



**ORIGINAL ARTICLE**

**Haematological Changes in *Clarias batrachus* in Relation with Ram Ganga River, Bareilly**

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**ABSTRACT**

Insecticidal use in agriculture gained momentum around the mid twentieth century. Fungicides are also used in agriculture for the prevention of fungal infection in seed grain. Later these compounds discharge in nearby water bodies and consumed by fishes and other aquatic life. These fat soluble contaminants concentrate in the adipose tissue of fishes by bioaccumulation and bio-magnification. The fishes, best indicator of water body pollution, are the most sensitive of all the aquatic animals towards the pollutant. The accumulation of effluents becomes hazardous to the aquatic organism because they are the most important factors of food chain. The fish selected is commonly used in laboratory because it is hardy and easily available throughout year. Walking catfish is the most common English name for this species. This is valued as a food fish owing to its ability to survive extended period out of water. Walking catfish can be sold and treated live with ease, ensuring fairly fresh food product. For the above said purpose present study is undertaken to assess the haematological changes in fishes.

**Key words:** Haematological Changes, *Clarias batrachus*, Ganga River, Bareilly

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**INTRODUCTION**

Pesticides are used extensively in every field of our surroundings. To small extent they enter the body. Sources of pesticides in aquatic environment may be categorized in to natural agricultural activities, which results in the waste water discharge in to water and land. Some of these pesticides are biological, but others containing heavy metals like cadmium, lead and mercury are highly hazardous to aquatic biota and normally occur in low concentrations.

Since a major part of the world's food is supplied from fish source, so it is secure the health of fish (Medhi, *et al.*, 2013). Fish are known to be richest source of high quality protein. In India, where protein deficiencies in wreaking havoc on the health of millions of underprivileged and malnourished population, the fisheries can prove to be boon in overcoming this problem. However, fisheries suffer greatly threat from environmental pressures, in particular deteriorating water quality and habitat. Aquaculture is facing an increase threat from water pollution. In general, studies of pesticides can be important in two main aspects, public health point of view and aquatic environment view point.

Hematological tests are important diagnostic tools and valuable as indicator of diseases or stress due to pollutant and environmental fluctuation (Bhatkar, 2011). The blood also plays as integrated and inevitable part in immune system. Fish blood is being studied increasingly in toxicological research and environmental monitoring as a possible

indicator of physiological and pathological changes in fisheries management and disease investigation (Javed and Usmani, 2012). The blood parameters in fishes are influenced by many factors. Quality of water, temperature, food availability and physiological status of fish either directly or indirectly influenced on blood constituents of fish (Malathi, *et al.*, 2012). Like in warm blooded animals changes in blood parameters of fish which occurs because of injuries or infection of some tissue or organ, can be used to determine and confirm the dysfunction and injuries of the latter (organ and tissue). However in fish, these parameters are more related to response of whole organism i.e. to the effect of survival, reproduction and growth. On the basis of studies on the warm blooded animals, specific indices reflect the effect of some substances on the organism (e.g. changes in the blood serum cholinesterase under influence of insecticides and changes in hematology under the influence of nitrites) have been determined. However, the complex of unspecified biochemical indicators of blood reveals more fully the general effect of pesticides on the fish.

## MATERIALS AND METHODS

### SELECTION AND COLLECTION OF FISH:

*Clarias batrachus*, the walking catfish, was selected as a test animal. It has an elongate body that is broader at the head, tapering toward the tail. It is readily recognizable as a catfish with four pairs of barbells (whiskers) and fleshy papillated lips. The teeth are villiform (small and bristle-like). It is nick-named as walking catfish. It is commonly available in local water. It is hardy fish and can withstand the aquarium condition very well. Adult live specimens of fish *Clarius batrachus* with the size range of 16-18cm. were collected from Ram Ganga River. Fishes were carried immediately to department laboratory avoiding stress and injury as much as possible.

### Test compound:

Commonly used pesticide in experimental area (Dithane)

### ESTIMATION OF MEDIAN LETHAL CONCENTRATION:

Median Lethal concentration is the statistically derived dose that causes mortality in 50% of given population of an organism under a definite set of experimental conditions after 96 hours of exposure period. To investigate the effects of pesticide in the body of *Clarias batrachus* LC<sub>50</sub> values of the respective pesticide, the experiment designed accordingly. Control group was also maintained during the experiment. For determination of 96 hours LC<sub>50</sub> (lethal concentration values, a group of six fishes was used for each concentration of pesticide. All experiments were run for 96 hours (4 days) and the concentration of pesticide that caused the 50% mortality in fish the named the LC<sub>50</sub> value of dithane. Fish mortality was recorded at regular intervals of 24 hours and dead fishes were carefully and immediately removed.

### EXPERIMENTAL PROTOCOL AND PREPARATION FOR EXPERIMENT:

Experiments employing different doses of sublethal concentration of pesticide were carried out in 100litre capacity aquarium in the departmental laboratory. The fishes were collected from Ram Ganga River and water was also taken from the same site. The acclimatized fishes were divided into 2 groups. The first group served as control and other group served as experimental group. The fish were exposed to sublethal concentrations of pesticide for 96 hours. At the end of exposure time, the blood was collected from the test fish and studied for different haematological parameters viz., haemoglobin and total erythrocyte count. Double oxalate vials were used for storing the blood samples for various haematological studies. 800 mg potassium oxalate and 1200 mg ammonium oxalate was dissolved in 100 ml distilled water. One drop of this solution was added to each empty sterilized vial, shaken and dried in the oven at 80°C. Six fishes from each set control and treated were sacrificed for the haematological studies of blood

after 24 hrs, 48 hrs, 72 hrs and 96 hrs of exposure to dithane. The blood was collected after severing the caudal peduncle of the living fish by a scissors. The blood samples were taken in double oxalate vials. In each vial two to three drops of an anticoagulant EDTA were added prior to collection of blood. The blood samples were analyzed individually for each fish.

