tissues of the bivalve *Indonaia caeruleus* when exposed temperature and pH stress.

A marked fall in the lipid level in all the tissues indicates a rapid initiation of breakdown of lipid. Lipid is a food reservoir, store and transport in the form of di, tri-glycerol's and esters. They are major structural components of the membrane. The liberated energy from the lipid can be used during crisis (Swami *et al.,* 1983). Decrease in lipid content in adductor muscle of *V. cyprinoides* was possibly due to stress conditions caused by toxicity of chlorpyrifos on lipid metabolism or due to enhanced lipolytic activity as a consequence of increased metabolic demands following exposure to the toxic stress of pesticide.According to the Muley and Mane while studying the endosulfan toxicity to freshwater mussel, *Lamellidens marginalis* stated that, pesticide might be act as the metabolic depressor. To cope up the increasing energy demand the lypolysis might have the accelerated due to chlorpyrifos toxicity.

V. cyprinoides shows shift in its metabolism for survival and maintenance and additional demand of energy leads to reduction in lipid synthesis. This affects the nutritive value of edible organism. The present study shows lipid reduction in adductor muscles of *V. cyprinoides*, when exposed to chlorpyrifos. The result shows that there was progressive decrease in the lipid content as exposure period was increased. It seems to be the physiological manifestation of the changed toxic environment, that is, the pollution stress caused by chlorpyrifos. It suggests that the pesticides like chlorpyrifos released in the freshwater bodies may be harmful to the aquatic organisms and may cause serious health problems in human beings.

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