Evaluation of Acute Toxicity of Chromium to Freshwater Air Breathing Teleosts, *Channa gachua* (Bloch)

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Abstract: ***Heavy metals are naturally occurring elements that have a density at least five times that of water and a high atomic weight. These metallic elements are considered systemic toxicants because they are known to harm different organs, even at lower exposure levels. Water, the most valuable natural resources, is essential to all life. During the experiment, the freshwater fishes Channa gachua were exposed separately to different doses of chromium ($K_2Cr_2O_7$,) for 24 hrs, 48 hrs, 72 hrs and 96 hrs. At first, we determined median lethal concentration (LC_{50}) of $K_2Cr_2O_7$, to fish using graphical interpolation method and confirmed it by probit analysis. Changes in locomotory and behavioral attributes were observed carefully and monitored properly. The treated fishes showed restlessness and rapid random movements. Rapid opercular movements were also observed.***

Keywords: *Channa gachua*, lethal concentration, graphical interpolation method, probit analysis, chromium

1. Introduction

Nowadays, we live in an environment where the purity of water, air, and all heavy metals and other components is in question. Our changing human habits are exacerbating the problem of environmental pollution and contamination to such an extent that the survival of plants and animals is becoming increasingly difficult over time. The anthropogenic causes of environmental degradation are being ignored, which are directly or indirectly harming existing civilization and causing massive damage to human health and the economy at large.

Chromium (Cr) metal is thought to be one of the most prevalent and pervasive contaminants in aquatic environments, it does not occur naturally in its pure metallic mode (Kotas, et al., 2000; Saha, et al., 2011; Shadreck, et al., 2013; Prasad, et al., 2021). In its pure metallic form, chromium exists in three oxidation states: Cr (II), Cr (III), and Cr (VI). Its most unstable form is Cr (II) while Cr (III), and Cr (VI) are described to be in stable oxidation states (Espenson, J. H., 1970; Shupack, S. I. 1991; Apte, et al., 2006; Jiang, et al., 2019; Ao, M. et al., 2022). Notably, chromium metal is frequently utilized by many industrialists for his desirable purpose as electroplating, ore mining, tanneries, textile dyeing and printing, photography in addition with medical sectors and its waste particulates enter into aquatic medium (Narayani, et al., 2013; Siddiquee, et al., 2015; Dasan, et al., 2020; Kerur, et al., 2021).

Exposure of water pollutant through aquatic animal and their health issues are global concern. Of all aquatic animals, fish are the most sensitive to the pollutants because they are the main organisms in the system (Cooper, C. M., 1993; Camargo, et al., 2006; Jayaswal, et al, 2018; Qadri, et al., 2020; Zahoor, et al., 2023). Heavy metals can enter the fish body through contaminated food through the gills, skin or digestive tract. Various research have shown that fish are particularly sensitive to aquatic pollutants because they absorb and store heavy metals such as nickel and chromium in their tissues (Kousar, et al., 2014; Javed, et al., 2019; Garai, et al., 2021; Naeem, et al., 2021; Agbugui, et al., 2022).

Metal bioaccumulation must be monitored in order to determine the process's temporal and spatial scope and to evaluate any possible effects on the health of the organism (Has-Schön et al., 2008). In fish exposed to acute and sub-lethal metal concentrations, bioaccumulation of numerous metals has been documented in the literature (Uysal et al., 2009; Kamunde and MacPhail, 2011). Whenever, fish exposed to various hazardous compounds can often exhibit sub-lethal effects that can be detected or diagnosed using biochemical and physiological biomarkers (Al-Ghanim, K.A. 2012; Keiter, S. 2013). Biochemical variables like antioxidants and enzyme activities and physiological variables like serum levels of metabolites, ions, and hormones are prominent among these biomarkers used as stress indicators (Speich, et al.,2001; Pasupathi, et al., 2009; Firat and Kargin, 2010; Atli, et al., 2015).

