



The effect of chlorpyrifos pesticides on the biochemical contents in the tissues of fresh water snail *Bellamyia bengalensis* (LAMARCK)

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Abstract

The extensive use of pesticides on crops and overfishing illegally by fisherman in recent years in the south of Iraq, causing serious problems on non-target organisms leading to a number of pathological and disturbed biochemical alterations. Effect of chlorpyrifos pesticides on glycogen, lipid and protein contents of the viscera, mantle and foot at the end of 48 h and 96h of LC₅₀ of the sub-lethal concentration (0.01, 0.06) mg / l, on the freshwater snail *Bellamyia bengalensis* was studied. Significant differences were noticed at the different tissues contents to the both concentrations at 24h and 96h.

Keywords: Chlorpyrifos, Biochemical alteration , *Bellamyia bengalensis*

1- Introduction

Use of pesticides illegally by fisherman to capture fish from the south of Iraq, represent the potential threat for many aquatic organisms(Al-Helfi, 2005; Yasser *et al.*, 2008; DouAbul *et al.*, 2009; Naser, 2009). Though there is limited local information on the acute toxicity of pesticides on gastropods(yasser *et al.*, 2008), but unfortunately there is no

information about the effect of these pesticides on the biochemical contents.

Carbohydrates are the chief and immediate source of energy while proteins are source of energy during chronic periods of stress (Umminger, 1977).

Molluscs are important as not only vectors of helminth parasites but also rich source of protein food for fish and man. They form an important link in the food

chain of aquatic systems. It is impertinent to mention here that the snail, *Bellamya bengalensis* is an economically important mollusc like *Bellamya dissimilis* which is used to feed ducks in India (David, 1963). In addition to an important food item of some commercially import fish (Sinha, 1972). So far little research information is available on the effect of sub-lethal dosages of chlorpyrifos on the body tissues of snail. Therefore in the present investigation an attempt has been made to study the effect of chlorpyrifos on alterations in total protein, glycogen and lipid of *Bellamya bengalensis* with particular reference to the concentrations of pesticides and duration of exposure.

2-Materials and Methods

Adult freshwater snail *B. bengalensis* is of almost uniform size with (20.5 ± 1.8) mm height were collected from Garmat Ali at $30^{\circ}34'16''$ N $47^{\circ}44'59''$ E and kept in glass aquarium containing 30 L decolorinated tap water at least for 96 hr for acclimatization under laboratory conditions. Some of parameters of the water used in the experiments were measured as follow: PH, DO, salinity, nitrate, nitrite and total hardness as CaCO_3 , 7.2, 7.4 mg/l, 0.8 PSU, 1.4mg/l, 0.004mg/l and 138 mg/l, respectively. Water was changed every day. Dead snails were removed as soon as

possible to avoid water fouling. The snail were fed daily on washed and dried *Ceratophyllum demersum*. during the whole acclimatization period. The 96 hr LC_{50} value is 1.232 mg/L which was estimated according to Finny (1971) from the concentrations (0.5, 1, 1.5, 1.7 and 2) mg/l (Table.1). Two sub-lethal concentrations (0.01, 0.06) mg / l of The 48h and 96 hr LC_{50} were used for biochemical experiments.

Commercial grade chlorofete, chlorpyrifos 48% TC (Made in Vapco/Jordon) was used in this study. Two different concentrations of chlorpyrifos (0.01 and 0.06)mg/l and a control with six replicates were used. The snail were exposed to sub-lethal concentration of (0.01, 0.06) mg /l. In control groups the water remains pesticides free. No food was given to the snails during the course of experiment. Snails were treated for 24 hr and 96 hr and after the completion of treatment they were dissected and viscera, mantle and foot tissues were removed for biochemical analysis.

The biochemical constituents (glycogen) was determined in all the three major tissues by the colorimetric method of Kemp *et al.*, (1954) the total lipid content by Pandey *et al.*, (1963) and the total protein content by Lowry *et al.*, (1951). Each assay was replicated six times,

values were expressed as mean values and Anova- test was applied to locate significant differences at ($p < 0.01$) between treated and control groups.

3-Results and Dissucion

96 LC₅₀ Acute toxicity of chlorpyriphose on the snail *Bellamya bengalensis* is shown in the (table, 1).

Table 1. Acute toxicity of chlorpyriphose to the snail *Bellamya bengalensis*

Point	Concentration (mg/l)	95% confidence limits	Slope± SE	Intercept± SE
LC/EC 1.00	0.160	0.001 - 0.390	2.6246±0.94	4.762±0.210
LC/EC 5.00	0.291	0.008 - 0.561		
LC/EC 10.00	0.400	0.024 - 0.686		
LC/EC 15.00	0.496	0.049 - 0.789		
LC/EC 50.00	1.232	0.761 - 1.891		
LC/EC 85.00	3.058	1.959 - 27.295		
LC/EC 90.00	3.792	2.256 - 55.744		
LC/EC 95.00	5.215	2.759 - 161.869		
LC/EC 99.00	9.482	3.977 - 209.693		

Note: Control group (theoretical spontaneous response rate)= 0.0000.

