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Research Article

ASSESSMENT OF TOXICITY OF COPPER AND MERCURY ON GOURAMI FISH, *TRICHOGASTER TRICHOPTERUS* (PALLUS)

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ABSTRACT

Aquatic pollution by heavy metals is a growing concern that has been proved because of its detrimental effects on biological life, extremely toxic to fish and some aquatic arthropods, including human beings. The present study was undertaken in order to find out the toxicological effects of copper and mercury on the *Trichopodus trichopterus* from the habitats characterized by different environmental condition. The investigation has centred mainly to study the toxic effects of copper and mercury individually at different lethal levels on the gourami fish (*Trichogaster trichopterus*). The heavy metals are some of the most toxic compounds to aquatic organisms, and of these, mercury and copper are among the most toxic. Gourami fish exposed to CuS04, ZnSo4 and HgCl2 have shown more or less identical response. Various concentrations, 0.3, 0.6 and 0.9 of the toxicants were used to determine the taxological effect on the fish. During the present investigations, copper was found to be most toxic among copper, zinc & mercury to all the tested Gourami fish. Further study is recommended studying the processes by which these heavy metals affect physiology and histology of fish and their accumulation in fish tissues.

Keywords: Toxicity, Copper and Mercury, *Trichogaster trichopterus.*

INTRODUCTION

The growing interest in aquarium fishes has resulted in steady increase in aquarium fish trade globally. In this situation, ornamental fish farming may be one of the most promising alternatives (Ghosh *et al.*, 2003). The top exporting country is Singapore followed by Hongkong, Malaysia, Thailand, Philiphines, SriLanka, Taiwan, Indonesia and India. The largest importers of ornamental fish are the USA, Europe and Japan. The emerging markets are China and South Afiica. Over US$ 500 million worth of ornamental fish are imported to USA each year. India's share in ornamental fish trade is estimated as Rs. 158.23 Lakhs which is only 0.008% of the global trade (New, 1999). The major part of the export trade is based on wild collection. There are very good domestic markets too, which are mainly based on domestically bred exotic species. The overall global domestic trade crosses 10 crores and is growing at the rate of 20 percent annually. The earning potential of this sector has hardly been understood and the same is not being exploited in a technology driven manner. Although no reliable data are available on the volume of the ornamental fish trade, a conservative estimate suggests that at least 200 million ornamental fishes are being traded globally (Sales & Janssens, 2003). In countries such as Germany, Heavy metals are the natural components of the Earth's crust which cannot be destroyed. As trace elements some of these heavy metals are required for the maintenance of metabolism of the body. However, with the increasing concentration these heavy metals become lethal and can cause poisoning. Heavy metals are dangerous since they are involved in bioaccumulation (Rand, 1995). Heavy metals can enter a water supply through various sources such as industrial waste, or even through acid rains. Copper is an essential trace element but its high concentration causes damage to kidney and liver. Mercury is a toxicant which plays no important role in the human physiology and biochemistry (Langston, 1990). The major sources of mercury are the volcanic eruptions, mining practices, etc. Release of such heavy metals into the water bodies, results in serious contamination of the

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streams, lakes, rivers, etc. Choosing Gourami fish (*Trichogaster trichopterus*) as a laboratory model is because of its unique feature to easily adjust to laboratory conditions and also since it is highly tolerant to environmental changes.

MATERIALS AND METHODS

Collection of fish

The ornamental gourami fish (*Trichogaster trichopterus*) having average length 15±1 cm and weight about 40±5 gm was procured from the Local fish vender in Mumbai and collected in the clean containers of 10 liters capacity, ensuring that they were not harmed either physically during collection and transportation. They were brought to the laboratory and transferred to aerated aquarium for acclimatization. The fishes were fed daily with commercial fish feed.

Laboratory Procedure

The adult ornamental gourami fish (*Trichogaster trichopterus*) with body weight of 3.2 g ± 2.12 g was purchased from local fish dealer to serve as laboratory models. They were placed in an aquarium and were allowed to acclimatize in the laboratory for a period of 7 days.

Toxicants

The toxicants selected for the experiment were copper and mercury.

Copper

Analar grade of copper sulphate (M.W. 249.68) was the source of copper. The salt was dissolved in distilled water and added to achieve the required concentration.

Mercury

Standard solution of mercury was freshly prepared using analar grade of mercuric chloride (Molecular Weight. 271.50) in distilled water and added to achieve the required concentration.

Toxicant concentration

Various concentrations (such as 0.3, 0.6 and 0.9) of the toxicants were measured in mg/L in terms of the individual heavy metal (copper and mercury). The experiment was conducted in 5L container with a volume of 3L with different concentrations of toxicants, where in each container contained 2L of water followed by the fish distribution and then adding the remaining water so as to make the total volume unto 3L. All containers were provided with artificial aeration and constant stocking density of 1 fish per container. One tank was kept as blank without any toxicant. Triplicates were run for each toxicant concentration. Readings were taken every 12hr to observe mortality. Dead fishes if any were removed and were fixed in formalin. The dead fishes were later dissected so as to remove their gills, liver and kidney prepared for histopathological analysis. Sections of 5-6µm thickness were cut and stained with haematoxylin-eosin and then used for investigation and analysis.