The morphological alterations that can be induced by Cr (VI) include epithelial hyperplasia, epithelial lifting, secondary gill lamellae degeneration, lamellar cell hyperplasia, and atrophy of the central axis in tissues. There is a lack of knowledge regarding the genetic and histological toxicity of Cr (VI) to aquatic species (Roberts, et al., 2004). The most significant industrial application for nickel is in the manufacturing of stainless steel. Because to its exceptional strength and durability, ability to resist corrosion, appealing appearance, advantageous thermal characteristics, electrical conductivity, and alloying ability. The production of alloys accounts for around 75% of all nickel consumption (Wilson et al., 1986, Nriagu and Pachana, 1988, Nicolaidou and Nott, 1989). The $LC_{5}o$ of chromium for common carp (Cyprinus carpio) was found to be 191 mgL-1 by Tayyabah et al. (2005). The dosage and time-dependent $LC_{5}o$ of Cr to Channa marulius was reported by Mallesh et al. (2015); Naz and Jawed (2015). As fish became older, the toxicity of chromium to them diminished.

Metal accumulation in the tissue and organs of fish (Oreochromis aureus) depends on the kind of metals and their concentrations, exposure duration, water physico-chemical characteristics, such as temperature, salinity, and hardness (Allen, 1995), metabolic processes, species type, and age (Pelgrom et al., 1995).

2. Materials and Methods

All of the purchased live *Channa gachua* (*C. gachua*) fish was cleaned by using 0.1% KMNO4 to remove cutaneous infections. Fish that were healthy (average length 20-25 cm, weight 45–60 g) were chosen and transferred to large glass tank (aquariums) for stocking and acclimatization to various test settings, including light, temperature (28 ± 2°C), and water physiochemical characteristics, for a period of 15 days.

Water was recycled on alternate day. Tanks always covered with fish netting to prevent the escape of fishes.

Fish were fed with ad libitum, provided with a formulated dite and natural food. Many of the arteficial food as wheat powder, prawn meal, artemia eggs, vitamin B complex, and vitamin A was fed to the fish. To rectify the deficiencies caused by arteficial diet, natural foods like earth worm, and chopped soft meat was provided thrice a week. After feeding the remaining food particles were immediately removed from the tank bottom. Food was stopped one day before the experimental animal (*C. guchua*) which was sacrificed at the end of the experimental period.

Sample water (aquarium's water that were filled with ground/Tap water) were analysed as per the stander method of APHA (2020) for the physiochemical parameters as pH, TDS, TH, alkalinity, chloride, and dissolve oxygen (DO).

Results to the determination of LC_{50} of chromium aquarium setup was arranged in laboratory premises of the Department of Zoology, Patna Science College, Patna University, Patna.

During the analysis of LC_{50}, stock solution of Potassium dichromate ($K_2Cr_2O_7$) was dissolved in deionized water and the stock solution (1000ppm) were prepared. The bioassay of acute toxicity for the chemical of this experiment, potassium dichromate ($K_2Cr_2O_7$) was carried out on the static system to determine the 24, 48, 72, and 96 hours LC_{50} in the experimental fish, C. gachua for the definitive test, the scale finding experiment was executed first to measure the concentration of the experimental solution.

In the definitive test, ten fishes were used for every group. Group 1, 2, 3, 4, 5, and 6 were exposed to rising concentration of, potassium dichromate of ($K_2Cr_2O_7$) whereas the group 1 was retained, potassium dichromate ($K_2Cr_2O_7$,) fresh water to serve as a control. The minimal concentrations of, potassium dichromate ($K_2Cr_2O_7$) tested were: 25ppm, 40ppm, 90ppm, 150ppm, and 180ppm. The exposure duration for the experiment was 24, 48, 72, and 96 hours. The test was replicated three times for the accuracy of this experiment.

