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BIOCHEMICAL AND ENZYMOLOGICAL CHANGES IN LIVER AND KIDNEY OF *CLARIAS BATRACHUS* FOLLOWING LITHIUM INTOXICATION

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The present paper deals with some enzymological alterations and glycogen-glucose relationship in the liver and kidney of *Clarias batrachus* in response to lithium nitrate administration. The toxin was administered by intramuscular injections and the dose of 500 μg was repeated every third day. The experimental animals received a total of 7500 μg $LiNO_3$ in 15 injections. The biochemical parameters were measured comparatively in the experimental and control animals.

The results show that the glycogenolysis in the liver was accelerated by lithium administration and that a decrease in hepatic glycogen was associated with increase in glucose. The activities of enzymes AlPase and AcPase increased significantly. RNase and 5'nucleotidase also increased. Decreased lactic dehydrogenase reflected a disturbance in the tricarboxylic acid cycle whereas a rise in GOT and GPT indicated an increase in transamination reactions in the tested tissues.

From industries and agricultural farms, many metals are randomly distributed in the environment affecting fauna and flora in a number of ways. The metals causing water pollution are highly toxic (1, 2, 3). The toxicity of metals to aquatic animals has been demonstrated by different authors (4—6).

Lithium is extensively used in ceramic industry, pharmaceutical works and air conditioning plants. In alloys it serves to improve the tensile strength and is also used as deoxidiser and lubricant. Very little is known about its toxicity to fish (7, 8).

In this paper, an attempt has been made to study some biochemical changes in the liver and kidney of *Clarias batrachus* treated with lithium.

MATERIAL AND METHODS

Live specimens were collected from the local river Kalinadi and acclimatised to laboratory conditions for a week. A total of 40 healthy fishes weighing between 50—70 g were selected irrespective of sex and classified into two groups. The animals from the first group were administered 500 μg of lithium as $LiNO_3$ every third day by intramuscular injection at the base of caudal peduncle. The fishes from the second group (control) were administered an equal amount of distilled water. Both groups of animals were kept at $22 \pm 2\,^{0}C$ and pH 6.8 in laboratory conditions.

After 15 doses (7500 μg $LiNO_3$), the fishes from the experimental group were killed and their livers and kidneys were carefully removed. The tissues were homogenized in respective solvents for biochemical analysis. The homogenates were centrifuged and the clear supernatant fluids were used for measuring alkaline phosphatase (AlPase) acid phosphatase (AcPase) (9), 5'nucleotidase (10), ribonuclease (RNase) (11), glutamic oxaloacetic transaminase, glutamic pyruvic transaminase (GOT and GPT) and lactic dehydrogenase (LDH) (12). The glucose and glycogen levels were also determined in the liver and kidney (13). The data were analysed by Student's t-test (14).

RESULTS AND DISCUSSION

The data of biochemical analysis in the liver and kidney of *Clarias batrachus* are presented in Table 1.

A significant increase in the activities of phosphatases (AcPase and AlPase) is evident. It is assumed to be due to the cytotoxic effect of lithium (15). A rise in phosphatase activity in the liver and kidney of *Channa punctatus* during exposure to 2,3,4-triaminoazobenzene has also been reported (16). However, a decrease in phosphatase activity is accounted for by a leakage from the affected tissues into the blood (17).

The activity of hepatic ribonuclease and 5'nucleotidase increased and was related to the malfunctioning of liver. However, the ribonuclease activity in the kidney significantly decreased. Elevated serum ribonuclease activity has been reported in lithium intoxicated fish, *Heteropneustes fossilis* (18). It has been demonstrated that decreased activities of these enzymes in animals are due to liver injuries (19). The activities of ribonuclease and 5'nucleotidase have been reported decreased in the gill of *Clarias batrachus* under aminoazostress (20). Enhanced activity of 5'nucleotidase is an indication of disrupted hydrolysis of nucleotides.

