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## Comparative toxicity of Butachlor, Imidacloprid and Sodium fluoride on protein profile of the walking cat fish *Clarias batrachus*

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### ABSTRACT

Freshwater cat fish *Clarias batrachus* was exposed to Butachlor, Imidacloprid and Sodium fluoride with lethal and sub-lethal concentration for 72 hrs and 21 days durations. Protein changes in liver and muscles were analyzed after exposure period. Imidacloprid and sodium fluoride caused remarkable protein loss lethal concentration but at sub-lethal level their toxicity was moderate. But Butachlor caused remarkable protein loss at lethal as well as sublethal concentration.

**Keywords:** Protein, Butachlor, Imidacloprid, Sodium fluoride,

### INTRODUCTION

The walking catfish, (*Clarias batrachus*), is a species of freshwater air-breathing catfish found primarily in Southeast Asia, so named for its ability to "walk" across dry land, to find food or suitable environments. This fish normally lives in slow-moving and often stagnant waters in ponds, swamps, streams and rivers, flooded rice paddies or temporary pools which may dry up. When this happens, its "walking" skill allows the fish to move to other sources of water. It is a voracious eater which consumes food rapidly and this habit makes it a particularly harmful invasive species. *Clarias batrachus* is generally considered to be one of the most important catfish species for aquaculture as well as for its economic value as food in almost all over India. Today we have attempted to increase the world's food production to solve the problem of malnutrition and we have achieved this by increased use of fertilizer to nourish the plant and by increased use of pesticides, insecticides to protect them from pests. Recently a large quantity of pesticides and fertilizers are used to nourish the plants and food production. These chemicals have entered into the aquatic system and create pollution, which poses a great threat to aquatic organisms. There are several reports regarding the effects of pesticides (Arunachalam *et al.*, 1985) on physiology of fish. The pesticides used in pest control programmes also produce many physiological and biochemical changes in freshwater organisms particularly the fish (Girija, 1984). Although some data available on the effects of different pesticides on the biochemical aspects of fish gill. The alteration in biochemical contents in different tissues of fish is due to toxic effects of different heavy metals and pesticides have been reported by many workers (Saxena *et al.*, 1989; Khan *et al.*, 1992; Virk and Sharma, 1999; Rawat *et al.*, 2002).

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Neuhold & Sigler (1960) reported that the accumulation of drinking water fluoride currently is being suspected as a cause for the decline of trout fishery. It was also reported that trout's critical habitat has been found to have measurable levels of fluoride that was involved with delayed migration. The fluoride minerals or fluoride-rich minerals in the rocks and soils and their dumping into the water bodies, are the main cause of high fluoride contents in fishes. Since the fluoride content in water bodies is increasing day by day due to enhance uses of domestics and industrial products, it has become a serious threat to aquatic organisms. In the present communication authors studied the toxicity responses induced by Butachlor, Imidacloprid and Sodium fluoride, in freshwater cat fish, *Clarias batrachus* and reported the adverse affects of these toxicants on the protein profile of the fish.

**MATERIAL AND METHODS**

*Clarias batrachus*, were collected and brought to laboratory and kept in aquaria for a week using aged water for acclimatization. During acclimatization they were fed on every day. The acclimated fishes were exposed to Butachlor (4.00 ppm lethal, 2.00 ppm sub-lethal), Imidacloprid (4.00 ppm lethal, 2.00 ppm sub-lethal) and Sodium fluoride (1200 ppm lethal, 600 ppm sub-lethal for 72 hrs for 21 days. Simultaneously a control group of healthy fishes were maintained under identical conditions. The fishes were sacrificed immediately at the end of exposure period, liver and muscle were isolated and used to investigate biochemical contents under toxicant stress. Protein content was estimated by Follin phenol reagent method (Lowry *et al.*, 1951).

**RESULTS AND DISCUSSION**

During present investigation Imidacloprid caused significant protein loss at lethal concentration (-19% in muscles and -49% in liver) where as at sub-lethal level protein loss was found (-5% in muscles and -27% in liver). In Sodium fluoride exposure protein loss was (-36% in muscles and -43% in liver) at lethal concentration and (-3% in muscles and -17% in liver) at sub-lethal level. Butachlor was found highly toxic at both lethal and sub-lethal concentration with (-53% in muscles and -50% in liver) at lethal exposure and (-36% in muscles and -29% in liver) at sub-lethal concentration. The results obtained in the present investigation are summarized in Table 1, Fig 1(A,B,C) and Fig 2.

