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IJPBS |Volume 5| Issue 3|JUL-SEPT|2015|145-149

Research Article
Biological Sciences

EFFECT OF HERBICIDE (PRIMEXTRA) ON TISSUE PROTEIN LEVELS IN *Clarias albopunctatus*

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ABSTRACT

The Chloroacetanilide and Traizine herbicide, primextra is widely used in agriculture for weed control in maize and sorghum farmlands in many areas of Nigeria. These chemicals, when discharged to receiving waters cause severe damage to aquatic ecosystem especially to fishes. The aim of this study is to evaluate the impact of primextra on tissue proteins levels in Clarias albopunctatus. Juvenile Clarias albopunctatus were exposed to sublethal concentrations (0.04 µg/L, 0.06 µg/L and 0.10µg/L) of primextra for 21 days in a static renewal bioassay system. The changes in some tissues (liver, muscle, blood) protein levels were determined every seven days. The result indicated that primextra had adverse effect on the tissue protein levels in C. albopunctatus. When compared with the control, the liver, muscle and blood protein levels decreased significantly (P>0.05) due to primextra exposure. The induction of hypoproteinemia in the liver, blood and muscle of the treated fish are indications of dysfunctional protein physiological processes occurring in the fish due to primextra exposure.

KEY WORDS

Blood, Clarias gariepinus, Liver, Muscle, Protein, Primextra.

INTRODUCTION

The contamination of aquatic ecosystem with a wide range of pollutants has become a matter of great concern, not only because of the threat to public water supplies, but also with the damage caused to the aquatic life [1]. Over the last few decades, in many African countries, a considerable population growth has taken place accompanied by a steep increase in urbanization, industrial and agricultural land use [2, 3, 4]. This has entailed a great increase in discharge of pollutants to receiving waters, causing undesirable effects on the aquatic environment. The water contamination cause damages to aquatic life especially to fishes which are very sensitive to wide range of toxicant in the water [5].

Pesticides, insecticides and herbicides used in agricultural production have been recognised as having deleterious effects on aquatic organisms including fish and fish eggs [6]. Herbicides originating from agricultural activity enter the aquatic environment through atmospheric deposition, surface run-off or leaching and frequently accumulate in soft-bottom sediments and aquatic organisms [7, 8, 9]. Biochemical changes induced by herbicides strain cause disturbance in the metabolism and also cause inhibition of some important enzymes, retardation of growth and reduction in longevity of the organs. Persistence of these toxic chemicals in aquatic environment is dangerous for the survival of fish and their food organisms [10]. The alterations of

biochemical contents in different tissues of fish due to toxic impact of different agrochemicals and heavy metals have been reported by number of researchers [11, 12, 13].

Primextra, a pre-emergent broad spectrum herbicide for weed control in maize and sorghum farmlands, can be toxic if inhaled, swallowed or absorbed through the skin. It has been found that empty containers of this herbicide retains product residue for a long time and those applied in the weed control programmes accidentally leach into the aquatic environment either through runoff and/or as aerosol carried by wind [14].Therefore this study was aimed at investigating the sublethal effect of primextra on the tissue protein levels of *Clarias albopunctatus* under laboratory conditions.

MATERIALS AND METHODS

Ninety six (96) healthy juveniles of *Clarias albopunctatus* with mean body weight 24.10±2.10 and length 6±1.20 cm were obtained from Freedom Fish Farm in Nsukka, Enugu State, Nigeria. They were transported to the Fisheries and Hydrobiology Unit Laboratory, Department of Zoology and Environmental Biology, University of Nigeria, Nsukka in a plastic fish transport container with water. The fish were acclimatized for three weeks before the commencement of the study. During the period of acclimatization and the experiment, the fish was fed *ad libitum* on 45% crude protein diet. The fish were randomly divided into three replicate groups of eight fishes per replicate in plastic aquaria. The fish in groups A, B and C were treated with 0.6 µg/L, 1.0 µg/L, and 1.5 µg/L of primextra respectively. The fourth group was exposed to tap water (Temperature-23.60± 2.0°C, pH-7.3) as the control experiment. The sublethal primextra concentrations were prepared from the commercial preparation containing 290g of metalachlor and 370g of atrazine as the stock. The primextra and water were changed daily in a static renewal bioassay system. The fish were sacrificed and the tissues (liver, muscle and blood) were extracted and taken to Clinical Pathology Laboratory, College of Veterinary Medicine University of Nigeria for estimation of protein according to [15]. The data obtained were analyzed statistically with one-way analysis of variance (ANOVA).

RESULTS

The results showed that primextra caused significant decrease in the protein levels in *C. albopuntatus* exposed to primextra when compared with the control (Table 1-3). The liver protein levels (Table 1) decreased from 3.36±1.96 mg/g on the 7th day to 2.47±0.24 mg/g on day 21 in the group treated with 0.6µg/l primextra. While liver protein levels of fish exposed to 1.5µg/l primextra slightly increased from 2.27±0.77 mg/g on the 7th to 4.53±1.76mg/g on the last day. The percentage decrease in the liver protein levels (Table 1) was highest on day 7 with 54.66%, 58.97% and 69.37 % values in the fish treated with 0.6, 1.0 and 1.5 µg/l respectively. At the end of the study the percentage decrease were 66.21 %, 39.26% and 38.03% in the groups treated with 0.6, 1.0 and 1.5µg/l, respectively.

Table 1: Liver protein levels of *Clarias albopunctatus* exposed to sublethal concentrations of primextra for 21 days.

