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SUBLETHAL EFFECTS OF CHROMIUM ON SOME BIOCHEMICAL PROFILES OF THE FRESH WATER TELEOST, *CYPRINUS CARPIO.*

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ABSTRACT: The Common carp, *Cyprinus carpio* was exposed to sublethal concentrations of chromium for various exposure periods (8, 16, 24 and 32days). Serum glucose, total protein, total cholesterol, serum GOT and GPT were measured both in control and experimental fish. During various exposure periods (8, 16, 24 and 32 days), the levels of serum glucose, total cholesterol, GOT and GPT levelss were ($P<0.05$) significantly elevated in the experimental fish over the control and the serum total protein was decreased significantly ($P<0.05$) in experimental fish.

Keywords: chromium, biochemical parameters, sublethal studies, *Cyprinus carpio.*

INTRODUCTION

Heavy metal contamination in the aquatic environment is a potential threat for aquatic organisms, when exposed to significant amounts of metals as consequences of industrial, agricultural and anthropological activities. Heavy metals at high concentrations can cause harmful effects on metabolic, physiological, and biochemical systems of fishes (Heath, 1987; Folmer, 1993; Death et al., 1999; Yang et al., 2003; Atli and Canli, 2007) and it causes long-term eco-toxicological effects (Starmark and Braunbeck 2000). Fish have been largely used as a bio-indicators for environmental pollutants (Barak and Mason, 1990; Harrison and Klaverkamp, 1990; Saiki, 1990; Winger et al., 1990; Saiki et al., 1993; Kock et al, 1996) and used to estimate the influence of environmental pollution due to the sensitivity of their biochemical and hematological parameters under such conditions (Lopes et al, 2001).

Chromium is also a compound of biological interest, probably having a role in glucose and lipid metabolism as an essential nutrient (Langard et al, 1979). Chromium (VI) compounds have been found to be mutagenic and carcinogenic (Forstner and Wittman, 1979; Venko, 1985). Among the heavy metal, chromium is an important pollutant from industrial effluents and causes deleterious effects on non-target aquatic organism resulting imbalance of an ecosystem (Arun kumar et al, 2004). It is also listed among 25 hazardous substances that pose threat to human health (Irwin et al., 1997).

Works on toxic effect of hexavalent chromium on survival and physiology of fishes (Anbrose et al, 1994; Vutukuru et al, 2000; Sornaraj et al, 1995; Abbasi et al, 1995; Sastry and Sunitha 1984; Mahipaul Singh, 1995), effect on gill of roach (Strike et al., 1975), inhibition of various metabolic processes (Nath and Kumar, 1987), osmoregulatory ability and respiration in fish (Artillo and Melodio, 1988) and various physiological processes of mudskipper B. detanus (Kundu et al.,1995). Biochemical profiles in fish and other aquatic organisms under heavy metal stress serves as important bio-indicators in monitoring of aquatic environment (Abbas and Mahmoud, 2004, Shalaby et al., 2005; Abbas, 2006; Abbas et al., 2007). A literature on biochemical study is limited in Indian context. Therefore, the present study aims to investigate the effect of sublethal concentration of hexavalent chromium on serum biochemical parameters - Glucose, Total protein, Total cholesterol, serum GOT and GPT in common carp *Cyprinus carpio* during different exposure periods (8, 16, 24 and 32 days).

MATERIALS AND METHODS

The Common carp, *Cyprinus carpio* (22gm) were procured from Tamil Nadu Fisheries Development Corporation Ltd. (Fish farm, Mettur, Tamil Nadu, India) and were acclimatized to laboratory conditions (24.2°C) for one month. Fish were fed *ad-libitum* with commercial feed once a day. Since physico- chemical parameters of water have significant influences on the biodegradability and toxicity of pollutants, hydro biological parameters were measured throughout the study period.
Five hundred fish were stocked in a large tank (120 x 180 x 90 cm) after it was cleaned and disinfected with potassium permanganate. Fish with an average weight of 22g were selected for the experiment. Chronic effects of sublethal concentration of the toxicant were studied in four 125-L plastic tubs, filled with clean dechlorinated water. The aquaria were labeled with 'C' and 'T' represented control and experiment respectively. Tub 'T' was filled with sublethal solution of chromium(1/10th of LC_{50} for 96 h) as suggested and tub 'C' was filled with clean dechlorinated water. Then 200 fish were randomly selected from the stock and added to the plastic tub, 100 in each group, 50 in each replicate. After the stipulated time period (8, 16, 24 and 32 days) fish were sacrificed and blood sample was collected from the caudal vein of both control and experimental animals by using non-heparinzed tubes for serum biochemical analysis. The blood was immediately centrifuged at 1500 rpm for 10 min. Serum was then removed and stored at 4ºC prior to immediate determination of biochemical parameters- total glucose, total protein, total cholesterol, Glutamic oxaloacetic acid transaminase (GOT) and Glutamic pyruvic acid transaminase (GPT).
Blood glucose was estimated by using the method of O-Toluidine (Cooper and McDaniel, 1970). Serum total cholesterol was estimated by using the method of Wybenga and Pileggi (1970) and serum total protein was measured according to the procedure of Lowery *et al.* (1951). The activities of serum GPT and GOT were estimated according to the methods of Retiman and Frankel (1957) colorimetric test.
The mean values of the various biological parameters for the control and experimental fish were analyzed for statistical significance of differences using the Student's t- test. $p<0.05$ was taken as the level of significance.