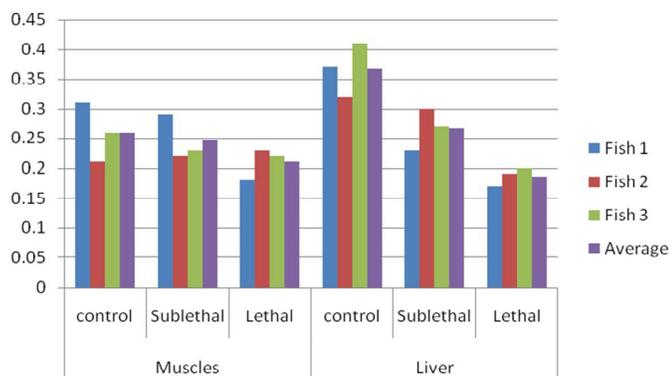


Fig. 1a: Effects of Imidacloprid toxicity on the fish protein.

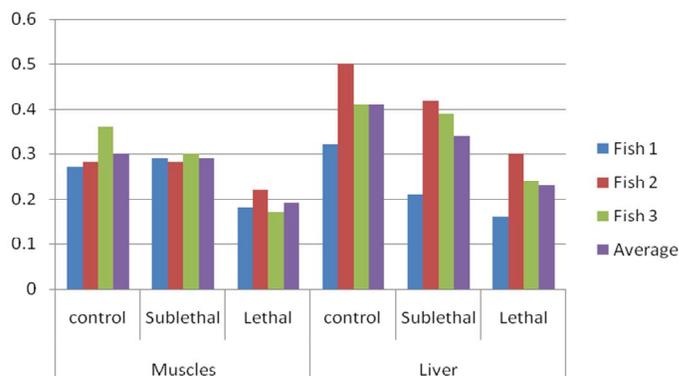


Fig. 1b: Effects of Sodium fluoride toxicity on the fish protein.

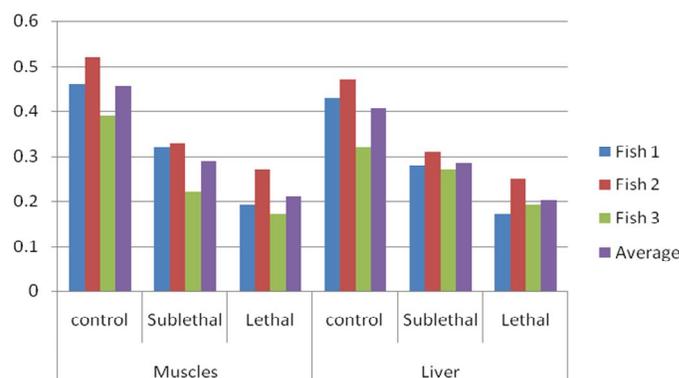


Fig. 1c: Effect of Butachlor toxicity on the fish protein.

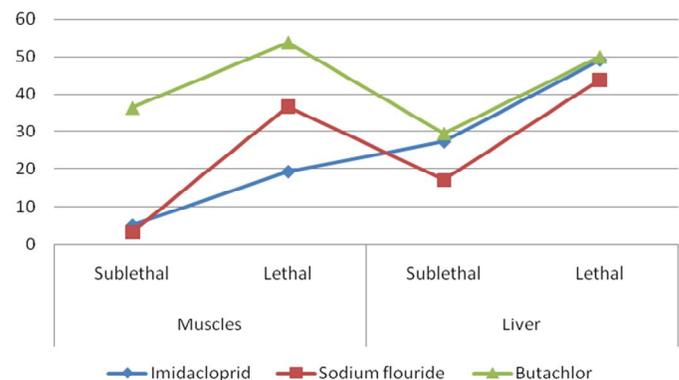


Fig. 2: Percentage of changes reported in fish proteins.

Liver is the primary organ for detoxification (Hulterer *et al.*, 1969) and hence it is expected that toxicant could reach there for detoxification and disposal. This results in structural changes in the liver, the arrangement of hepatic cords leading to the alteration of liver metabolism and its biochemical content. The pollutants acts as one kind of stress and organism respond by developing necessary potential occurring in body give first indication of stress. During stress as organism needs sufficient energy which is supplied from reserve food material *i.e.* protein, glycogen, cholesterol etc. Decrease in the protein content was observed throughout the exposure period. The sublethal exposure results shows the protein content decreases are depend upon the concentration. There is progressive decrease in the protein content with increase in concentration. The toxicity of Imidacloprid, Butachlor and Sodium flouride showed a direct correlation with the

concentration and time exposure. Similar observation has been observed by Singh and Bhati (1994).

