Toxic effect of lead on the fingerlings of *Cirrhinus mrigala* (Hamilton, 1822)

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Abstract
Pollution of water bodies caused by heavy metals such as lead is a major problem affecting aquatic fauna. The effect of lead on the fingerlings of fishes has not been widely investigated. The present study was undertaken to evaluate the impact of heavy metal lead on the fingerlings of the freshwater fish, *Cirrhinus mrigala*. Different concentrations of lead nitrate solutions, viz., 1 ppm, 10 ppm, 40 ppm, 100 ppm, 120 ppm, 160 ppm, 200 ppm, 240 ppm, 300 ppm, 400 ppm and 500 ppm were prepared. Ten fingerlings were introduced into each concentration. The mortality rate and the time of survival of fingerlings in the lead nitrate solution during next 96 hours were recorded. The mortality rate increased to a sudden plateau from 100 ppm to 200 ppm (from 10% to 100%) and total mortality was attained at 200 ppm. LC₅₀ for the fingerlings was found to be 160 ppm. There was almost instantaneous mortality – absolute intolerance – of the fingerlings at 500 ppm. Our study results show that the fingerlings of *Cirrhinus mrigala* can survive the toxic effect of lead nitrate up to 40 ppm, without physiological stress.

Keywords: *Cirrhinus mrigala*; lead nitrate; mortality; LC₅₀; toxicity

1 | INTRODUCTION

Rapid industrialization, increase in population density, and changes in agricultural practices in the past few decades have contributed significantly to the contamination of water bodies with hazardous pollutants (Chavan and Muley 2014). Effluents released from industrial wastes contain heavy metals such as lead, chromium, mercury and arsenic, which pollute the water bodies (Korai et al. 2008). Heavy metal pollution is a major concern as they are resistant to degeneration, and cause morphological as well as physiological damage to the aquatic fauna (Shahriari 2005; Monteiro et al. 2005). Lead enter the aquatic habitat from effluents associated with industries like mining, smelting, refining, battery manufacturing and so on. Moreover, a wide range of chemicals are being used in aquaculture that sometime contains heavy metals (Mohsin et al. 2012a). Bioaccumulation of lead in fishes poses a severe problem because fish occupies an important position in the aquatic food chain and is routinely consumed by humans worldwide (Pauly et al. 1998).

Toxicity is dependent on factors such as species, age, diet etc. and environmental factors such as temperature, pH, dissolved oxygen, salinity etc. *Cirrhinus mrigala* is one among the three Indian major carps, cultivated widely in...
Asian countries (e.g. Mohsin et al. 2009, 2012b; Asaduj-jaman and Hossain 2006; Talukder et al. 2017). The easy availability and simple rearing techniques have made this fish a favourite model for ecotoxicity studies among researchers. Early investigators have shown that treatment with lead on the fish *Arrhina mrigla* caused changes to the haematological parameters of the fish (Iqbal et al. 1997), whereas it caused changes to the gill lamellae in *Channa punctatus* (Jha and Sathyendra Pandey 1990). However, the above-mentioned studies have been conducted by using adult fishes, and similar studies on fingerlings are lacking. It may be important to study the effect of heavy metals on the fingerlings of fishes as they are highly sensitive to subtle changes in the aquatic environment. The present study was undertaken to evaluate the impact of the heavy metal, lead, on the fingerlings of the freshwater fish, *C. mrigala*.

### 2 | METHODOLOGY

The fingerlings of *C. mrigala* were procured from Malampuzha Fish Hatchery in Kerala (mean ± SD SL = 2.4 ± 0.7 cm) and were allowed to acclimatize to the laboratory conditions for one week, before using them for experiments. Plastic troughs, of 6 L capacity, filled with non-chlorinated water, were used to house the fingerlings. The troughs were placed in the lab at room temperature (28°C), and the animals were maintained at a 12-hour light and 12-hour dark cycle. The animals were fed commercial fish feed (protein content: 40%, carbohydrate content: 23%, oil content: 10%, and vitamin and mineral content: 1%) at the rate of 5–6% of biomass, in two rations, during the experimental period.