Acute toxicity test

The acute toxicity test was performed according to semi-static methods described in the OECD procedure. The fishes were not fed 24 h before the experiments and during the acute toxicity test. The experiments consisted of a control group and five experimental groups. Acute test was performed to determine the appropriate toxicity range. Assuming acute toxicity, 10 fishes per group were exposed to different heavy metals concentrations (0.0, 0.3, 0.6 and 0.9mg/L paraquat, purity 30%) in 85L aquarium. During the 96 h acute toxicity experiment, water in each aquarium was aerated and had the same conditions as the acclimation period. Test solutions were renewed every 24 h to maintain the chemical and the water quality. Every 24 h the dead fishes were removed and the numbers of survivals were recorded. The experimental was repeated in triplicate. LC50 values were calculated by the Probit Analysis test (Aydın & Köprücü, 2005).

RESULTS AND DISCUSSION

The results of the present study indicate that both heavy metals cupper and mercury varied in their acute toxicity to blue gourami, *Trichogaster trichopterus*. The toxicity of cupper and mercury on blue gourami increased with increasing concentration and exposure time. No fish died during the acclimation period before exposure no control fish died during acute toxicity tests. The mortality of blue gouramies for cupper and mercury doses 0, 0.3, 0.6 and 0.9 (mg/l) were examined during the exposure times at 24, 48, 72 and 96 h (Table 1 and 2). The mortality of blue gouamies was increased significantly with increasing concentrations from 0.9mg/l to higher concentrations for Cupper and 0.9mg/l to higher concentrations for mercury. Because mortality (or survival) data are collected for each exposure concentration in a toxicity test at various exposure durations (24, 48, 72, or 96 hours), data can be plotted in other ways; the straight line of best fit is then drawn through the points. These are time mortality lines. The LT50 (median lethal survival time) can be estimated for each concentration (Figure 1 and Figure 2).

Trichogaster trichopterus exposed to $CuS0_4$, and $HgCl_2$ have shown more or less identical response. During the present experiment, the *Trichogaster trichopterus* opened their operculum for a short duration in order to avoid high concentrations of heavy metals therefore the contact with the dissolved metal solution was also for a short duration. Hence very high concentration of metal is required for their death within 24 hrs.

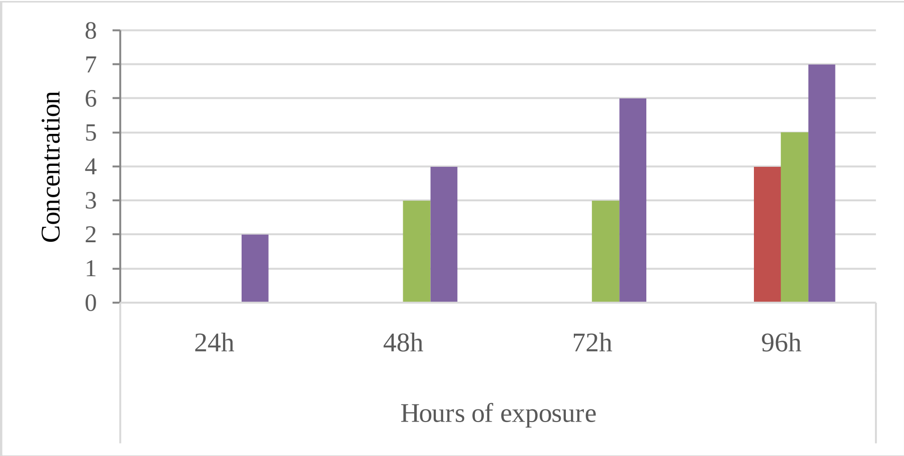

Figure 1. Acute toxicity testing statistical endpoints in blue gourami Fish exposed to Cupper in different times (24h, 48h, 72 h and 96 h respectively).

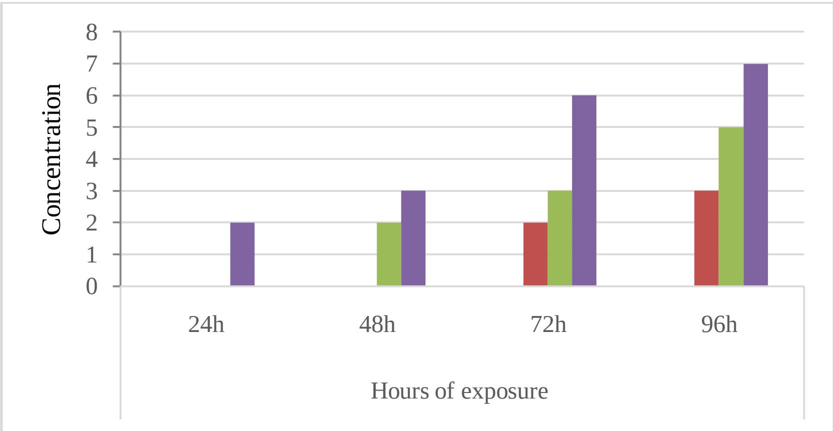

Figure 2. Acute toxicity testing statistical endpoints in blue gourami Fish exposed to Merquery in different times (24h, 48h, 72 h and 96 h respectively).

