Histological Alterations in the Tissues of *Catla Catla* Exposed to Lethal Concentrations of Copper as Copper Sulphate

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ABSTRACT

The study deals with the acute toxicity of copper as copper sulphate, as a component of industrial waste and its effect. Copper is an essential trace metal for life, present in natural waters and sediments and virtually in other media including air, water and soil. It is abundant in industry effluents from manufacturing of electronic goods, fertilizers, fungicides and metal plating by products. This trace metal is a vital part of many biological enzymes as a co-substrate. However, higher concentration of copper in an aquatic ecosystem would become toxic to organism due to increased production of free radicals in the body. Even though copper is an essential element in low concentration, if it is discharged into the water body it cause deleterious effects to fauna, especially fish. Fish are the simple and reliable biomarkers of copper pollution of aquatic bodies. The fresh water fish Catla catla were subjected for acute toxicity studies. The present study deals with the acute toxic assay of fresh water fish Catla catla exposed to copper sulphate and histological analysis. The lethal concentration (LC_{50}) of copper as copper sulphate for 24, 48, 72 and 96hrs were found to be 32, 28, 23 and 20ppm respectively. The histological alterations in intestine and muscle were studied after the exposure of fish to copper sulphate and results revealed extensive histological alterations in the internal structure of intestine and muscle.

Keywords: Catla catla, Copper sulphate, Lethal Concentration (LC₅₀), Histology.

1. INTRODUCTION

Heavy metals like copper, lead, barium etc are one of the major components of the industrial wastes, which along with other products from industrial operations are discharged into the aquatic environment and it is toxic to aquatic life [1]. Contamination by heavy metal in aquatic system is a severe problem and fish are more repeatedly exposed to these pollutants and might be taken in through gills, skin and contaminated foods [2,3]. Aquatic organisms are often exposed to mixtures of toxicants because it is believed that regardless of where the pollution occurs, it will eventually

end up in the aquatic environment [4]. Metal pollution may damage aquatic organisms at the cellular level and possibly affect the ecological balance. The problem of metal pollution is considered to be the most serious one that faces mankind in the twenty-one century. The trace element Copper is a vital part of many biological enzymes as a co-substrate. Even though copper is an essential element, higher concentration causes deleterious effects to flora and fauna especially fish which is the major source of protein for the community. Contaminated fish through the food chain also affects the human being. Metal pollution may damage organisms at the cellular level and possibly affect the ecological balance Fish are the simple and reliable biomarkers to assay the level of pollution [5]. Hence, the present study was focused to assay the acute toxicity and its effect on the histology of intestinal and muscle tissue of *Catla catla* treated with copper as copper sulphate, as a component of industrial waste.

2. MATERIAL AND METHODS

COLLECTION AND MAINTENANCE OF FISH

Live fingerlings of *Catla catla* were collected from Bharat fish farm, Poondy, Thiruvallur, Tamil Nadu, India and brought to laboratory in plastic bags to avoid any injury and disinfected by giving a bath for 2mins in 0.05% KMnO₄ solution. Thereafter, they were transferred to tubs for two weeks, for acclimatization in the laboratory conditions. Fish were fed with live *Tubifex* worm. Dead fish were removed whenever located. Water was changed at regular interval. Physico-chemical parameters of the tap water were analyzed to confirm the permissible limits.

ACUTE TOXICITY

After two weeks of acclimatization fingerlings were starved for 24hr prior to exposing them to copper sulphate. The stock solution was prepared by dissolving copper sulphate in distilled water. The LC_{50} values were found by Finney Probit analysis [6] and the values were tabulated in Table 1.

HISTOLOGY

Muscle and intestinal tissues of both control and treated with copper sulphate *Catla catla* fingerlings for 96hr were subjected for histology study. Histological analysis was performed according to Culling [7] with few modifications.

3. RESULT

ACUTE TOXICITY

The lethal concentration (LC₅₀) of copper as copper sulphate for 24, 48, 72 and 96hrs were 32, 28, 23 and 20ppm respectively and its values are tabulated in table 1. For 96hr lethal concentration, the upper and lower limits are 50.56 and 20.25 respectively.

HISTOLOGICAL ALTERATIONS

In control, intestine of *Catla catla* showed a normal structure that persisted through the 96hr of the experimental period and the four layer of intestine were clearly observed in control group (Plate.1a). Intestine of the treated fish showed several histological alterations (Plate.1b) such as disjoinment of layers, shortened villi and mild cattarhal enteritis, also wide gap between muscular and vacuolization were seen.

Muscle of control group of *Catla catla* showed a normal structure that persisted through the 96hr of the experimental period (Plate.2a). In treated muscle edema and mild lymphocyte infiltration, vacuolar degeneration in muscle bundles and atrophy of muscle bundles were observed. Edema between muscle bundles and splitting of muscle fibers were seen (Plate.2b).

4. **DISCUSSION**

The quality of the water was checked before using it for the acclimatization of fish in the laboratory conditions. The lethal concentration of the copper sulphate was studied by exposing the fresh water fish *Catla catla* for various concentrations and different time intervals. As the time of exposure increases there is a decrease in LC_{50} value which is inferred from the Table 1. The 96 hr LC_{50} value of copper sulphate in *Catla catla* was found as 20ppm in the present work. Copper enters the fish bodies in three ways, through body surface, gill and the digestive tract. Copper in an aquatic ecosystem would become toxic to organism due to increased production of free radicals in the body [8].

Histological studies are recommended for the evaluation of fish health. Various alterations were observed in the muscle and intestinal tissues of *Catla catla* treated with lethal concentration of copper sulphate for 96 hours might be due to the toxic stress developed by copper. Sajda *et al.*, [9] also observed disjoinment and edema of *Clarias batrachus* treated with rogorin. Jeshesha *et al.*, [10] also observed edema and mild lymphocyte infiltration, vacuolar degeneration in muscle bundles, atrophy of muscle bundles and edema between muscle bundles and splitting of muscle fibers of *Catla catla* when treated with naphthalene.

5. CONCLUSION

Industrialization in the developing and developed countries have resulted in heavy metal contamination in to the ecological system. The aquatic flora and fauna are affected by the toxic substances which eventually enter into water body. The biological response of an aquatic organism to pollutants frequently induces changes at cellular and biochemical levels, leading to changes in the structure and function of the cells, tissues, physiology and behavior of the organism.

Exposure Time (hr)	LC ₁₆ (ppm)	LC ₅₀ (ppm)	LC ₈₄ (ppm)	Regression	Slope	Confidence Limit	
						Upper Limit	Lower Limit
24	17	32	41	Y=15.97-1.77X	1.58	50.56	20.25
48	15	28	35	Y=24.61-2.74X	1.55	43.40	18.06
72	12	23	30	Y=23.28-3.14X	1.60	48.30	18.63
96	10	20	25	Y=37.95-4.74X	1.62	32.50	12.30

Table 1: Acute toxicity (LC₅₀) showing tolerance of fish Catla catla.





Fig 1a Photomorphograph of the muscle section of control fish Catla catla showing normal structure of muscle

Fig 1bPhotomorphograph of the muscle section of *Catla catla* fish treated with copper sulphate showing edema and mild lymphocyte infiltration





Fig 2a Photomorphograph of the intestine section of control fish *Catla catla*showing normal structure of intestine

Fig 2b Photomorphograph of the intestine section of fish *Catlacatla* treated with copper sulphate showed mononuclear cell infiltration in the lamina propriva

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