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Effect of cypermethrin on blood glucose and urea levels of *Heteropneustes fossilis* (Bloch)

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ABSTRACT

The present study was designed to assess the impact of cypermethrin, a type 2 synthetic pyrethroid insecticide on blood glucose and urea levels of *Heteropneustes fossilis*. Fishes were subjected to sub-lethal concentration (0.96 µg/L) of cypermethrin 10% emulsified concentration for 24 h, 48 h, 72 h and 96 h. There was a significant increase (p<0.05) in blood glucose and urea levels of cypermethrin exposed fishes compared with the control group. Therefore, proper care should be taken to minimize the contamination of freshwater bodies while spraying insecticides.

Key words: Cypermethrin, *Heteropneustes fossilis*, Glucose, Urea.

Introduction

Synthetic pyrethroids are insecticides that have been introduced over the past two decades for agricultural and domestic use (Sanchez-Fortun and Barahona, 2005). These chemicals are potentially more toxic to fish and other aquatic organisms and are list toxic to mammals. Owing to the excessive use of synthetic pyrethroids, the environment and water resources are being polluted, thus endangering aquatic life directly and human life indirectly (Hill, 1989). Due to their lipophilicity, pyrethroids have a high rate of gill absorption even when present at very low concentrations in the water. This in turn is a contributory factor to the sensitivity of the fish to aqueous pyrethroid exposures, because fish seem unable to metabolize the pyrethroids efficiently (Viran *et al.*, 2003). Cypermethrin is a type of cyanophenoxy-benzyl pyrethroid and is categorized as restricted use pesticide (RUP) by United States Environmental Protection Agency (USEPA), because of its high toxicity to fish (Extension Toxicology Network, 1996). It is used to control pests of cotton, fruits and vegetable crops (Pedigo, 1996). The excess use of this pesticide may enter into natural waters through agricultural run-off and ultimately cause damage to non-target organisms such as fish. (Prashanth and Neelagund, 2008; Singh *et al.*, 2010). The freshwater catfish *Heteropneustes fossilis* is widely cultivated in rice fields, swamps and derelict water bodies (Chondar, 1999) and is thus frequently exposed to agricultural runoff. Fish mortality may occur because of the use of cypermethrin in normal agricultural practice (Shires, 1983).

Blood is a pathophysiological reflector of the whole body, and therefore, blood parameters are important in diagnosing the structural and functional status of fish exposed to toxicants (Adhikari *et al.*, 2004). Changes in the biochemical blood profile indicate alterations in metabolism and biochemical processes of the organism, resulting from the effects of various pollutants and they make it possible to study the mechanisms of the effects of these pollutants (Luskova *et al.*, 2002). Changes in macromolecules like glycogen, protein and lipid are considered to be sensitive indicators of pesticides stress (Peter, 1973). Hence, the present study an attempt has been made to study the effects of cypermethrin on blood glucose and urea levels, selecting *H. fossilis* as an experimental model

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Material and Methods

Animals:

Healthy and sexually matured living fishes of length 17-20cm and weight 24-35g were procured from a local fish farm in Guwahati, Assam and disinfected in 0.1% solution of potassium permanganate for 5 minutes to avoid dermal infection. The fishes were allowed to acclimate in a glass aquarium in the laboratory for one month. The water of the aquarium was changed daily. Fishes were fed daily with commercial dry feed pellets (Tokyo pellets). The feeding was discontinued 24h prior to exposure. Commercial grade cypermethrin (10%EC) of liquid formations manufactured by United Phosphorus Ltd. was purchased from local agro-chemical stores.

Study design:

Twenty healthy acclimatized fishes were divided into two groups and were transferred to two separate aquaria (size 75 X 45 X 45 cm) of which one with 10 fishes served as control containing only tap water while the other with 10 fishes containing 0.96µg/L of Cypermethrin 10% EC referred as test group. The test water was changed every other day during the experimentation and proper oxygenation in the test solution was ensured. The experiments were conducted for 24h, 48h, 72h and 96h to study the short-term exposure effects and each treatment experiment was repeated for 7 times.

Biochemical analysis:

Blood samples were taken from the caudal vein of fish as described by Congleton and La Voie (2001). The blood was collected in anticoagulant-free centrifuge tubes. Serum was obtained by centrifugation of blood at 3000 rpm for 10min. Levels of glucose and urea in the serum samples were analyzed in semi automatic biochemical analyzer "Lablife ChemMaster" using Biosystem Kits.