LC=Leathel concentration EC=Effect concentration

Table 2: The mean values of total Protein contents, Glycogen and lipid in the viscera, mantle and foot of *B. bengalensis* exposed to sublethal concentrations (0.01, 0.06) mg / l of chlorpyriphose during 24h and 96h . (n=6)

Concentration	T	Protein			Glycogen			Lipid		
		C	24 h	96	C	24 h	96	C	24 h	96
0.01 mg/l	V	2.83	2.68**	2.41**	6.90	6.58**	5.96**	5.35	5.25**	4.43**
	M	2.73	2.56**	2.16**	6.01	5.86**	5.53**	3.46	3.40**	2.75**
	F	4.86	4.71**	4.20**	3.70	3.70*	3.20*	3.83	3.51**	2.48**
0.06 mg/l	V	2.85	2.43**	2.05**	6.81	6.21**	5.50**	5.25	4.76**	4.25**
	M	2.75	2.35**	2.06**	5.90	5.05**	4.71**	3.20	2.68**	2.18**
	F	4.88	4.20**	3.80**	3.78	3.13*	2.65*	3.95	3.45**	2.13**

T = tissue; V= viscera; M= mantle; F= foot; C=control **= p < 0.01; *= p < 0.05

The biochemical parameters (glycogen, total lipid and total protein) contents decreased in all the three tissues of the snails exposed to the pesticides. This decreased was noticed at sub-lethal concentration 24hr LC₅₀ animals and was studied highest at sublethal concentration at 96 hr LC₅₀ concentration. In general, chlorpyrifos, an organophosphorus pesticides has maximum effect on biochemical contents. The means of (Protein, glycogen and lipid) contents were changed during (24, 96)h are statically significant at ($p < 0.01$). Except total glycogen at foot of snail changes statically significant at ($p < 0.05$) (table 2). Depletion of glycogen may be due to the direct utilization for energy generation, a demand caused by pesticides induced hypoxia. The reduction in glycogen content in both the body tissues indicates it is rapid utilization by the respective tissue as a consequence of pesticides toxic stress. Similar decrease in tissue glycogen content of the dreissinid bivalve *Mytilopsis sallei* exposed to mercury (Uma Devi, 1996). Carbohydrates are the primary and immediate source of energy for fish exposed to stress condition (Umminger, 1977).. Snail exposed to sub-lethal concentration of (0.01, 0.06) mg /l required more energy, which was immediately obtained from the increased

metabolism of glycogen (Padmaja and Rao, 1994; Tripathi and Singh, 2002).

Behavioral responses of snails exposed to sub-lethal concentration of pesticides showed that they were stressed. During stress, the snails needed more energy to detoxify the toxicants and to overcome stress. Since snails have a small amount of carbohydrates, the next alternative source of energy to meet the increased energy demand is protein. The depletion of the protein fraction in viscera, mantle and foot tissues may have been due to their degradation and possible utilization for metabolic purposes. Singh *et al.*, (1996) have also reported decline in protein constituent in different fish tissue exposed to sub-lethal concentrations of pesticides.

Decrease in tissue lipid and proteins under pesticide stress could be due to several mechanisms, these include, 1) formation of lipoproteins which are utilized for repair of damaged cell and tissue organelles 2) direct utilization by cells for energy requirements and 3) increased lipolyses (Ghosh and Chatterjee, 1989).

The results of the above study indicate the highly toxic nature of chlorpyrifos on snail showed that the snail was very sensitive to the presence of chlorpyrifos even in low concentrations and are under severe metabolic stress. This study also shows the

significance of biochemical parameters in assessing pesticides hazard to snail.

4- Conclusion:

The present study shows as result of exposure to the pesticides the biochemical constituents were reduced. As a result of decreased in biochemical constituents growth and reproduction of the snail will be affected. Thus even low level of pesticides concentration residues in the biota would result in decrease of snail population which would in turn deprive the nutrition of aquatic fauna.

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تأثير مبيد الكلوربيرفوس chlorpyrifos على المحتوى البايوكيميائي في انسجة قوقع المياه العذبة *Bellamyia bengalensis* (LAMARCK)

الخلاصة

الاستخدام الواسع للمبيدات على المحاصيل و بصورة غير قانونية بواسطة الصيادين في السنوات الحالية في جنوب العراق تتسبب بمشاكل جديه على الاحياء المائية وبالتالي تؤدي الى عدد من التغيرات الامراضيه و البايوكيميائيه . تم دراسة تأثير مبيد الكلوربايروفوس على المحتوى البايوكيميائي الكلايوجين، الدهن والبروتين في الاحشاء والجبه والقدم عند نهاية تعريض فترات 48 و 96 ساعة من التركيز تحت المميت (0.01, 0.06) ملغم / لتر على القوقع المياه العذبة *Bellamyia bengalensis* . لوحظت اختلافات معنوية في محتوى الانسجة المختلفة لكلا التركيزين خلال (24 و 96) ساعة.