	Duration of exposure (days)		
Treatment group (µg/l)	7	14	21
Control(0.0)	7.41±1.56^{b}	6.38±1.33^{c}	7.31±2.55^{b}
Group A(0.6)	3.36±1.96^{a} (54.66)	2.20±0.44^{a} (65.52)	2.47±0.24^{a} (66.21)
Group B(1.0)	3.04±1.21^{a} (58.97)	4.35±1.03^{b} (31.82)	4.44±1.03^{ab} (39.26)
Group C(1.5)	2.27±0.77^{a} (69.37)	3.00±0.70^{ab} (52.98)	4.53±1.76^{ab} (38.03)

Results are expressed as mean of the samples ± standard deviation, mean values with different superscript are significantly different (P>0.05). Values in parenthesis indicate % decrease.

The changes in the muscle protein levels are shown in Table 2. The muscle protein levels decreased along days of exposure in the treatment groups when compared to the control. The percentage change (Table 2) in the muscle protein indicated that on the 7th day, the percentage decrease over the control were 48.75%, 74.85% and 72.84% in the fish exposed to 0.6, 1.0 and 1.5µg/l, respectively and 54.40%, 67.11% and 76.65% decrease over the control in the fish treated with 0.6, 1.0 and 1.5µg/l, respectively were recorded on day 21.

Table 2: Muscle protein levels of *Clarias albopunctatus* exposed to sublethal concentrations of primextra for 21 days.

	Duration of exposure (days)		
Treatment group (µg/l)	7	14	21
Control(0.0)	8.43±1.49^{b}	7.69±1.45^{b}	8.18±3.72^{b}
Group A(0.6)	4.32±3.75^{a} (48.75)	1.77±0.77^{a} (76.98)	3.73±1.97^{a} (54.40)
Group B(1.0)	2.12±0.87^{a} (74.85)	1.85±0.44^{a} (75.94)	2.69±0.96^{a} (67.11)
Group C(1.5)	2.29±1.00^{a} (72.84)	1.85±0.39^{a} (75.94)	1.91±0.14^{a} (76.65)

Results are expressed as mean of the samples ± standard deviation, mean values with different superscript are significantly different (P>0.05). Values in parenthesis indicate % decrease.

The changes in the blood protein levels are shown Table 3. The protein levels in the treatment groups remained generally lower than the control (P≤0.05). The percentage decrease (Table 3) on the 7th day was 23.35%, 31.48% and 23.65% in the fish exposed to 0.6, 1.0 and 1.5µg/l, respectively. At day 21 the percentage decrease in the blood protein was highest (66.70%) in the fish treated with 1.5µg/l and least (62.87%) in the group exposed to 1.0µg/l**.**

Table 3: Blood protein levels of *Clarias albopunctatus* exposed to sublethal concentrations of primextra for 21 days.

	Duration of exposure (days)		
Treatment group (µg/l)	7	14	21
Control(0.0)	7.02±1.61^{b}	7.61±1.29^{b}	8.86±0.70^{b}
Group A(0.6)	5.10±1.45^{a} (23.35)	4.86±3.21^{ab} (36.14)	3.05±1.57^{a} (65.58)
Group B(1.0)	4.81±1.21^{a} (31.48)	3.06±0.94^{a} (59.79)	3.29±1.19^{a} (62.87)
Group C(1.5)	5.36±1.01^{a} (23.65)	2.65±0.98^{a} (65.18)	2.95±0.86^{a} (66.70)

Results are expressed as mean of the samples ± standard deviation, mean values with different superscript are significantly different (P>0.05). Values in parenthesis indicate % decrease.

DISCUSSION

The present study revealed reduction in protein levels in the tissues (liver, muscle and blood) of *C. albopunctatus* following sublethal exposure to primextra. The decreased trend of the protein content as observed in the present study in the analysed fish tissues may be due to metabolic utilization of the ketoacids to gluconeogenesis pathway for the synthesis of glucose; or due to the directing of free amino acids for the synthesis of necessary proteins, or, for the maintenance of osmotic and ionic regulation and this indicates the physiological adaptability of the fish to compensate for pesticide stress. To overcome the stress the animals require high energy. This energy demand might have led to the stimulation of protein catabolism as stated by [16, 17, 18, 19, 20]. Similar change was observed in *C. punctatus* exposed to technical grade malathion by [21]. [22] reported reduction of protein content of liver, brain and ovary *of* C. *punctatus* exposed to fenvalerate.[23] reported decrease in protein content of dimethoate intoxicated fish *Clarias batrachus* indicating the physiological adaptability of these fish to compensate for pesticide stress. The findings of this study was consistent with the report of observed decreased protein content in freshwater fish, *Labeo rohita* exposed to sub lethal concentrations of pesticide mixture of monocrotophos and fenvalerate [24]. This study finding also coincides with that of [25] who reported decrease in protein content in the liver, muscle, kidney, intestine, brain and gill of *C. punctatus* treated with quinalphos. The changes and decrease in protein level might also be due to inhibition of metabolizing enzymes by administration of toxicants.

CONCLUSION

The present work indicates that primextra caused alterations in the protein metabolism of *Clarias albopunctatus*, Treated fish tissues showed more reduction in protein levels than the control and this may be due to more pesticidal stress. The low content of proteins reflects a change in the rate of synthesis and degradation of protein, lowered working capacity under the impact of accumulation of pollutants leading to an alteration in function indicating the vulnerability of the organ. Therefore, Protein levels in liver, muscle and blood of *Clarias albopunctatus* could serve as useful biomarkers of herbicide toxicity and could be of immense value in policy formulation regarding safe levels of the compound in the aquatic environment.

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