A significant decrease has been recorded in the activity of lactic dehydrogenase, which seems associated with the disturbance in tricarboxylic acid cycle and glycolytic pathway. Similarly, a decrease in

Table 1

Biochemical alterations in the liver and kidney of Clarias batrachus treated with lithium nitrate. All values are the means ± standard errors of six estimations.

Enzymes and constituents	Liver			Kidney		
	Control	Experimental	Change (%)	Control	Experimental	Change (%)
Glycogen (mg/g)	35.75±0.22	27.94±0.56[a]	—22	10.12±1.80	7.34±0.49	—27
Glucose (mg/g)	24.99±0.22	30.23±2.03	+21	13.17±0.13	17.39±1.05[b]	+26
AlPase (mg iP/g)	0.82±0.03	0.96±0.03[b]	+17	4.27±0.00	5.04±0.06[a]	+18
AcPase (mg iP/g)	0.55±0.03	0.63±0.02	+15	0.790±0.02	1.00±0.20	+27
RNase (mg iP/g)	0.94±0.02	1.18±0.03[a]	+25	1.79±0.00	1.37±0.06[a]	—23
5'nucleotidase (mg iP/g)	0.80±0.08	1.48±0.17[b]	+83	1.79±0.27	2.56±0.10[b]	+43
Lactic dehydrogenase (unit/g)	132.04±0.76	110.12±2.31[a]	—16	71.04±0.66	53.26±1.39[a]	—25
GOT (unit/g)	24.36±0.60	28.71±0,61[b]	+18	13.85±0.63	16.30±0.46	+18
GPT (unit/g)	20.80±1.48	22.54±0.40	+8	10.65±0.34	11.59±0.23	+9

Levels of significance: [a]$P<0.01$; [b]$P<0.05$

lactate dehydrogenase activity has been observed in the liver, brain and testis of manganese treated rats (21). The LDH activity has also been decreased in the kidney of rats treated with mercury (22) and in the digestive system of *Channa punctatus* exposed to $PbNO_3$ (23).

Increased activities of transaminases (GOT and GPT) indicate the increase in transamination reactions i.e. transfer of $—NH_2$ group from an amino acid to a keto acid. A similar rise in the activities has been noted under the effect of azodye (24). However, a decrease in the activities of both enzymes has been reported during the treatment with phenylene brown in teleost fish (20).

A decrease in glycogen is associated with its regular breakdown in glucose resulting in a marked rise in glucose level. The exhaustion of glycogen as a source of energy may be considered as a possible cause of cell death. A decrease in glycogen under the effect of methyl mercury and lead has also been demonstrated (25, 26). A decrease in tissue glycogen after prolonged exposure to water contaminants may be due to their interference with the activities of the enzymes actively involved in its synthesis (27).

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Sažetak

BIOKEMIJSKE PROMJENE U JETRI I BUBREZIMA RIBE *CLARIAS BATRACHUS* TRETIRANE LITIJEVIM NITRATOM

U radu se iznose promjene enzimskih aktivnosti te odnos koncentracija glikogena i glukoze u jetri i bubrezima ribe *Clarias batrachus* nakon višekratne intramuskularne aplikacije litijeva nitrata. Pokusne ribe primile su ukupno 750 μg $LiNO_3$ u 15 injekcija, dok su kontrolne životinje primale destiliranu vodu.

Rezultati su pokazali da je glikogenoliza i u jetri i u bubrezima bila ubrzana davanjem litija, jer je nastalo smanjenje glikogena uz istodobno povećanje koncentracije glukoze. Aktivnosti alkalne i kisele fosfataze značajno su porasle. Ribonukleaza i 5'nuklcotidaza također su imale veću aktivnost u tretiranih životinja. Smanjenje aktivnosti mliječne dehidrogenaze pripisuje se poremećenju ciklusa trikarboksilnih kiselina. Porast aktivnosti GOT i GPT upućuje na pojačanje transaminacijskih reakcija u jetri i bubregu.

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