Physico-chemical analysis of the water was done at the beginning of the experiment (APHA 1989), and mean of two consecutive measurements were considered. Temperature of the water was measured using a glass thermometer, and pH using Systronics pH meter (model: 335). Hardness of water was determined by titrating with EDTA, free CO₂ was determined by titrimetric method using phenolphthalein as indicator, and dissolved oxygen (DO) was measured by Azide modification method.

Lead nitrate, the heavy metal salt (Qualigens), was used as the toxicant in the experiment. Different concentrations of lead nitrate solutions, viz., 1 ppm, 10 ppm, 40 ppm, 100 ppm, 120 ppm, 160 ppm, 200 ppm, 240 ppm, 300 ppm, 400 ppm and 500 ppm were prepared with 5 L of non-chlorinated water. The lead nitrate solutions were prepared in 6L plastic troughs. One trough, without lead nitrate, was maintained as control. Ten fingerlings were introduced into each concentration. The period of experiment was fixed for 96 hours. The mortality rate and the time of survival of fingerlings in the lead nitrate solution during the period of experiment were observed and noted by manually entering the data in a log book.

Percentage mortality was plotted against concentrations, and Microsoft Excel, version 1997, was used for data analysis.

### 3 | RESULTS AND DISCUSSION

Water temperature, pH, total hardness, free CO₂ and DO from 27.3–28 °C, 7–7.5, 52–61 mg L⁻¹, 3.9–4.1 ppm and 4.9–5.3 ppm respectively. It was observed that the fingerlings exposed to 10 ppm and 40 ppm survived indefinitely and did not demonstrate any mortality at all. The mortality rate increased to a sudden plateau from 100 ppm to 200 ppm (from 10% to 100%) and total mortality was attained at 200 ppm (Figure 1). LC₅₀ for the fingerlings was found to be 160 ppm. There was almost instantaneous mortality – absolute intolerance – of the fingerlings at 500 ppm. When concentrations were plotted against time for 100% mortality, it was found that the time for mortality descends from 24 hours to 4 hours between 200 ppm and 300 ppm.

![FIGURE 1 Plot of 24-hour percentage mortality against concentrations](image-url)
lead nitrate became sluggish, eventually lost equilibrium, and death followed.

Our study results show that fingerlings of *C. mrigala* can survive the toxic effect of lead nitrate up to 40 ppm, without physiological stress. Above 40 ppm, the physiological stress sets in, as indicated by increasing surfacing of fishes. In addition, gills show high degree of dysfunction with increasing concentration of the toxicant. Death seemed to be caused by asphyxiation due to the reaction of heavy metal ions on the mucous secreted by the gills. It has been shown earlier that the primary effect of lead nitrate is the prevention of the diffusion of oxygen into the gill capillaries, leading to anoxia (Chavan and Muley 2014). Thus apart from the interference of lead ions with the body homoeostasis, physical obstruction to oxygen diffusion also occurs. Studies by a previous research group showed that *C. mrigala* showed restlessness, increase in rhythm and depth of respiration, when exposed to 0.1 to 0.4 mg/L of lead (Metelev et al. 1983), which agrees with our findings. Though lead does not cause mortality at sub-lethal concentrations, prolonged exposure can lead to undesirable effects like shorter life span, bioaccumulation etc. (Brown 1976; Lavie and Nevo 1987).

Heavy metals have been reported to accumulate in intestine, liver, kidney, muscles etc. (Senthilkumaar et al. 2012) other than gills. Lead has high permeability and high affinity for SH-group of various enzymes. The disruptions in membrane permeability caused by lead would lead to dysfunction of not only gills, but also, liver, brain, kidney, muscles etc. A study on *C. mrigala* has shown that the haematological parameters of the fish are affected due to nickel exposure (Parthipan and Muniyan 2013). Though the present study did not investigate the effect of lead on other organs, severe degenerative changes to gills were observed.

Thus, the present study indicates that concentrations of lead nitrate up to 40 ppm form the sub-lethal concentrations for the fingerlings of *C. mrigala*.

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Toxic effect of lead on *Cirrhinus mrigala* fingerlings
J Fish 6(1): 549–552, Apr 2018; Palazhy et al.

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| CONTRIBUTION OF THE AUTHORS |
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| SP research design, primary data collection and manuscript (MS) writing; JKT data analysis and preparation of MS; SS research design, supervision and MS preparation. |

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