Table 1. Cumulative mortality of blue gourami Fish (n=7, each concentration) exposed to acute $CuSO_4$.

Concentration mg/L	Np. of mortality			
	24h	48h	72h	96h
0.0	0	0	0	0
0.3	0	0	0	4
0.6	0	3	3	5
0.9	2	4	6	7

Table 2. Cumulative mortality of blue gourami Fish (n=7, each concentration) exposed to acute $HgCl_2$.

Concentration mg/L	Np. of mortality			
	24h	48h	72h	96h
0.0	0	0	0	0
0.3	0	0	2	3
0.6	0	2	3	5
0.9	2	3	6	7

Table 3. Lethal Concentrations (LC1-50) of Cupper (Mean ± Standard Error) depending on time (24 - 96h) for blue gourami in (mg/l).

Method	24hrs	48hrs	72hrs	96hrs
LC_1	2.89 ± 0.10	2.02± 0.04	1.74± 0.07	1.11± 0.05
LC_{10}	2.95 ± 0.10	2.45± 0.04	2.37± 0.07	2.26± 0.05
LC_{20}	3.18 ± 0.10	3.11± 0.04	3.02± 0.07	2.84± 0.05
LC_{30}	3.56 ± 0.10	3.44± 0.04	2.88± 0.07	2.72± 0.05
LC_{40}	4.91 ± 0.10	4.56± 0.04	3.64± 0.07	2.59± 0.05
LC_{50}	5.23 ± 0.10	4.71± 0.04	3.84± 0.07	2.79± 0.05

Table 4. Lethal Concentrations (LC1-50) of Mercury (mean ± Standard Error) depending on time (24 - 96h) for blue gourami in (mg/l).

Method	24hrs	48hrs	72hrs	96hrs
LC_1	14.67± 1.15	12.47± 1.10	11.12± 1.09	10.38± 1.07
LC_{10}	16.23± 1.15	15.62± 1.10	13.72± 1.09	10.42± 1.07
LC_{20}	19.43± 1.15	17.57± 1.10	14.61± 1.09	11.43± 1.07
LC_{30}	20.57± 1.15	18.33± 1.10	16.83± 1.09	15.31± 1.07
LC_{40}	21.61± 1.15	18.93± 1.10	16.62± 1.09	13.72± 1.07
LC_{50}	22.42± 1.15	20.83± 1.10	17.14± 1.09	12.34± 1.07

Therefore, the difference was found in the heavy metals for longer durationLC_{50} (mg/l) values at 24 & 96 hr (Table in turn bringing mortality at 96 hr.1), (Table 3 and 4). However, at low concentrations the Another visible sign of their reaction to animals showed free movements, thus, the stimulus by metallic pollution was bodies of these animals remained in contact higher rate of mucus secretion. This is probably for protection from heavy metals as the sticky mucus forms a protective layer on the margins of operculum. During the present investigations copper was found to be most toxic among mercury to all the tested fish. Whereas, (Kulkarni *et al*., 2004; Waldichuk, 1974) have found mercury to be most toxic than other heavy metals to the crabs, prawns & clams. Fernandes has detected average copper and zinc in the considerable quantities (69-98 mm/g) in *N.oryzarum* and *P. sulcatus* from Mumbai coast. The level of heavy metals detected in the animals during the experiment might be the combined effect of experimental concentration and high concentration of these metals already accumulated in the body of animals from surrounding environment; as high level of copper and zinc has been reported in sediments of Bombay coast (Zingde, 1985). Similarly, accumulation of greater quantity of Zincthan Mercury in the animals from surrounding may be the explanation Zinc being more toxic than Mercury. Such type of variations in relative toxicity of heavy metals for different test organisms have also been reported (Kulkarni *et al*., 2004; Portmann, 1972). Waldichuk, (1974) reported toxicity in the order of copper>zinc> cadmium to sensitive larval stages of some marine molluscs and crustaceans, which strengthens present findings.

CONCLUSION

The occurrence of heavy metals in high concentrations in water and their toxicity to aquatic organisms especially fish species is observed in the study. Contamination of aquatic environment with heavy metals via rainfall runoff is very possible. Both heavy metals cupper and mercury varied in their acute toxicity to blue gourami, *Trichogaster trichopterus*. The toxicity of cupper and mercury on blue gourami increased with increasing concentration and exposure time.

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