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Mercuric Chloride Induced Enzymological Changes in Kidney and Ovary of a Teleost Fish, *Channa punctatus*

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Heavy metal input into the media, either terrestrial or aquatic is an important aspect of environmental pollution. Heavy metals are reported to produce toxic effects on the tissues of various terrestrial and aquatic animals. According to MATHIS & KEVERN (1975) among the aquatic fauna fishes are the most sensitive group. Toxicity of mercurial compounds to fishes has been reported by a number of workers (GRESSON 1970, REHWOLDT et al. 1972). Kidney, liver and brain are the most sensitive organs easily vulnerable to intoxication by mercurial compounds (KLEIN et al. 1972, TAKAHASHI et al. 1971). The toxic effects of mercurial compounds in the digestive system of fishes has been reported by SASTRY & GUPTA (1978a,b). Kidney is one of the most sensitive organs readily responding to mercury intoxication. Very little information is available on the changes in enzyme activities produced by mercury in fishes. Further, mercury can also affect the reproductive system of fishes. Therefore, in the present paper the changes in the activity of a few enzymes in the kidneys and ovaries of a teleost fish, Channa punctatus have been examined.

MATERIAL AND METHODS

Living fishes were collected from local fresh water sources in the month of February, 1978 and maintained in laboratory aquaria. The fishes were allowed to acclimatize for four days to the laboratory conditions prior to experimentation. Preliminary bioassays conducted in the laboratory under static conditions have shown that the LC(50) for 96 hr and the sublethal concentration for 30 days are 1.8 mg/L and 0.30 mg/L of mercuric chloride respectively. 75 fishes weighing 70 $\pm$ 6 g were selected and divided into three equal groups. The first two groups of fishes were treated with LC(50) and sublethal concentration respectively while the third group maintained in mercury free dechlorinated tap water served as controls. 10% (W/V) homogenates of kidney and ovary were prepared in 0.25 M sucrose solution in cold. The homogenates were centrifuged for 20 min at 1500 G and the clear

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supernatant fluids were used as the source of enzymes. 0.016 M sodium-β-glycerophosphate was used as the substrate at pH 5.0 and 9.3 for acid and alkaline phosphatases respectively. The enzyme activity was estimated according to the method of MORTON (1955). For the estimation of glucose-6-phosphatase activity, 0.01 M glucose-6-phosphate solution was incubated for 15 min at pH 6.5. The method of SWANSON (1955) was followed. The inorganic phosphate liberated was quantitatively estimated by the method of FISKE & SUBBAROW (1925). The activity of urease was determined by the method of HOFMANN & SCHMIDT (1953) and lipase activity was estimated following the method of BIER (1955) with Tween 20 as substrate. The protein content in the homogenates was determined by the method of LOWRY et al. (1951) with bovine serum albumin as standard. The test described by FISHER (1950) was employed to calculate the statistical significance between control and experimental values.

RESULTS AND DISCUSSION

The effect of mercuric chloride on the acid and alkaline phosphatases, glucose-6-phosphatase, lipase and urease activities in the kidney and ovary of Channa punctatus has been investigated. The results presented in Table 1 reveal that in fishes treated with LC(50) of mercuric chloride for 96 hr all the enzymes except acid phosphatase are significantly inhibited below the normal level in both the tissues while the latter enzyme showed significant increase in activity in the kidney. Treatment with a sublethal concentration for 30 days also resulted in significant inhibition of enzyme activities in the two tissues but this inhibition was less marked than in fishes treated for 96 hr showing that acute treatment is more toxic. This condition may be due to the new synthesis of enzymes and repair of the damaged tissue. According to PICKERING & HENDERSON (1964) heavy metal toxicity appears to be different at higher concentration and short term treatment than at long term exposure to low concentration. Heavy metals, particularly mercury and its compounds are well known inhibitors of enzymes (BETTIGELLI 1960). According to PASSOW et al. (1961) toxic effects of heavy metals result from their binding with biologically active constituents of the body like lipids, aminoacids and proteins. WEBB (1966) demonstrated alterations in enzyme activities and metabolic processes in animals treated with mercury. CHANG et al. (1973) demonstrated a fall in the activities of alkaline phosphatase and glucose-6-phosphatase and a moderate increase in acid phosphatase in kidney. HINTON et al. (1973) have also noted an initial increase

