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Effect of Endosulfan on ATPase Activity in Liver, Kidney and Muscles of *Channa punctatus* and Their Recovery Response

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Abstract

The specific activities of Na^+-K^+ ATPase and Mg^{++} ATPase were investigated in liver, kidney and muscles of a fresh water fish, *Channa punctatus* at the interval of 15 and 30 days exposure to 1/5th of 96 hours TL_m of endosulfan, i.e., 0.00033 mg/l. The activities were found to be inhibited significantly. The recovery response of adverse effects of the exposure was also carried out. The inhibition in activity of Na^+-K^+ ATPase after 15 days exposure was 37.56%, 31.65% and 34.34% which was recovered after 15 days in toxicant free water up to the levels of 12.24%, 8.68% and 11.17% in liver, kidney and muscles, respectively The activity of Mg^{++} ATPase was inhibited by 41.28%, 38.42% and 39.85% which was found to be recovered up to 13.38%, 11.21% and 12.72%in the liver, kidney and muscles respectively. The activity of Na^+-K^+ ATPase after 30 days exposure was inhibited to 52.79%, 43.63% and 51.11% and after 30 days in normal water that was recovered up to 14.89%, 10.67% and 14.37% and inhibition of Mg^{++} ATPase was found to be 63.86%, 58.26% and 59.79% and after recovery it was found to be 13.11%, 10.33% and 12.15% in liver, kidney and muscles respectively. The order of inhibition in the activity of ATPase was found to be liver>muscles>kidney and the order of recovery of the activity of ATPase was found to be contradictory like kidney>muscles>liver. This alteration in the activity of ATPase may alter cellular metabolism which may in turn result to reallocate the fish physiology.

Keywords: *Channa punctatus*, endosulfan, liver, kidney, muscles, ATPase, recovery.

1. INTRODUCTION

The large scale use of chemicals to protect crops from pests and fungi, and to control vectors of diseases such as insects and increasing use of weedicides and growth promoting substances to increase food production have begun to cause serious concern about chronic toxic effects in living beings. The introduction of these pesticides into environment seriously caused deterioration especially to the aquatic environment (Dalela et al., 1978; Reidel and Christenson, 1979).There are several evidences which show that pesticides caused alterations in physiological and biochemical activities (Vermaet al., 1979; Bansal and Chandra, 1985; Sharma, 1988).Since all the cellular processes are essentially enzyme catalyzed, the consequence of the interaction in between the toxic agents and living systems are generally cause the disturbance of enzymatic reactions such as adenosine triphosphatase (Anjum and Siddiqui, 1990; Sharma et al., 2010; El-Elaimyet al., 2014).The effects of the pesticide endosulfan on enzymatic activity of Adenosine Triphosphatase (ATPase) have been studied

in present work. The experiments for enzymatic study were carried out in liver, kidney and muscles of *Channa punctatus*. The reversal effects for activities of these enzymes have also been studied.

2. MATERIALS AND METHODS

A fresh water fish *C. punctatus* has been selected as test animal for the present work. One of the very widely used pesticide endosulfan (trade mark as "mentor") has been used as test chemical. The test fish *C.punctatus* ranging in size from 7to 12 cm (wt. 35 to 80 g) were collected from the neighboring fresh water resources. The fish were observed for any pathological symptoms and were washed with 0.1 % potassium nitrate ($KMnO_4$) solution for 2 minutes to avoid dermal infection. The fish were then rinsed in water and acclimatized to the laboratory conditions for a period of two weeks in a rectangular tank containing tap water.

After acclimatization the fish were exposed chronically to the 1/5th of 96 hours TLm of endosulfan for fish i.e. 0.00033 mg/l. for a period of 15 and 30 days. The solutions were renewed after every 48 hours interval and fish were fed on alternate days to avoid any histological and biochemical change due to starvation. Any fish found dead was removed immediately with the fear that such mortality may deplete dissolved oxygen (DO) with resultant effects on other fish. Control experiments were also run side by side for 15 and 30 days under similar conditions. After definite period of exposure, six fish were sacrificed and liver, kidney and muscles were removed carefully, washed, blotted and weighed. ATPase activity was measured by a Continuous method (Pullman et al., 1960; Fritz and Hamarik, 1966). Protein concentration was measured using Biuret Method (Henry and Winekelman, 1974).