Statistical anaylsis:

Student's t-test was used to analyze the statistical significance between the control and cypermethrin exposed fishes.

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Results and Discussion

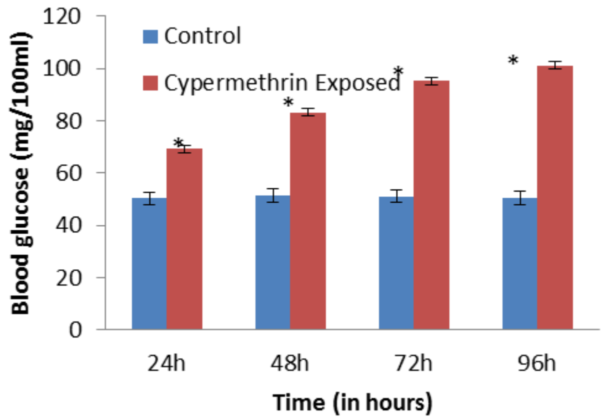
The blood glucose of cypermethrin exposed *H. fossilis* were significantly (p<0.05) increased from their corresponding control groups. In the control group the blood glucose was found within the range of 50.24 ± 2.46 mg/100ml to 51.37 ± 2.51mg/100ml while in the cypermethrin exposed group it was found to increase from 69.26 ± 2.41mg/100ml to 101.15± 2.03mg/100ml of blood with increase of the exposure periods (Table-1.; Figure-1).

Table-1. Blood glucose of control and cypermethrin exposed *Heteropneustes fossilis* expressed in mg/100ml of blood

Exposure Periods	Control	Cypermethrin exposed
24 h	50.24± 2.46	69.26 ± 2.41*
48h	51.37± 2.51	83.29 ± 1.36*
72h	51.10 ± 2.73	95.24 ± 2.49*
96h	50.45± 3.22	101.15 ± 2.03*

*Values are means ± SD of 7 observations. Significant differences are indicated by asterisks * (p<0.05).*

Figure-1. Blood glucose of control and cypermethrin exposed *Heteropneustes fossilis* expressed in mg / 100ml of blood



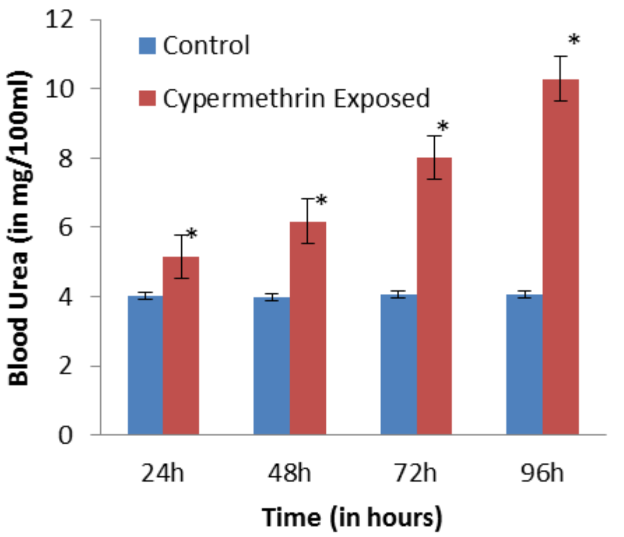
The blood urea of cypermethrin exposed *H. fossilis* were significantly (p<0.05) increased from their corresponding control groups. The blood urea in the control group of fishes was found within the range of 3.97 ± 0.11mg/100ml to 4.05 ± 0.10mg/100ml of blood whereas in the cypermethrin exposed group it was found to increase from 5.16 ± 0.68mg/100ml to 10.29 ± 0.89mg/100ml of blood with increase of exposure periods (Table-2.; Fig-2).

