

Changes in Serum Glucose and Liver and Muscle Glycogen of *Clarias batrachus* Exposed to Lethal and Sub-Lethal Concentrations of NiSO₄

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ABSTRACT

Haematological changes in the blood of *Clarias batrachus* exposed to selected lethal and sub-lethal concentrations of Nickal Sulphate (NiSo₄.6H₂O) at selected periods with its control was studied. The exposure hours were 24, 48, 96, 240, 480, 960 and 1440. Changes in serum glucose, liver and muscle glycogen and its values were investigated in the present study.

Keywords: *Haematological changes, Lethal and sub-lethal concentrations, Nickel sulphate, Serum glucose, muscle glycogen.*

INTRODUCTION

Heavy metals occur naturally in freshwater environment. Some metals are essential for normal growth and development of several organisms in minute concentrations, like Zinc and Selenium, but may be lethal after the limit. Contamination due to various sources has increased the natural levels of metals and now it is known that several heavy metals such as mercury, lead, chromium, copper *etc.* are highly toxic to most organisms and that these are easily soluble in water. Therefore, input into the environment becomes an important aspect of environmental pollution. On the heavy metal toxicity, Pictkering and Handerson (1965) indicated that higher concentration and short-term exposure was more effective than long term or chronic exposure. Kristofferson *et al.*, (1973) have stated that the effects of metal toxicity is depended on involvement of a number of variables such as , test animal, biological parameters, pollutants and organs. Considering the aforesaid facts in mind, the present investigation was undertaken.

MATERIALS AND METHODS

For experiment required number of fish were taken out from each selected concentrations one by one along with control and after blotting the trunk with filter paper, fresh blood samples were collected from the caudal artery by serving the tail and/or by direct heart puncture without using any anaesthesia or anticoagulant between 8-9 AM. Five pooled blood samples were used for the experimental determinations of blood for each concentrations and control separately. Immediate after collecting the blood, required amount of liver and muscle (mid dorso-lateral sides) for each tissue biochemical tests were measured accurately to the nearest milligrams and processed accordingly for glucose/glycogen and other contents following the methods as described after Osker (1965), Natelson (1971) and Varley *et al.*, 1980. For the determination of serum glucose and muscle glycogen O-Toluidine method was applied.

RESULTS AND DISCUSSION

The glucose content in the serum (blood) of control fish varied in between 59.32 ± 2.71 to 65.75 ± 3.46 mg/dl with overall average value of 61.33 ± 2.99 mg/dl, while the glycogen content in the liver and muscle of the normal fish varied in between 29.92 ± 1.42 with an overall average of 31.47 ± 2.08 mg/gm and 7.98 ± 0.19 to 9.00 ± 0.25 with overall average value of 8.43 ± 0.22 mg/gm wet wt. respectively (Table 1). The serum glucose level in the fish exposed to lethal and sublethal concentrations of NiSO_4 , showed an increasing trend which was found statistically significant $P < 0.05$ and $P < 0.01$ at 96 hr and 240 hr (80.80 ± 4.96 & 90.36 ± 3.23 mg/dl *i.e.* 31.75 & 47.33% respectively) of exposures of 175.5 mg/l concentrations and in sublethal concentrations *i.e.* 87.8 & 21.9 mg/l, the increases were found statistically significant ($P < 0.05$) at 480 hr of exposures (84.14 ± 3.76 & 80.95 ± 4.65 mg/dl *i.e.* 37.19 & 31.99% respectively) while it was found significantly increased ($P < 0.01$) at 1440 hr of exposure (82.55 ± 3.14 mg/dl *i.e.* 34.60%) in 87.8 mg/l concentration, whereas the increase was found statistically significant ($P < 0.01$) at 1440 hr (94.27 ± 4.10 mg/dl *i.e.* 53.71%) in 21.9 mg Ni SO_4 /l concentrations.

