

CHLORDANE INDUCED CHANGES IN CARBOHYDRATE METABOLISM OF THE INDIAN CATFISH *HETEROPNEUSTES FOSSILIS* (BLOCH)

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Anil K. Srivastava and Ashok K. Srivastava (1988) Chlordane induced changes in carbohydrate metabolism of the Indian catfish *Heteropneustes fossilis* (Bloch). Bull. Inst. Zool., Academia Sinica 27(2): 211-215. The Indian catfish *Heteropneustes fossilis*, when exposed to subacute concentrations (0.077 and 0.038 ppm) of chlordane for 15-70 days, showed both hepatic and muscle glycogenolysis with concomitant hyperglycemia. Lower concentrations (0.025 and 0.019 ppm) evoked hyperglycemia but the decrease in tissue glycogen was not significant.

Key words: Chlordane, Toxicity, Fish, Carbohydrate metabolism.

The organochlorine insecticide chlordane (1, 2, 4, 5, 6, 7, 8, 8-octachloro-4, 7-methano-3a, 4, 7, 7a-tetrahydro-indane) is widely used for the eradication of soil pests. It adversely affects non-target organisms, especially freshwater fish (Henderson *et al.*, 1959), which are an important source of protein for human nutrition in developing countries. The highly toxic effects of chlordane on the spawning, hatching success and growth of the sheepshead minnow, *Cyprinodon variegatus* (Parrish *et al.*, 1977) as well as changes in the blood and tissue chemistry of the Indian catfish *Heteropneustes fossilis* (Mishra and Srivastava, 1984) after acute exposure have already been shown. The aim of this investigation was to examine as to what extent short and long term exposure to both subacute and sublethal concentrations of chlordane affects the carbohydrate metabolism in the freshwater Indian catfish *Heteropneustes fossilis*.

MATERIALS AND METHODS

The catfish *Heteropneustes fossilis* (weight, 40.32 ± 3.20 g), collected locally from a freshwater lake, were brought to the laboratory and acclimated in tap water for 15 days under natural photoperiod and ambient temperature ($25.47 \pm 2.4^\circ\text{C}$) in 50-l aquaria. They were fed daily with wheat flour pellets and ground dried shrimp (Srivastava, 1966); the aquaria were cleaned and only the water was replenished once daily. Only healthy fish of both the sexes were used in the experiments.

A static acute toxicity bioassay (APHA *et al.*, 1975) was performed to determine the LC_{50} value (Litchfield and Wilcoxon, 1949) of chlordane; this value for the catfish was 0.386 ppm. A stock solution of chlordane (1 mg/ml) was prepared in acetone. For the study of the effect of the insecticide on carbohydrate metabolism, groups of 30-36 fish (6 fish/20-l glass jar) were exposed to sub-

acute (0.077 and 0.038 ppm) and sublethal (0.025 and 0.019 ppm) concentrations of chlordane in tap water for both short (15-30 days) and long (50-70 days) terms. The average mortality among the treated fish was 10%. Six fish from each group were selected randomly for the analyses of selected variables. Parallel groups each of six fish kept in tap water and receiving equal volume of acetone as the treated fish were sampled at specified time intervals for comparison with the exposed fish.

The fish were anesthetized with 1 g/31 MS 222 (tricaine methanesulfonate) and blotted dry with absorbant paper. The caudal peduncle was cut off with a sharp razor blade and freeflowing blood, collected in citrated tuberculin syringes, was used for the determination of blood glucose levels (Oser, 1965). Muscle and liver glycogen concentra-

tions were measured by the method of Van der Vies (1954). The statistical significance between the treated and control groups was calculated by Student's *t*-test.

RESULTS

The effects of subacute (0.077 and 0.038 ppm) and sublethal (0.025 and 0.019 ppm) concentrations of chlordane on carbohydrate metabolites of *Heteropneustes fossilis* over four different durations are shown in Tables 1 and 2, respectively.