#### HAEMATOLOGICAL PARAMETERS:

Total erythrocytes were counted with the help of improved Standard Neubauer Haemocytometer (Dacie and Lewis, 1975). The haemoglobin concentration was measured with Sahli's method (Wintrobe *et al.*, 1981).

#### STATISTICAL CALCULATIONS:

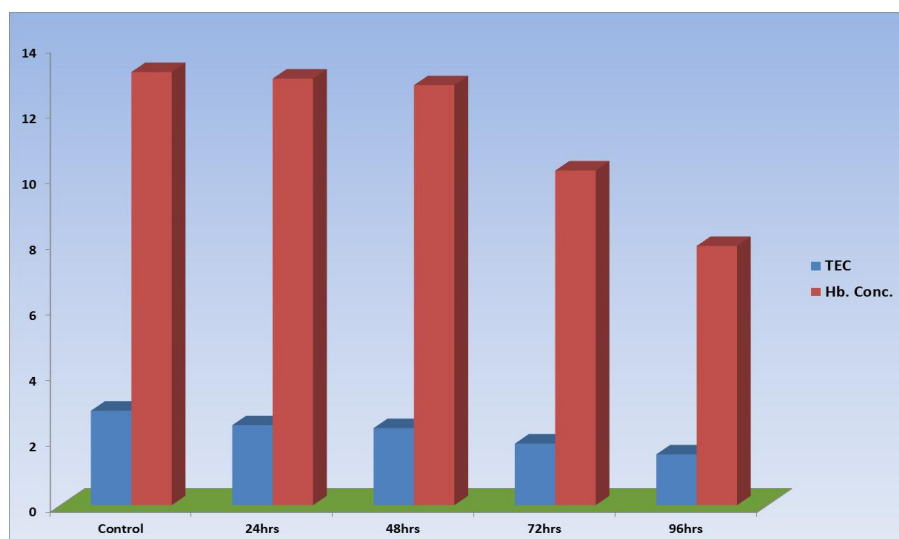
Statistical calculations were done with the help of computer software.

### RESULTS AND DISCUSSION

The results are shown in Table-1 and Fig. 1 which show decreasing trend in total erythrocyte count and haemoglobin concentration after pesticide treatment.

**Table 1:** Value of TEC and Hb. Conc. In *Clarias batrachus* in control and after pesticide treatment

| Set     | TEC ( $10^{12}/l$ )<br>(Mean Value signified at 5%, 1%) | Hb. Con. (g/dl)<br>(Mean Value signified at 5%, 1%) |
|---------|---------------------------------------------------------|-----------------------------------------------------|
| Control | 2.88                                                    | 13.2                                                |
| 24hrs   | 2.44 (P>0.05)                                           | 13.0 (P>0.05)                                       |
| 48hrs   | 2.35 (P<0.05)                                           | 12.8 (P<0.05)                                       |
| 72hrs   | 1.88 (P<0.01)                                           | 10.2 (P<0.05)                                       |
| 96hrs   | 1.55 (P<0.01)                                           | 7.9 (P<0.01)                                        |



**Fig 1:** Showing Value of TEC and Hb. Conc. In *Clarias batrachus* in control and after pesticide treatment

The total erythrocyte count showed decreasing trend on exposure to dithane at different time intervals (24 hrs, 48 hrs, 72 hrs and 96 hrs) at sublethal concentration. The findings are strongly supported by the studies of Agrawal, *et al.* (1994), Nath and Banerjee (1994); Jaipurkar and Nath (1996) who recorded the effect of different pesticides and heavy metals on *Heteropneustes fossilis* (Bloch.) reporting a decrease in the total erythrocyte count. Trivedi and Rajbanshi (1999) and Das and Mukherjee (2000) reported a decrease in TEC in *Labeo rohita* (Ham.) due to the exposure to zinc, copper and quinalphos

respectively, thus supporting the present findings. Svobodova, *et al.* (2003) also recorded a decrease in TEC in the blood of different fishes after treatment with different pesticides, Das, *et al.* (2004) studied the effect of nitrite and chromium respectively on *Labeo rohita* (Ham.) and recorded a decrease in TEC from the control set, thus supporting the present investigation. The significant decrease of RBC count may be due to the retarded erythropoiesis, which triggered as a typical stress response.

The hemoglobin concentration showed decreasing trend on exposure to dithane at different time intervals (24 hrs, 48 hrs, 72 hrs and 96 hrs) at sublethal concentration. The same result were observed by Rai and Qayyam (1984) in *Catla catla* due to intoxication of lead; in *Channa punctatus* exposed to BHC; Garg and Tyagi (1989) in *Heteropneustes fossilis* due to manganese poisoning, Goswami and Dutta (1991) in *Heteropneustes fossilis* due to vit. A deficient diet; while Singh and Shrivastava (1991) in *Heteropneustes fossilis* due to propoxur toxicity; Nath and Banerjee (1995) in *Heteropneustes fossilis* treated with devithion. The reduction in haemoglobin content may be due to the formation of a mucous envelope on the entire body surface and particularly on gills.

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