On the other hand the glycogen content in the liver & muscle showed a gradual decrease in both lethal and sublethal concentrations, which were found statistically significant ($P < 0.05$) & ($P < 0.01$) at 96 & 240 hr of exposure (24.38 ± 1.78 & 19.21 ± 1.60 mg/gm *i.e.* 22.53 & 38.96%) in liver and ($P < 0.01$) at 240 hr (7.08 ± 0.15 mg/gm wet wt. *i.e.* 16.01%) in muscle respectively in the fish exposed to 175.5 mg Ni SO_4 /l and in sublethal concentration (87.8 mg/l) the decline was found statistically significant ($P < 0.05$ and $P < 0.01$) at 960 & 1440 hrs (22.72 ± 2.38 & 19.70 ± 1.42 mg/gm *i.e.* 27.80 & 37.40% in liver and 7.58 ± 0.19 & 7.13 ± 0.17 mg.gm wet wt. *i.e.* 10.08 & 15.42% respectively in the muscle of the fish) exposures, whereas the decline was observed significant ($P < 0.05$) at 1440 hr of exposure to 21.9 mg/l concentration in liver (21.10 ± 2.68 mg/gm *i.e.* 32.95%) and 7.38 ± 0.20 mg/gm wet wt. *i.e.* 12.45% in muscle when compared with the overall respective normal values (Table 1).

Blood glucose and tissue glycogen especially liver, representing energy reserves of the fish, could be used as reliable tools to evaluate the severity of toxication. The elevation of blood glucose level due to toxicants have been reported by several workers (Shalini, 1989; Kumari, 1990; Suraj, 1998 and Gupta, 2003). Shalini (1989) in *H. fossilis* exposed to CaCl_2 and NiSO_4 observed an elevated blood glucose and decreased glycogen contents in the liver of the fish depended on concentrations and exposure periods and stated that the raised value of the blood glucose might be due to enhanced glycogen breakdown (glycogenolysis) due to either anaerobic stress or due to accumulation of respective pollutants in the liver and other organs thus affecting carbohydrate metabolism. On the other hand Sastri and Shukla (1990) in *Channa punctatus* exposed to lethal and sublethal concentrations of cadmium observed a decrease in blood glucose level, indicative of decreased rate of glycogenolysis. Nath and Kumar (1988) in *H. fossilis* exposed to nickel, observed hyperglycemia. Carbohydrate as energy sources are readily utilized during a number of different situations in fish. Muscle and liver glycogen contents decline during stress or acute hypoxia (Burton *et al.*, 1972). Srivastava (1982) evaluated the comparative effects of Cu, Cd and Hg on the tissue glycogen content of the fish *H. fossilis* and suggested that heavy metals in low concentrations act through the endocrine system creating hormone and /or enzyme imbalance rather than other route, while higher concentration may act by producing intense and rapid damage to various systems resulting in hypoxia and electrolyte imbalance.

In the present study, the experiment showed an increasing trend in the blood glucose level which was found statistically significant ($P < 0.05$ and $P < 0.01$) at 96 and 240 hrs of exposure to lethal and ($P < 0.05$ and $P < 0.01$) at 480 and 960 hrs of exposure in sublethal concentrations followed by a decline, but, still significantly ($P < 0.05$) more than the normal value at 1440 hrs of exposures. Whereas, the glycogen content in both liver and muscle showed a gradual decline which were found significant ($P < 0.01$) at 240 hrs in lethal and at 960 and 1440 hrs ($P < 0.00$ and $P < 0.01$) in both liver and muscle in sublethal concentrations when compared with their overall normal values. Thus, an increased blood glucose level with decreased tissue glycogen levels as observed during present study, reveals that the glycogenolysis in the tissue might be due to stress induced increased secretion of catecholamines as it is known that the toxicant, exposure time and strength of stimulus are some important factors governing hormonally mediated changes in tissue glycogen and either glycogenolysis or glycogenesis results.