Table-2. Blood Urea of control and cypermethrin exposed *Heteropneustes fossilis* expressed in mg/100ml of blood

Exposure Periods	Control	Cypermethrin exposed
24 h	4.04± 0.31	5.16 ± 0.68*
48h	3.97± 0.11	6.18 ± 0.67*
72h	4.05 ± 0.12	8.01 ± 0.63*
96h	4.05± 0.10	10.29 ± 0.89*

*Values are means ± SD of 7 observations . Significant differences are indicated by asterisks * (p<0.05)*

Figure-2. Blood Urea of control and cypermethrin exposed *Heteropneustes fossilis* expressed in mg/100ml of blood



Discussion

Increase of blood glucose in the present study might be resulted from disruption in carbohydrate metabolism, possibly due to enhanced glucose 6-phosphatase activity in liver, elevated breakdown of liver glycogen or the synthesis of glucose from extra hepatic tissue protein and amino acids. Raja *et al.* (1992) suggested that the increase in blood glucose by pesticide treatment might indicate disrupted carbohydrate metabolism due to enhanced breakdown of liver glycogen, possibly mediated by increase in adrenocorticotrophic and glucagon hormones and / or reduced insulin activity. Similar observations were reported in *Labeo rohita* (Das and Mukharjee, 2003), *Sebastes schlegeli* (Jee *et al*., 2005) and *Oreochromis niloticus* (Firat *et al*., 2011) exposed to cypermethrin.

Hyperglycaemic response is an evidence of stress due to cypermethrin. Stress is an energy demanding process and the animal mobilizes energy substrates to cope with stress metabolically (Vijayan *et al*., 1997). Glucose is one of the most sensitive indices of the stress state of an organism; its high concentration in blood indicate that the fish is in stress and it is intensively using energy reserves i.e. glycogen in liver and muscles (Vosyliene, 1999; Sunil Kumar Guru, Rajesh Behera and Milan Kumar Behera, 2014). The stress hormone cortisol has been shown to increase glucose production in fish, by both gluconeogenesis and glycogenolysis and likely play an important role in the stress associated increase in plasma glucose concentration (Iwama *et al*., 1999). In the present study, glucose levels might elevate to cope with the increased energy demand during pesticide induced stress, which is an important pathway for the recovery from stress. Increased in the glucose levels were reported in *Prochidolus lineatus* (Martinez *et al*., 2004) and *Oreochromis niloticus* (Monteiro *et al*., 2005) in response to lead and copper respectively. Borges *et al*. (2007) suggested that cypermethrin induced hyperglycemia in *Rhamdia quelen* is likely to be a sign of stress. Similar result was reported by Ansari and Kumar (1988) in *Heteropneustes fossilis* exposed to cypermethrin. The findings of the present study are in close consortium with the above observations.

The significant increase of blood urea in cypermethrin exposed fishes for all exposure periods might be inadequate excretion due to kidney damage or excessive protein breakdown due to toxic stress. Goel *et al*. (1984) suggested that the increase in blood urea in *Clarias batrachus* exposed to alachrol toxicity was possibly due to the anomalies in kidney functioning. Similar observation was reported by Sharma (1989) and Himansu Bhusan Mahananda (2014) in *Clarias batrachus* induced to lithium. Jyothi and Narayan (2000) reported that the increase in blood urea in *Clarias batrachus* exposed to phorate could be due to reduction in the protein content. Jayantha *et al*. (1984) who recorded same observation in *Tilapia mossambica* exposed to phosphamidon suggested protein degradation or biochemical transformation of protein, nitrogen into other nitrogenous products.

Sivaramakrishna and Radhakrishnaih (1998) observed an increase in blood and liver urea level in *Cyprinus carpio* exposed to mercury. They ascribed it to part of excess ammonia converting into less toxic urea in the liver during active operation of urea-ornithine cycle. Similar results were described by Venkataramana *et al*. (2005) in fish *Glossogobius giuris* exposed to malathion and Kumar *et al*. (2012) in *Channa punctatus* exposed to lambda-cyhalothrin, REEVA-5. Increase in blood urea was also reported by Philip and Rajasree (1996) and David *et al*. (2004) in *Cyprinus carpio* and Kumar *et al*. (2011) in freshwater fishes exposed to cypermethrin and Balasubramaniam and Kumar (2013) in *Heteropneustes fossilis* induced to sodium arsenate.

Conclusion

From the present study it is obvious that exposure of fishes to cypermethrin produces degraded metabolic changes which make them less fit for survival. This in turn will affect the fecundity of the fish population and also other organisms including human beings through food chain.

Conflict of Interests:

The authors declare that there is no conflict of interests regarding the publication of this paper.

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