By using probit analysis method; above experimental data represents the level of LC_{50}, along with toxicity level of potassium dichromate ($K_2Cr_2O_7$,) upon the *C. gachua.*

Test Chemical

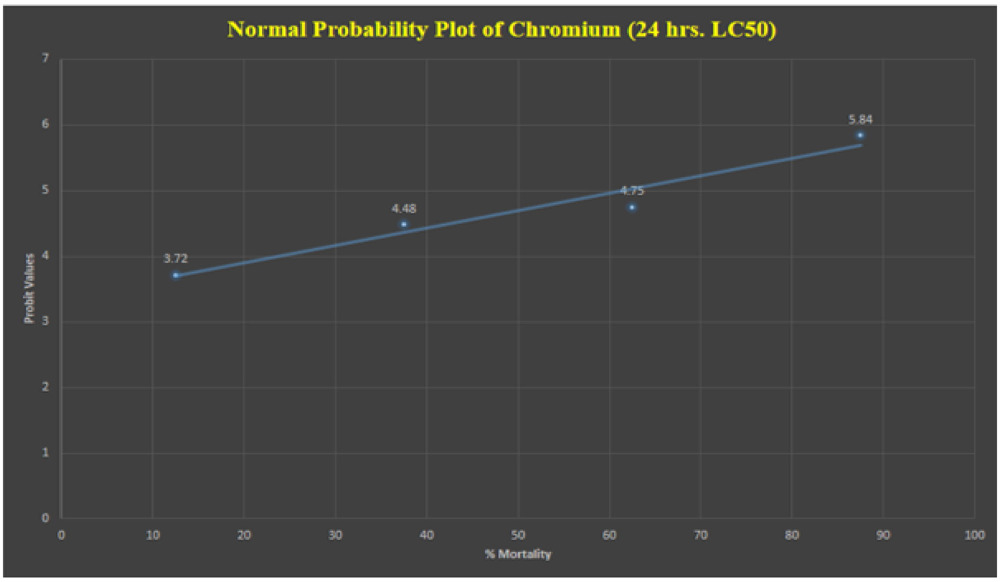
Analytical-grade Potassium dichromate ($K_2Cr_2O_7$,) was used to prepare a stock solution in dechlorinated water, from which the required nominal test concentrations were obtained by serial dilution.

Table 1: Data shows LC_{50} value of chromium as $K_2Cr_2O_7$, for a period of 24, 48, 72, and 96 hrs. in freshwater fish *C. gachua*.

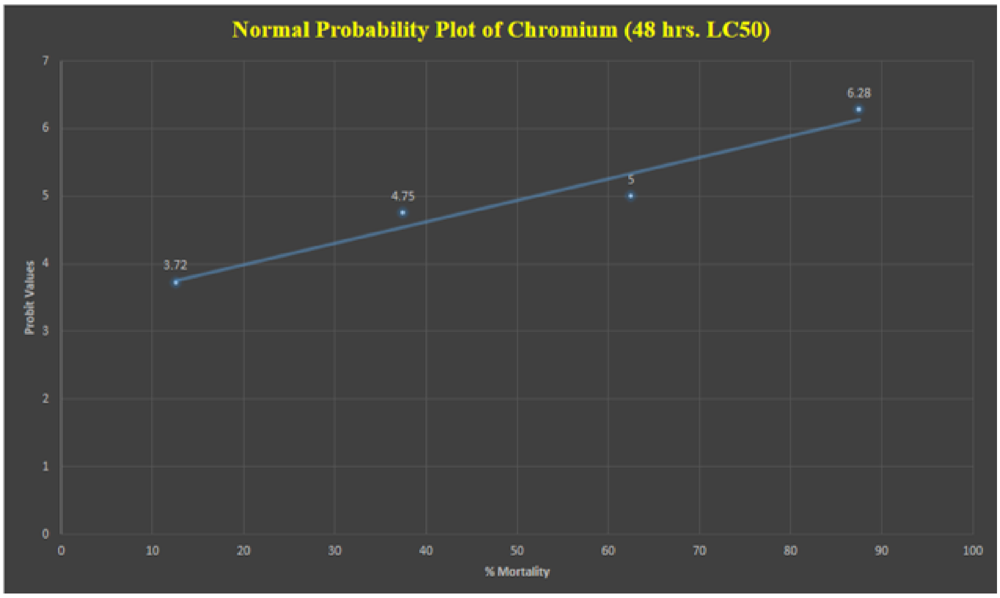
Aquarium No.	No. of test fish C.Gachua	Chromium Concentration	Mortality During							
			24hrs.		48 hrs.		72 hrs.		96 hrs.	
			No.	%	No.	%	No.	%	No.	%
Gr. I	10	Control	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
Gr. II	10	180ppm	8	70	9	90	10	100	-	-
Gr. III	10	150 ppm	4	40	5	50	7	70	8	80
Gr. IV	10	90 ppm	3	30	4	40	4	40	5	50
Gr. V	10	40 ppm	1	10	1	10	2	20	3	30
Gr. VI	10	25 ppm	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
Gr. VII	10	10 ppm	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil

Probit Regression Analysis (Cr)

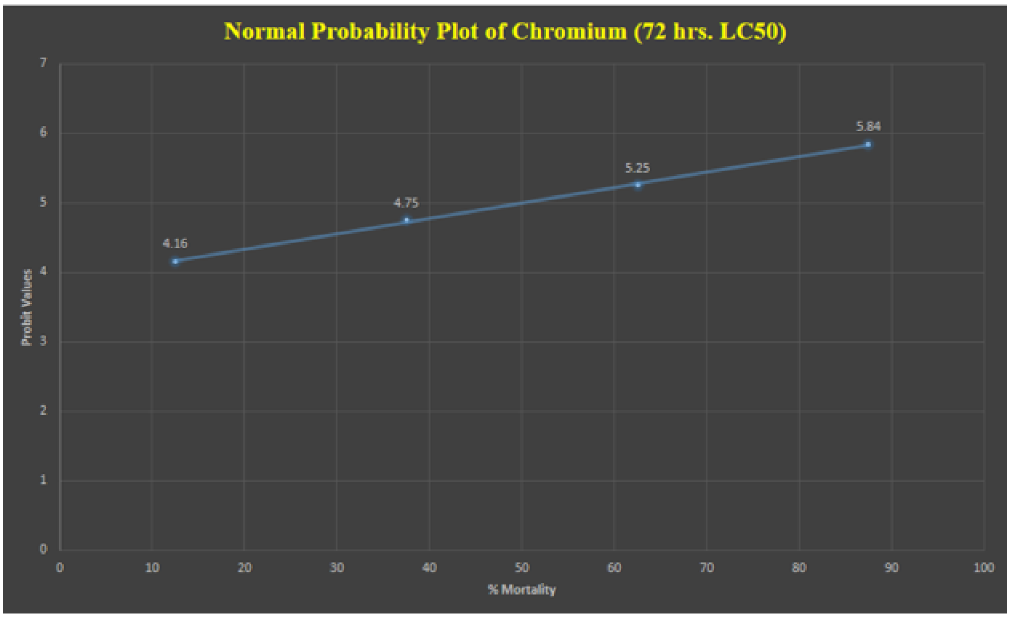
@ 24h


Normal Probability Plot of Chromium (24 hrs. LC50)
Probit Values
% Mortality
3.72
4.48
4.75
5.84