TABLE 1

Enzyme activities in experimental and control fishes[a]

Enzyme	Tissue	Control	Experimental	
			96 hr	30 days
Acid phosphatase[c]	Kidney	0.189 ± 0.0020	0.203 ± 0.0041(+)b	0.171 ± 0.0014(+)
	Ovary	0.160 ± 0.0030	0.136 ± 0.0014(+)	0.165 ± 0.0010(-)
Alkaline phosphatase[c]	Kidney	0.226 ± 0.0017	0.081 ± 0.0007(+)	0.099 ± 0.0045(+)
	Ovary	0.225 ± 0.0119	0.143 ± 0.0017(+)	0.211 ± 0.0021(-)
Glucose-6-phosphatase[c]	Kidney	0.217 ± 0.0014	0.132 ± 0.0020(+)	0.138 ± 0.0029(+)
	Ovary	0.223 ± 0.0071	0.168 ± 0.0027(+)	0.147 ± 0.0040(+)
Lipase[d]	Kidney	51 ± 0.574	37 ± 0.707(+)	41 ± 1.08(+)
	Ovary	87 ± 1.224	66 ± 0.812(+)	52 ± 0.812(+)
Urease[e]	Kidney	0.0071±0.00077	0.0044±0.000316(-)	0.0059±0.000316(-)

a. Values are Mean ± S.E.
b. (+) indicates statistically significant difference from control values at 95 percent confidence interval.
c. Activity is expressed in mg of inorganic phosphate liberated per mg of enzyme protein per hour at 37°C.
d. Activity is expressed in lipase units.
e. Activity is expressed in mg of nitrogen per mg of enzyme protein per hour at 37°C.

and later fall in acid phosphatase activity due to mercury intoxication. In our earlier histochemical studies (SASTRY & AGRAWAL 1977) similar increase in acid phosphatase activity was observed after 12 hr of mercury treatment but alkaline phosphatase activity was significantly inhibited. The elevation in acid phosphatase activity is probably related to the increase in lysosomal activity in the injured cells occurring as part of prenecrotic changes (DEDUVE 1963, NOVIKOFF 1961). The greater inhibition in alkaline phosphatase activity after 96 hr of treatment with LC(50) in the present study may be due to the increased excretion of mercury by the kidney. Alkaline phosphatase is a brush border enzyme localized in the intestinal mucosa and kidney tubules. According to HICKMAN & TRUMP (1969) this enzyme is involved in the reabsorption of glucose from the renal tubular lumen. The inhibition in activity indicates that the transphosphorylation reactions and absorption of glucose are adversely effected by mercury treatment. The decrease in glucose-6-phosphatase activity may be attributed to the disturbances in the general metabolism of the cell due to mitochondrial damage. According to SUMNER & SOMERS (1953) urease is readily inhibited by heavy metals and the present results also reveal that its activity is significantly inhibited by mercuric chloride.

SUMMARY

The effect of LC(50) (1.8 mg/L) and a sublethal concentration (0.30 mg/L) of in vivo mercuric chloride exposure on the activities of acid and alkaline phosphatases, glucose-6-phosphatase, lipase and urease in the kidneys and ovaries of a teleost fish, Channa punctatus has been studied after 96 hr and 30 days respectively. It has been observed that the activities of all the enzymes except acid phosphatase were significantly inhibited in both the tissues. However, treatment for 96 hr resulted in more marked inhibition than 30 days of treatment. Acid phosphatase showed elevation in activity in the kidney after 96 hr of treatment.

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