The average muscle glycogen concentration of control fish was between 0.71 and 0.90 mg/100 mg wet weight of tissue. Exposure of fish to 0.077 ppm chlordane resulted in significant reduction in muscle glycogen concentrations from 15 to 70 days. A concentration of 0.038 ppm chlordane caused

TABLE 1
Carbohydrate metabolite values for the catfish *Heteropneustes fossilis* following exposure to subacute concentrations 0.077 and 0.038 ppm of chlordane

Parameters	Exposure period (days)							
	Short term				Long term			
	15		30		50		70	
	Control	Treated	Control	Treated	Control	Treated	Control	Treated
0.077 ppm Chlordane								
Muscle glycogen (mg/100 mg wet wt)	0.90 ±0.13	0.77*** ±0.01	0.86 ±0.03	0.66*** ±0.01	0.75 ±0.01	0.65* ±0.01	0.75 ±0.02	0.64*** ±0.01
Liver glycogen (mg/100 mg wet wt)	13.26 ±0.34	11.33*** ±0.25	12.76 ±0.42	11.36** ±0.23	12.70 ±0.39	10.86 ±0.34	12.97 ±0.36	11.46** ±0.24
Blood glucose (mg/100 ml)	38.68 ±0.96	48.82*** ±0.47	40.56 ±0.35	53.70*** ±1.20	43.57 ±0.61	51.34*** ±0.63	40.12 ±0.75	59.94*** ±0.77
0.038 ppm Chlordane								
Muscle glycogen (mg/100 mg wet wt)	0.88 ±0.05	0.81 ±0.03	0.83 ±0.02	0.79 ±0.06	0.80 ±0.03	0.73 ±0.03	0.81 ±0.02	0.73** ±0.01
Liver glycogen (mg/100 mg wet wt)	13.60 ±0.33	10.86*** ±0.31	12.86 ±0.42	11.00** ±0.37	12.60 ±0.43	11.26 ±0.28	12.35 ±0.32	10.60** ±0.29
Blood glucose (mg/100 ml)	38.21 ±0.86	42.19** ±0.72	36.41 ±0.78	40.92** ±0.81	37.44 ±0.68	47.30*** ±1.10	39.55 ±0.90	46.89* ±1.11

Values are expressed as mean ± SE (N=6), * $p < 0.05$, ** $p < 0.01$ and *** $p < 0.001$.

TABLE 2
Carbohydrate metabolite values for the catfish *Heteropneustes fossilis* following exposure to sublethal concentrations 0.025 and 0.019 ppm of chlordane

Parameters	Exposure Period (days)							
	Short term				Long term			
	15		30		50		70	
	Control	Treated	Control	Treated	Control	Treated	Control	Treated
0.025 ppm Chlordane								
Muscle glycogen (mg/100 mg wet wt)	0.80 ±0.02	0.72 ±0.03	0.78 ±0.01	0.75 ±0.01	0.76 ±0.04	0.71 ±0.03	0.77 ±0.02	0.68 ±0.01
Liver glycogen (mg/100 mg wet wt)	13.80 ±0.48	14.50 ±0.72	13.62 ±0.51	11.80 ±0.35	12.80 ±0.37	11.20** ±0.20	12.75 ±0.31	10.20* ±0.51
Blood glucose (mg/100 ml)	37.13 ±0.01	42.22 ±1.12	40.36 ±0.92	46.25** ±1.15	41.18 ±1.10	45.27 ±1.21	42.68 ±1.14	51.31* ±1.25
0.019 ppm Chlordane								
Muscle glycogen (mg/100 mg wet wt)	0.84 ±0.03	0.78 ±0.06	0.81 ±0.05	0.77 ±0.03	0.79 ±0.02	0.75 ±0.04	0.74 ±0.02	0.60 ±0.05
Liver glycogen (mg/100 mg wet wt)	14.10 ±0.56	12.80 ±0.62	13.86 ±0.47	12.40 ±0.38	13.75 ±0.52	11.96 ±0.61	13.68 ±0.71	11.50 ±0.72
Blood glucose (mg/100 ml)	39.19 ±0.48	44.25*** ±0.62	39.44 ±0.57	41.36 ±0.29	40.92 ±0.28	44.72* ±0.71	40.19 ±0.62	43.36** ±0.58

Values are expressed as mean±SE (N=6); * <0.05, ** $p < 0.01$ and *** $p < 0.001$.

significant decrease in muscle glycogen value at 70 days only, while the sublethal concentrations of 0.025 and 0.019 ppm did not cause any marked reduction in muscle glycogen values at the specified time intervals.