3. RESULTS

The activity of Na^+-K^+ ATPase and Mg^{++} ATPase was found to be inhibited significantly and the recovery response of the adverse effects of exposure was also carried out which is given in Tables 1 and 2.

Table 1: Activity of Na^+-K^+ ATPase in liver, kidney and muscles of *Channa punctatus* in control, and experimental conditions endosulfan and their recovery for 15 and 30 days

Days	Treatment	Specific Activity µ moles of inorganic phosphate liberated /mg of protein /h		
		Liver	Kidney	Muscles
15 days exposure	Control	0.4145±0.053488	0.8186±0.082098	0.2347±0.014639
	Endosulfan treated	0.2588±0.016146	0.5595±0.03230	0.1541±0.009246
	% Alteration	37.56**	31.65**	34.34**
15 days recovery	Control	0.4116±0.053484	0.8182±0.08207	0.2336±0.014639
	Endosulfan treated	0.3612±0.015702	0.7472±0.059381	0.2075±0.012480
	% Alteration	12.24**	8.68**	11.17**
30 days exposure	Control	0.4116±0.053484	0.8182±0.08207	0.2336±0.014639
	Endosulfan treated	0.1943±0.009706	0.4612±0.014187	0.1142±0.007779
	% Alteration	52.79**	43.63**	51.11**
30 days recovery	Control	0.4108±0.053432	0.8173±0.082099	0.2331±0.014426
	Endosulfan treated	0.3496±0.014229	0.7301±0.045889	0.1996±0.010152
	% Alteration	14.89**	10.67**	14.37**

*mean±S.E. of six observations; **significant at P<0.01

The inhibition in the activity of Na^+-K^+ ATPase after 15 days exposure was 37.56%, 31.65% and 34.34% which was recovered after 15 days in toxicant free water up to the levels of 12.24%, 8.68% and 11.17% in liver, kidney and muscles, respectively (Fig. 1). The activity of Mg^{++} ATPase was inhibited by 41.28%, 38.42% and 39.85% which was found to be recovered up to 13.38%, 11.21% and 12.72% in

liver, kidney and muscles respectively in a single fortnight exposure (Fig. 2). The activity of Na^+-K^+ ATPase after 30 days exposure was inhibited to 52.79%, 43.63% and 51.11% and after 30 days in normal water that was recovered up to 14.89%, 10.67% and 14.37% in liver, kidney and muscles respectively (Fig. 1) The inhibition of Mg^{++} ATPase for one month exposure was found to be 63.86%, 58.26% and 59.79% and after recovery it was found to be 13.11%, 10.33% and 12.15% in liver, kidney and muscles respectively (Fig. 2). The order of inhibition in the activity of ATPase was found to be liver>muscles>kidney. However, order of recovery of the activity of ATPase was found to be kidney>muscles>liver.