RESULTS

Total Glucose

Total Glucose level in serum of the common carp, *Cyprinus carpio* during sublethal exposure to chromium toxicity recorded overall elevations in its activity level over that of the control (Table-1). The increase of serum glucose was directly proportional to the exposure periods showing a minimum percentage increase of 93.36% at the end of 8th day and a maximum percent increase of 239.40% at the end of 32nd day of treatment.

Total Cholesterol

During sublethal exposure of chromium, total cholesterol level in serum significantly increased in experimental fish than the control (Table-1). The increase of serum cholesterol was directly proportional to the exposure periods. At the end of 8th day, recorded percent increase of 77.24% and at the end of 32nd day of treatment, recorded percent increase of 172.70%.

Total Protein

Total Protein level in serum of common carp exposed to sublethal concentration of chromium toxicity was found to be decreased moderately in experimental fish than the control (Table-1). The decrease of serum protein was directly proportional to the exposure periods. At the end of 8th day, minimum percent decrease of 9.22% and a maximum percent decrease of 43.61% at the end of 32nd day of treatment were recorded.

Table-1.Changes in biochemical parameters of *Cyprinus carpio* exposed to varying periods of sublethal concentrations of chromium.

Parameter	Treatment	Exposure period (days)			
		8	**16**	**24**	**32**
Total Glucose (mg\dl)	C	80.9 ± 14.47	81.8 ±14.63	82.4 ±14.74	86.8±15.53
	T	156.43± 27.96*	177.2± 31.69*	227.8± 40.75*	294.6 ±52.70*
		(+93.36)	(+116.62)	(+176.46)	(+239.40)
Total protein(gm/dL)	C	3.58± 0.595	3.57± 0.639	3.586 ±0.64	3.958 ±0.71
	T	3.25± 0.58	3.01 ±0.539	2.66 ±0.48	2.232± 0.399
		(-9.22)	(-15.69)	(-25.82)	(-43.61)
Total Cholesterol (mg/dl)	C	89.33± 15.98	89.88± 16.08	89.96± 16.09	90.33± 16.16
	T	158.33± 28.32	198.66± 35.54*	211.65± 36.79*	246.33± 44.86*
		(+77.24)	(+121.03)	(+135.27)	(+172.70)
GOT (U/L)	C				
	T	206.66 ±36.97	207.14 ±37.06	208.33± 37.27	207.2± 37.07
		360.36± 64.47	373.33± 66.78	393.66± 70.42*	401.33 ±71.79*
		(+74.37)	(+80.23)	(+88.96)	(+93.69)
GPT(U/L)	C				
	T	48.48±8.69	48.672 ±8.707	48.33 ±8.65	48.66 ± 8.71
		69.54± 12.44	88.35 ±15.81	108.66 ±19.44*	133.66 ±23.91*
		(+43.44)	(+81.52)	(+124.83)	(+174.68)

Values are Mean ±SEM (n=5) * Significant difference (P<0.05).

Serum GOT and GPT

Serum GOT and GPT activity of common carp, during sublethal exposure to chromium toxicity observed overall increases in its activity than the control (Table-1). The increase of serum GOT and GPT was directly proportional to the various exposure periods showing a minimum percent increase of 74.37% and 43.44% at the end of 8th day and a maximum percent increase of 93.69% and 174.69% at the end of 32d day of treatment respectively.