Table 1
Changes in the serum glucose and liver & muscle glycogen contents in the fish, *C. batrachus* exposed to selected lethal & concentrations of NiSO_4 at selected periods.

concentration of $\text{NiSO}_4 \cdot 6\text{H}_2\text{O}$	log value	Hour of Exposure (hr)	Serum Glucose		Liver Glycogen		Muscle Glycogen	
			(mg/dl)	% Change	(mg/gm)	% Change	(mg/gm)	% Change
Control		24	60.62 ± 2.86		30.66 ± 1.73		8.20 ± 0.23	
175.5	2.444		66.30 ± 3.75	8.1	27.54 ± 2.12	-12.49	7.54 ± 0.18	-7
87.8	1.943		63.58 ± 3.02	3.67	29.18 ± 2.26	-7.28	8.35 ± 0.24	-0.95
21.9	1.34		61.76 ± 3.24	0.7	30.34 ± 2.08	-3.59	8.40 ± 0.25	-0.35
Control		48	61.76 ± 3.24		32.00 ± 1.98		7.94 ± 0.25	
175.5	2.444		59.45 ± 3.38	22.48	26.95 ± 1.85	-14.36	7.94 ± 0.25	-5.81
87.8	1.943		75.12 ± 4.20	15.01	28.46 ± 2.22	-9.56	8.16 ± 0.17	-3.2
21.9	1.34		68.96 ± 3.75	12.44	29.50 ± 1.80	-6.26	8.48 ± 0.26	-2.13
Control		96	62.00 ± 2.64		32.54 ± 2.30		8.48 ± 0.26	
175.5	2.444		80.80 ± 4.96	31.75	24.38 ± 1.78	-22.53	7.60 ± 0.21	-9.84
87.8	1.943		74.34 ± 3.45	21.18	27.75 ± 2.04	-11.82	8.00 ± 0.19	-5.1
21.9	1.34		72.00 ± 2.92	17.4	28.90 ± 1.96	-8.17	7.94 ± 0.23	-5.81
Control		240	64.75 ± 3.46		31.45 ± 2.26		8.36 ± 0.20	
175.5	2.444		90.36 ± 3.23**	47.33	19.21 ± 1.60**	-38.96	7.08 ± 0.15**	-16.01
87.8	1.943		76.14 ± 4.60	24.15	27.52 ± 1.98	-12.55	7.82 ± 0.18	-7.24
21.9	1.34		74.50 ± 5.47	21.47	28.05 ± 1.75	-10.86	7.96 ± 0.26	-5.57
Control		480	59.32 ± 2.71		29.92 ± 1.42		8.25 ± 0.22	
175.5	2.444							
87.8	1.943		84.14 ± 3.76*	37.19	25.69 ± 2.78	-18.37	7.61 ± 0.26	-9.73
21.9	1.34		80.95 ± 4.65*	31.99	26.86 ± 2.02	-14.65	7.90 ± 0.24	-6.29
Control		960	60.20 ± 2.37		30.48 ± 2.27		9.00 ± 0.25	
175.5	2.444							
87.8	1.943		90.23 ± 4.72 **	47.12	22.72 ± 2.38 *	-27.8	7.58 ± 0.19*	-10.08
21.9	1.34		87.80 ± 4.93*	43.24	33.26 ± 2.64	-20.88	7.72 ± 0.22	-8.42
Control		1440	62.96 ± 3.53		33.26 ± 2.64		8.76 ± 0.19	
175.5	2.444							
87.8	1.943		82.55 ± 3.14**	34.6	19.70 ± 1.42**	-37.4	7.13 ± 0.17**	-15.42
21.9	1.34		94.27 ± 4.10**	53.71	21.10 ± 2.68*	-32.95	7.38 ± 0.20*	-12.45
Overall Average value of normal fish as 100%			61.33 ± 2.99		31.47 ± 2.08		8.43 ± 0.22	

“±” is standard error of five observation.

“*” & “**” are significance at 5% & 1% levels respectively

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