

SODIUM FLUORIDE TOXICITY IN FRESH WATER FISH CARASSIUS AURATUS (GOLD FISH): EFFECT ON RED BLOOD CELL COUNT

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Abstract: Fluoride is naturally occurring compound present in the earth's crust which enters the ground water through natural and anthropogenic sources. A wide range of environmental and genetic factors cause aquatic organisms to respond differently to given level of fluoride, but they do display characteristic fluoride intoxication signs. Fluoride tends to accumulate in the exoskeleton of invertebrates and in the bone tissues of fishes. The fluoride ions act as enzymatic poisons, inhibiting enzyme activity and ultimately interrupting metabolic processes such as glycolysis and synthesis of proteins. The present study consists of toxic effects of lethal concentration of sodium fluoride on the red blood cell count of the ornamental fish *Carassius auratus* (Gold fish) under laboratory conditions. Results indicate duration of exposure period and change in the Red blood cell count at regular period.

Keywords: *Carassius auratus*, sodium fluoride, RBC, RBC count, Chronic.

I. INTRODUCTION

Fluoride is present in the environment as a stable form of the super reactive element fluorine. Fluoride is the 17th most abundant element in the earth's crust, with fluoride detectable in all most all minerals. The main minerals are Fluorspar (CaF_2), Cryolite (NaAlF_6) and fluorapatite [$\text{Ca}_{10}\text{F}_2(\text{PO}_4)_6$]. Fluoride naturally enters the aquatic system through weathering of alkalic and silicic igneous and sedimentary rocks, primarily shale's as well as from emissions from volcanic activity. Fluoride is typically found in fresh water at concentrations less than 1mg/l, however, natural concentrations may exceed even 50mg/l.(McNeely et al 1979). The benefits of fluoride was seen mostly in the hardening of teeth and protection from caries (Barbier et al .,2010).Aquatic life is continuously exposed to toxic concentration of fluoride in surface waters and harmful effects ensure when fluoride enters food chain and accumulates in the exoskeleton of invertebrates and bone tissues of fishes. A review by Barbier et al. (2010) has outlined a number of cellular processes in which fluoride can have deleterious effects. Identified effects include disruption of enzyme activity (mostly inhibition), inhibition of protein secretion and synthesis, alternation of gene expression. Fluoride disrupts enzyme activity by binding to functional amino acid groups that surround the active centre of an enzyme. This includes the inhibition of enzymes of the glycolytic pathway and the kreb's cycle (Barbier et al 2010). Studies by Mendoza-Shulz et al (2009) indicate that fluoride at micro molar concentrations can act as an anabolic agent and promote cell proliferation, whereas at milli molar concentrations it acts as an enzyme inhibitor on e.g. phosphates, which play an important role in ATP production cycle and cellular respiration.

The present study describes the toxicity of sodium fluoride on the fresh water gold fish *Carassius auratus* in terms of Red blood cell count.

II. MATERIALS AND METHODS

The fish *Carassius auratus* weighing about 8 -10 gm were collected from fishery centre and studied in spacious aquarium. The water in aquarium was aerated continuously with constant oxygen supply device. The temperature of the aquarium was 30° C and the same was maintained at normal temperature throughout the course of investigation and they were

exposed to natural photo period. The fish were given one week time to get adapted to the laboratory conditions before executing further investigations. The range of variations in the size of fish selected was minimized by selecting those of uniform size. The animal was starved for 24 hours period prior to each estimation so as to eliminate possibility of differential factors, if any influencing the estimation. After acclimatization, the fishes were subsequently divided into two groups of one control and other experimental group, which was exposed to different lethal exposure periods like 1,7,15,20 and 30 days.

III. COLLECTION OF BLOOD SAMPLES AND ESTIMATION OF RBC COUNT

The blood was collected through a caudal incision and it is immediately diluted with Hayem's fluid (5g of sodium sulphate, 1g of sodium chloride, 0.5 g of mercuric chloride dissolved in 200ml of distilled water) The Hayem's fluid was taken up to 0 marks in RBC pipette and it was mixed thoroughly by rotating the pipette and the mixture was allowed to stand for about 2-3 min for thorough mixing. The counting chamber and cover glass were cleaned and the cover glass was placed over the portified area. Again the solution was mixed gently and the sample of solution was expelled and a drop of fluid was allowed to flow under cover slip, handling the pipette at an angle of 90. After a gap of 2-3 min, the RBC settled and portified area of the counting chamber was focused under the microscope and RBC was counted in five small squares of RBC columns. The RBC were counted in the 4 corner squares and the control square under high power microscope and the number of RBC per cubic millimeter were calculated using the following formula

$$\text{RBC count} = \{(\text{Number of cells} \times \text{Dilution factor (200)} \times \text{Depth factor}) / \text{Area counted}\}$$

IV. STATISTICAL ANALYSIS

All the results obtained in this investigation were subjected to statistical analysis. For this, the data were fed to the computer, the standard deviation to each mean and percent change over the means of controls and experimental were derived. The significance of the data among controls and experimental were derived at 5% level using the DMR test, and is represented in the respective tables.