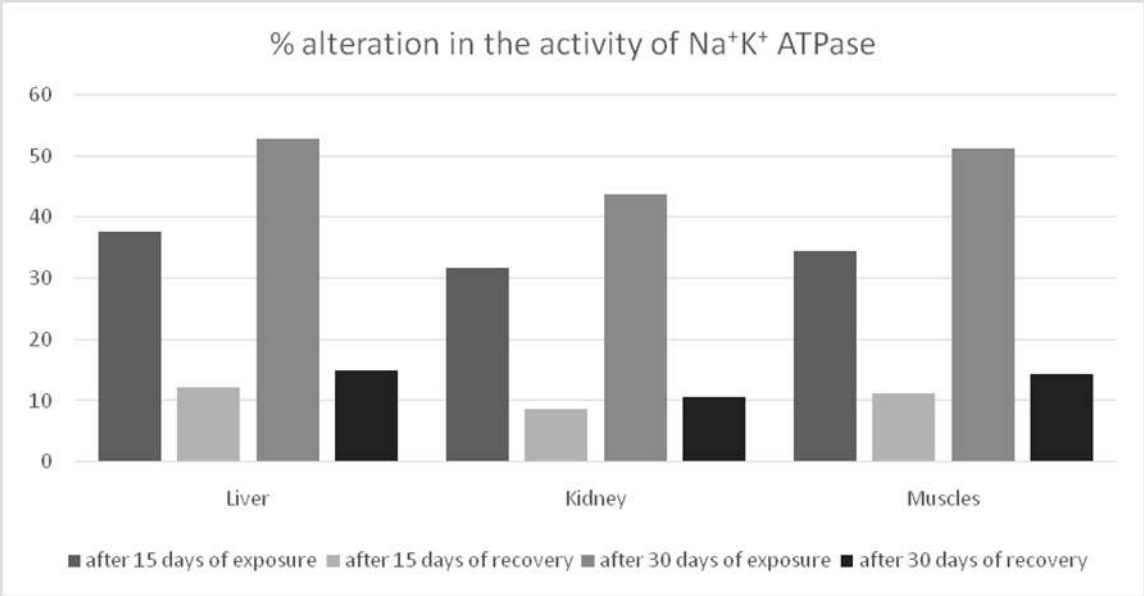

Fig. 1: Histogram showing % alteration in activity of Na^+-K^+ ATPase in liver, kidney and muscles of *Channa punctatus* after treatment with endosulfan and their recovery.

Table 2: Activity of Mg^{++} ATPase in liver, kidney and muscles of *Channa punctatus* in control, and experimental conditions endosulfan and their recovery for 15 and 30 days

Days	Treatment	Specific Activity μ moles of inorganic phosphate liberated /mg of protein /h		
		Liver	Kidney	Muscles
15 days exposure	Control	0.8440±0.082624	0.7161±0.045372	0.8136±0.080460
	Endosulfan treated	0.4956±0.038492	0.4409±0.037164	0.4893±0.036429
	% Alteration	41.28**	38.42**	39.85**
15 days recovery	Control	0.8371±0.063768	0.7071±0.045118	0.8092±0.06463
	Endosulfan treated	0.7251±0.045083	0.6278±0.044748	0.7063±0.045061
	% Alteration	13.38**	11.21**	12.72**
30 days exposure	Control	0.8371±0.063768	0.7071±0.045118	0.8092±0.06463
	Endosulfan treated	0.3025±0.012961	0.2951±0.012147	0.3254±0.011522
	% Alteration	63.86**	58.26**	59.79**
30 days recovery	Control	0.8365±0.063768	0.7063±0.045118	0.8084±0.06463
	Endosulfan treated	0.7268±0.045083	0.6333±0.044748	0.7102±0.045083
	% Alteration	13.11**	10.33**	12.15**