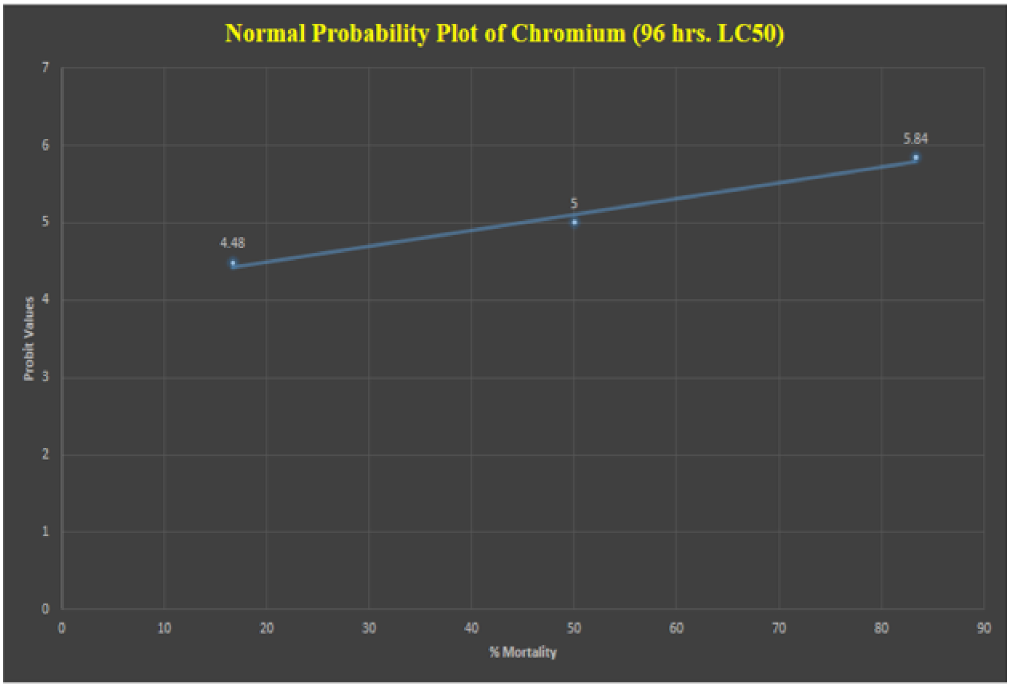
@ 48 h


Normal Probability Plot of Chromium (48 hrs. LC50)
Probit Values
% Mortality
3.72
4.75
5
6.28

@ 72 h


Normal Probability Plot of Chromium (72 hrs. LC50)
Probit Values
% Mortality
4.16
4.75
5.25
5.84

@ 96 h



3. Discussion

The present study establishes, for the first time, the acute toxicity profile of potassium dichromate in *Channa gach*ua, with a 96-h LC50 of 90 mg/L. Mortality increased in a clear concentration- and time-dependent manner, consistent with the classical acute toxicity pattern reported for cr across freshwater teleosts.

The concentration-dependent behavioural alterations noted in the present study– including surfacing/air-gulping changes, altered opercular movement, and abnormal swimming, Increased mucus secretion and surface discoloration, when observed, are consistent with a generalized stress and epidermal irritation response reported across chromium exposed fish species.

Because *C. gachua* relies on periodic surfacing for aerial respiration through its suprabranchial chamber, an ecological consideration previously highlighted in comparative toxicity work on this species5. This physiological trait strengthens the ecological relevance of *C. gachua* as a sentinel species for surface-water metal contamination in shallow wetland habitats, even though its acute LC_{50} in the present study did not indicate exceptional sensitivity relative to its congener**.**

4. Conclusion

The fish *Channa gachua* were exposed to different concentrations of potassium dichromate. The clear concentration- and time-dependent mortality of this air-breathing species as a bioindicator for chromium contamination in freshwater The LC_{50} values were calculated at 24 hours, 48 hours, 72 hours and 96 hours. We found here high values of the LC_{50} which might have been due to various factors. Further studies are needed to understand the key factors that are playing vital role in variations in the LC_{50} values in C. gachuиa 50%

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Paper ID: SR26729225514 DOI: https://dx.doi.org/10.21275/SR26729225514

Archives of environmental contamination and toxicology, 58, 151-157, 187 Anukriti (An International Peer Reviewed Multidisciplinary Refereced Research Journal) Vol. 15, No. 10, October 2025 Impact Factor: 8.0 ISSN: 2250-1193

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