V. RESULTS

The data for the red blood cell number (RBC) in millions / cubic mm at different lethal exposure periods like 1,7,15,20 and 30 days including the control in fresh water fish *Carassius auratus* is presented in Table and Fig. The RBC count decreases at lethal concentrations of sodium fluoride. There is an increase in RBC from its earlier maximum decrease at 15 day and reached nearer to the control value at 30 day exposure period with a fairly good amount of recovery indicating the adaptive capacity of *Carassius auratus* during lethal exposure of sodium fluoride.

Table: RBC count (millions/mm³) the sodium fluoride treated individuals of *Carassius auratus* at different lethal exposure periods. The mean and standard deviation are taken from 6 individual measurements. The percent change in RBC count at different exposure periods is calculated in relation to the level in the control medium which is fixed at 100%

RBC Count	Control	Lethal exposure periods				
		24 h	7 day	15 day	20 day	30 day
Mean	3.96	5.95	3.59	2.41	3.45	3.72
S.D. ±	0.416	0.821	0.525	0.471	0.396	0.451
(% Change)	-	(+50.25)	(-9.34)	(-39.14)	(-12.87)	(-6.06)
% Recovery	-	-	-	-	-	93.93
t-Test	-	P<0.001	P<0.05	P<0.01	P<0.01	P<0.05

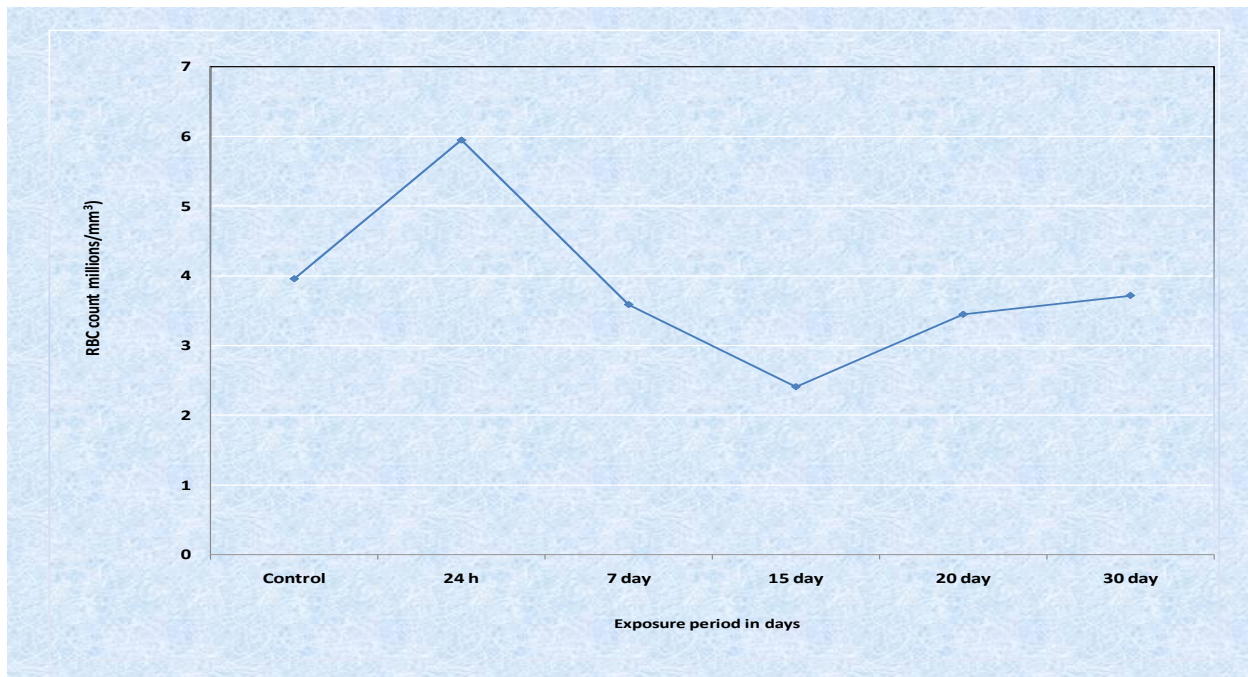


Fig: RBC count in *Carassius auratus* (Gold Fish) in control and at different lethal exposure periods of Sodium fluoride

VI. DISCUSSION

In the present study at 24 hr exposure, the RBC registered a steep rise when compared to the control medium. But in the middle of the sodium fluoride exposure period i.e. at 15 day, the RBC showed a decline in its number. This indicates that the presence of sodium fluoride in the medium of the present study might have induced hypoxia which in turn accelerates the haemopoietic tissue. Changes in the erythrocytic profile suggest a compensation of oxygen deficit in the body due to gill damage and the nature of the changes shows a release of erythrocytes from the blood depots (Drastichova et al 2004). The other reason for RBC suppression might be due to the failure of detoxification mechanisms, initially in the early half of the 30 day exposure where the fish probably experience greater stress.

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