*mean±S.E. of six observations; **significant at P<0.01

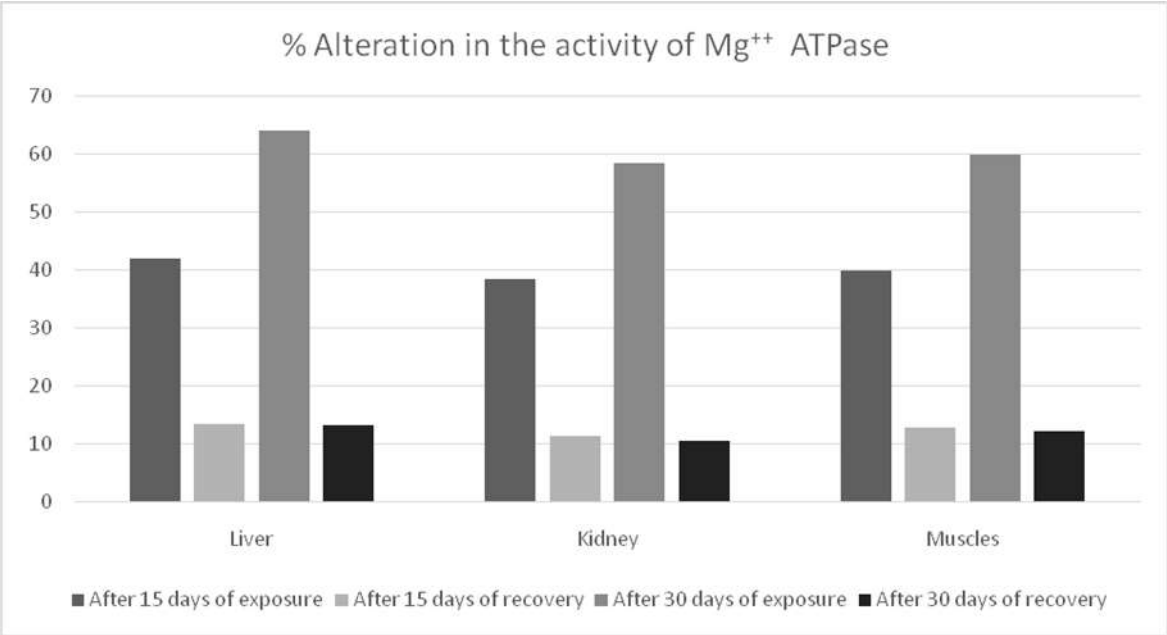

Fig. 2: Histogram showing % alteration in activity of Mg^{++} ATPase in liver, kidney and muscles of *Channa punctatus* after treatment with endosulfan and their recovery.

4. DISCUSSION

ATPase was involved in osmoregulation and intracellular functions. Na^+-K^+ ATPase was a biochemical expression of active transport which was responsible for reciprocal transfer of Na^+ and K^+ across the plasma membrane of cells (Skou, 1961; 64). The enzyme was therefore, fundamental to such functions as the generation of membrane potentials and maintenance of tissue osmolarity. Mg^{++} ATPase was involved in oxidative phosphorylation and thus responsible for biosynthesis of ATP in mitochondrial membrane. Data showed that the order of inhibition in activity of ATPase was found to be liver>muscles>kidney. However, order of recovery of activity of ATPase was found to be kidney>muscles>liver. The inhibition was more pronounced in liver than kidney and muscles. Vermaet al., (1979) observed inhibition in activity of ATPase in brain and liver of *Labeo rohita* and *Saccobranchus fossilis* following *in vitro* treatment of chlordane and found more inhibition in brain than liver. Sharma (1988) found decline in the activity of Na^+-K^+ ATPase and Mg^{++} ATPase in liver and muscles of *C.gachua*, exposed chronically to endosulfan and the order of inhibition was liver>muscles. Chandravathy and Reddy (1995) also noticed similar order of inhibition in *Anabas scandens* due to lead exposure. Balajiet al. (2015) also observed decreased activity of Na^+ and K^+ ATPase in cypermethrin exposed *Cyprinus carpio*. Na^+-K^+ ATPase was involved in active transport of Na^+ and K^+ of cells and the alteration in activity of it results variation in metabolism of cells, especially in nerve cells. The decline in ATPase also affected cellular metabolism (Chitraet al., 1983) Since the inhibition of Mg^{++} ATPase activity disturb the energy yielding process it might cause alteration in the contraction mechanism of muscles due to less energy liberation for contraction. This view was in agreement with Bansal and Chandra, (1985). Chinoy (1991a; b) has also observed the changes in physiology of various animals and human beings due to the inhibition of ATPase. Alteration in ATPases activity in *Cirrhinus mrigala* after exposure to deltamethrin was also observed by David et al. (2014).

Significant recovery was observed in activity of Na^+-K^+ ATPase and Mg^{++} ATPase in all the tissues of treated fish after transfer to uncontaminated water. Reddy et al. (1991) also observed recovery in the

activity of Na^+-K^+ ATPase and Mg^{++} ATPase in endosulfan treated crab after transfer to the clean water. Recovery of ATPase poisoning in *Clarias batrachus* exposed to carbamate as studied by Begum (2009). The activity of Na^+-K^+ ATPase and Mg^{++} ATPase found to be recovered in different tissues of carbofuran treated *C.batrachus* by Begum (2011). Peter et al. (2013) also reported ATPase recovery in several tissues of carbaryl treated *A. testudineus*. Singh et al. (2016) also observed the recovery of activity of Na^+-K^+ ATPase and Mg^{++} ATPase in cyfluthrin exposed *C. punctatus*.

5. CONCLUSION

The findings of the current investigation concluded that small changes in ionic composition that are regulated by ATPase would result in change in physiological patterns and could cause reallocation and modification of behavioral response of fresh water snake headed fish, *Channa punctatus*.

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