The mean hepatic glycogen content in controls ranged between 12.35 and 14.10 mg/100 mg wet weight of tissue during the experiments. The fish following treatment of 0.077 and 0.038 ppm chlordane showed significant reduction in liver glycogen content from 15 to 70 days. Exposure to 0.025 ppm chlordane evoked hepatic glycogenolysis at 50 and 70 days only, whereas the liver glycogen values were unaltered throughout the experimental period in the fish subsequent to exposed to 0.019 ppm chlordane.

The mean blood glucose level in control fish ranged from 36.41 to 43.57 mg/100 ml. The fish following treatment with 0.077 and

0.038 ppm chlordane showed hyperglycemia from 15 to 70 days after exposure. The fish showed significant increases in glucose concentrations at 30 and 70 days after exposure to 0.025 ppm chlordane. The fish were hyperglycemic at 15, 50 and 70 days of treatment with 0.019 ppm.

DISCUSSION

The results suggest that stress of exposure to high chlordane concentration evokes glycogenolysis with concomitant hyperglycemia. Glycogen is the form of carbohydrate stored in the animals, mainly in the liver and muscles. It may provide a reserve for demands occurring as a result of transient stress (Love, 1980). Previously reported chlordane-induced hyperactivity in *H. fossilis* (Mishra and Srivastava, 1984) would place

additional energy requirements and it is well known that changes in glycogen and glucose follow muscular activity in fish (Black *et al.*, 1962; Beamish, 1968). Furthermore, both organochlorine and organophosphorus pesticides (Srivastava and Singh, 1981; Srivastava and Mishra, 1983; Mishra and Srivastava, 1984) have been reported to decrease muscle and hepatic glycogen content of fishes.

The presence of an insecticide in the body constitutes a stress condition in fish and the common physiological response which follows is an increased stimulation of pituitary and adrenal glands (Murphy, 1969). Thus, the glycogenolysis in muscle and liver in the catfish after exposure to chlordane was most likely due to stress induced increase in circulating catecholamines. Catecholamines deplete tissue glycogen stores in fish (Nakano and Tomlinson, 1967; Larsson, 1973).

Blood sugar levels are maintained at the expense of carbohydrates and protein stores. However, glycogen is the only immediately available reserve of blood glucose. Alteration of blood sugar level is the primary metabolic symptom in vertebrates subjected to stressful situations. Both organochlorine and organophosphorus pesticides (Grant and Mehrle, 1973; Mishra and Srivastava, 1984) produce hyperglycemic effects in fish. Hyperglycemia could also be mediated through inhibited insulin release (Yau and Mennear, 1977). In view of the involvement of certain hormones and/or regulating substances such as insulin, catecholamines, glucagon, and cyclic AMP in carbohydrate metabolism (Terrier and Perrier, 1975; Nilsson *et al.*, 1976), an imbalance in these regulating substances can not be discounted in chlordane exposed catfish.

This work also demonstrated that exposure of fish to sublethal concentrations of chlordane did not bring about marked changes in the tissue glycogen. Apparently, as exposure to chlordane is decreased to 0.019 ppm the catfish are able to overcome at least some of the initial stress imposed by chlordane and maintain tissue glycogen at levels closer to

those found in the untreated fish. Perhaps, the sublethal concentration of 0.019 ppm is near presumably harmless or safe concentration.

It is difficult to assess the causes of disturbed carbohydrate metabolism in fish subjected to chlordane treatment. It may be due to stress or a biochemical mechanism of toxic action of the pesticide.

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Chlordane 致使印度鯰 *Heteropneustes fossilis* (Bloch) 碳水化合物代謝的改變

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當印度鯰暴露於稍為高濃度 (0.077 及 0.038 ppm) 的 Chlordane 中 15~70 天時，肝臟及肌肉都有醣解現象，並伴隨產生多醣症；在低濃度 (0.025 及 0.019 ppm) 時會導致多醣症，但組織中肝醣的減少量則不顯著。