Decrease in protein may be due to the impairment of protein synthesis or increase in the rate of its degradation to amino acids. This may be fed to TCA cycle through amino transferase probably to cope with high energy demands in order to meet the stress condition. The decrease in proteins might be due to their utilization in cell repair and tissue organization with the formation of lipoproteins, which are important cellular constituents of cell membranes and cell organelles present in cytoplasm (Harper, 1983). The decrease in protein content as a result of toxicity stress has already being reported by Borah and Yadav (1995) and Muley *et al.*, (2007). Singh (1988) reported reduction in protein content of liver of *Clarias batrachus* in response to Malathion and Y-BHC. Saxena *et al.*, (1989) attributed the decrease in protein content due to decreased protein synthesizing capacity of liver of *Channa punctatus* exposed to carbaryl and malathion. Jones and Kumar (1996) also observed decline in protein content in liver of *Etrophis maculates* under Ekalux stress. Choudhary and Gaur (2001) observed decline in liver protein of *Cyprinus carpio* under sodium fluoride stress. Shobha *et al.*, (2007) reported decrease in protein, glycogen and lipid contents in the liver of freshwater fish, Catla catla under Cadmium Chloride stress.

The decrease in protein liver during dimethoate exposure may be due to increased catabolism (Begum and Raghawan, 1995) and decreased anabolism of proteins (Khare and Singh, 2002). The loss of protein under the Imidacloprid, Butachlor and Sodium flouride stress noticed in the present study may be due to the utilization of amino acids in the various catabolic reactions (Jones and Kumar, 1996). Decrease in protein content may be due to increased proteolysis (Muley *et al.*, 2007) or it may be due to metabolic utilization of the ketoacids to glucogenesis pathway for synthesis of glucose (Schmidt Nielson, 1975). The alteration in protein value may also be related to some structural changes

in the liver, the arrangement of hepatic words leading to the alteration of liver metabolism.

The decrease in liver protein is also attributed to the inhibition of protein synthesis. Liver glycogen content decreased progressively during exposure period, this may be due to toxic stress. During stress an organism needs sufficient energy which is supplied from reserved glycogen. Glycogen is stored in the organism mainly in the liver and muscles in the form of carbohydrate. It may provide a reserve food for acute demands recurring as a result of transient stress (Love, 1980). A fall in the glycogen level clearly indicates its rapid utilization to meet the enhanced energy demands in fish exposed to toxicants through glycolysis or hexose monophosphate pathway. It is assumed that decrease in glycogen content may be due to the inhibition of hormones which contribute to glycogen synthesis. Radhaiah *et al.*, (1987) observed decreased carbohydrate content in heptachlor intoxicated fish *Tilapia mossambica* and stated this may be due to the rapid utilization of carbohydrates by the tissue possibly to overcome the pesticides induced stress. James and Sampath (1995) observed decreased liver glycogen in the *Heteropneustes fossilis* (Bloch) under mixtures of copper and ammonia and reported glycogenolysis releasing glucose in to the circulatory system to meet increased energy demand during stress conditions Susan *et al.*, (1999) observed drastic decreased glycogen content in liver of *Catla catla* under fenvalerate toxicity stress. Rawat *et al.*, (2002) have reported continuous decrease in quantity of glycogen in the liver of *Heteropneustes fossilis* exposed to endosulfan. Tiwari and Singh (2009) observed decrease in total protein and glycogen in the liver of *Colisa fasciatus*, exposed to ethanolic extract of Nerium indicum mill latex. Decrease in the glycogen level in liver suggests the possibility of glycogenolysis. A study indicating such depletion in fish models (Mishra and Srivastava, 1984) during organophosphorus toxicity offers an excellent support to the decreasing levels of proteins in the present study.

**Table. 1:** Change in Proteins induced by Imidacloprid, NaF and Butachlor in *Clarias batrachus*.

Toxicant	Control	Sub-lethal	Change in %	Lethal	Change in %
<b>Imidacloprid</b>	<b>Muscles</b>				
	0.261*±0.051	0.246**±0.037	-5.12	0.212*±0.026	-19.23
	<b>Liver</b>				
	0.366*±0.045	0.266*±0.035	-27.27	0.186*±0.015	-49.09
<b>Sodium fluoride</b>	<b>Muscles</b>				
	0.312**±0.049	0.285*±0.011	-3.33	0.191*±0.026	-36.66
	<b>Liver</b>				
	0.412*±0.092	0.342*±0.113	-17.07	0.231**±0.070	-43.92
<b>Butachlor</b>	<b>Muscles</b>				
	0.456*±0.065	0.293*±0.060	-36.44	0.214**±0.052	-53.94
	<b>Liver</b>				
	0.406*±0.077	0.286**±0.020	-29.55	0.203*±0.041	-50.0

The values are expressed in mg/100 mg dry weight (mean±SD) [\*-P<0.05; \*\*=P<0.01.]

## CONCLUSION

With the help of present investigation, Imidacloprid and sodium fluoride were reported highly toxic but at lethal dose because remarkable protein loss was reported at lethal concentration but at sub-lethal level their toxicity was moderate. But Butachlor caused remarkable protein loss at lethal as well as sub-lethal concentration. Hence, it may be concluded that the agricultural use of butachlor must be checked especially near the water bodies.

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