DISCUSSION

Environmental stressors such as metal exposure (Yang and Chen; 2003; Levesque et al; 2002; Zikic et al., 2001) may change the biochemical parameters. Therefore the measurement of serum biochemical parameters can be useful as diagnostic tool in toxicology to find their general health status and target organs affected by toxicants (Zikic et al, 2001; Mc Donald and Grosell, 2006).

The present study demonstrated that the common carp, *Cyprinus carpio* exposed to sublethal concentrations of chromium displayed a significant elevation in the level of blood glucose after all the exposure periods. Similar observations have been reported by Benson et al (1987), Early et al, (1990), Partap and Bonga (1990), Hontela *et al,* (1996), in fish and rat treated with cadmium. Additionally significant hyperglycemia were noted by Nath and Kumar (1988) in the catfish, *Heteropneustes fossilis* exposed to nickel, Radhakrishaniah *et al*., (1992) and Van Vuren *et al* (1994) in the fish *Labeo rohita and Claries gariepinus* subjected to copper and James *et al,* (1992) in the teleost, *Oreochromis mossambicus* exposed to lead.

Wedemeyer and McLeay (1981) reported that high levels of blood glucose are caused by disorders in carbohydrate metabolism appearing in the condition of physical and chemical stresses. A variety of stressors stimulate the adrenal tissues resulting in increased level of circulating glucocorticoids (Partap and Bonga, 1990; Hontela *et al.,* 1996; Gluth and Hanke, 1985; Ricard *et al,* 1998 and catecholamine's (Nakano and Tomlinson, 1967; Witters *et al,* 1991). The elevation in the blood glucose level is response to the increased rate of glycogenolysis or gluconeogenesis. Hyperglycaemic response in this study is an indication of a disrupted carbohydrate metabolism possibly due to enhanced breakdown of liver glycogen and mediated perhaps by adrenocortical hormones and reduced insulin secretory activity. Similar hyperglycemic response has been reported in the flounder Pleuronectes fleusus following exposure to sublethal concentrations of cadmium (Larsson, 1975).

Plasma proteins were decreased significantly with exposure period of chromium. This could be attributed to renal excretion or impaired protein synthesis or due to liver disorder (Kori-Siakpere, 1995). On the other hand, the observed decrease of plasma protein could also result from the breakdown of protein into amino acids first and possibly into nitrogen and other elementary molecules. Similar reduction in protein has also been reported in *Saccobranchus fossils* following exposure to chlordane (Verma *et al.,* 1979). Vutukuru, (2005) reported that there is an appreciable decline in different biochemical constituents in various tissues in fresh water fish *Labeo rohita* under chromium stress. Kori-Siakpere *et al,* (2006) reported that the plasma protein was lowered when *Heterobranchus bidorsalis and Clarias gariepinus* were exposed to sublethal effect of cadmium toxicity.

Cholesterol concentrations in the serum of metal-exposed fish generally increased when compared to the control value (Muazzez et al., 2009). The report of many investigators (Yang and Chen, 2003; Sing and Reddy; 1990; Canli, 1995) support the increase of serum cholesterol concentrations in the metal-exposed fishes. The concentrations of cholesterol is an essential structural components of membranes and the precursor of all steroid hormones, may increase due to the liver failure causing the release of cholesterol into the blood. Heavy metals are known to have hazardous effects on cell structure, especially on the membranes. Therefore, increase in cholesterol may be the indications of environmental stress.

The data of present study showed that the exposure of chromium caused significant elevations in the activities of blood serum GOT and GPT levels. Several reporters showed that these blood enzymes were highly increased in the fish treated with cadmium, zinc and copper (Benson *et al.,* (1987); Atef, (2005); Singh and Reddy, (1990); Hilmy and El-Domaity, (1987); Karan *et al.,* (1998). Shakoori *et al* (1990), reported that the increase in blood enzymatic activity is either due to (i) leakage of these enzymes from hepatic cells and thus raising levels in blood, (ii) increased synthesis and (iii) enzyme induction of these enzymes.

Tietz (1987) and Campbell (1984) reported that these enzymes liberate to the blood stream when the hepatic parenchyma cells are damaged. De smet and Blust (2000) reported that there is an increase in the activities of GOT and GPT in *Cyprinus carpio* exposed to cadmium.

The present study showed that chromium altered the entire biochemical metabolism in *C. carpio* by changing the levels of Total glucose, total protein and total cholesterol and GOT and GPT in serum. Such changes in biochemical levels under the effect of chromium toxicity might results in impairment of energy requiring vital processes, and hence give an idea about the health status of the fish population.

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