

Toxic Effect of Heavy Metal, Cadmium Concentration in Baraila Lake Fish Water *Channa Punctatus*

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Received: 2025, 15, Jul

Accepted: 2025, 21, Aug

Published: 2025, 30, Sep

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Annotation: Pesticides are synthetic chemical used for pest control. A major source of pesticides in water bodies is the runoff from agricultural fields. Some pesticides also enter inland waters from industries, which use pesticides in their manufacturing processes or from the manufacturing of pesticides themselves. Pesticides adversely affect a wide range of organisms including insects and fish. Pesticides accumulate in the tissues of aquatic organisms through bio concentration and biological magnification and adversely affect their metabolic processes including reproduction.

The experimental fish sub lethal concentration of Cadmium chloride caused various behavioral abnormalities in fish such as an erratic increased swimming, surfacing and hyperactivity, restlessness, abnormal swimming, and secretion of mucous which was followed by loss of balance and succumbing of fish, when they are initially exposed to cadmium chloride test solution. Throughout the exposure period the fishes showed various aggressive

behavior abnormalities such as nudge and nip, fin flickering, partial and S-jerk and burst swimming increased significantly ($P < 0.05$) compared with control. It is also observed that the aforementioned responses are significantly and gradually.

The sublethal concentration of Cadmium chloride caused changes in various behavioural and haematological indices of *Channa punctatus*, including the accumulation of bluish white mucous-like substance clogged on the vital external areas of the fish skin. These deposits were suspected to be salts of cadmium. They were noticed only in the Cd treated fishes, which indicated contamination through water.

Keywords: Toxic, effect, cadmium, concentration, Lake water, *Channa punctatus*.

INTRODUCTION

Pesticides are synthetic chemicals used for pest control. A major source of pesticides in water bodies is the runoff from agricultural fields. Some pesticides also enter inland waters from industries, which use pesticides in their manufacturing processes or from the pesticides themselves. Pesticides adversely affect a wide range of organisms, including insects and fish. Pesticides accumulate in the tissues of aquatic organisms through bioconcentration and biological magnification and adversely affect their metabolic processes, including reproduction.

Some pesticides kill not only the target organisms but they also adversely affect many non-target organisms. Hence the term biocide is often used for them in the environment. A biocide may be detoxified or its toxicity is changed in some way. A good example of the second possibility is DDT, which is quite toxic to many insects and relatively nontoxic to birds. DDT is metabolized by detritivores along two pathways: under anaerobic conditions to TDE (also called DDD) and under aerobic conditions to DDE.

Cadmium is a biotoxic environmental pollutant which accumulates in the body tissues such as the lungs, liver, kidneys, bones, reproductive organs and the immune system. The toxic effects of cadmium on organisms include nephrotoxicity, carcinogenicity, teratogenicity and endocrine disruption. Cadmium may also cause the deterioration of cell membranes by binding to metallothionein (MT) or glutathione and consequently interfere with the ability of these proteins to avoid oxidative stress. Cadmium can also replace essential metals such as copper and zinc in several metalloproteins, altering the protein conformation and affecting their activity because this element interacts ubiquitously with sulfhydryl groups of amino acids, proteins and enzymes. Cadmium can cause oxidative stress through several mechanisms; the Fenton reaction, depletion

of cellular antioxidants, alterations in the mitochondrial electron transfer chain and inhibition of antioxidant enzymes.

With the discovery of new uses of cadmium, its presence in the environment has increased. Cadmium is used in the production of television picture tube phosphorus, nickel cadmium batteries, motor oils, curing agents for rubber, fungicides, phosphate fertilizers, stearate stabilizers for plastics (polyvinyl chloride) and shields for nuclear reactors. Cadmium is used primarily for electroplating other metals or alloys to protect them against corrosion and in the manufacture of low melting point alloys or solders.

Fish is generally appreciated as one of the healthiest and cheapest sources of protein and it has amino acid compositions that are higher in cysteine than most other sources of protein. Heavy metals like copper, iron and zinc are essential for fish metabolism while some others such as mercury, cadmium, arsenic and lead have no known role in biological systems.

Notable contributions are those of Prasad, B. (1932); Rao, K. (1969); Rao, Vasishta, I.S. and S. Gulati (1971); Roy, S.P. (2003); Sharma, U.P., Roy, S.P. and Rai, D.N. (1983); Seth, R.N. (2003); Bhumika, Shah S. and Mitra A. (2000); Bhatnagar, S.K. (2000); Berube, L. (1964); Brown, D.S. (1965); Choudhary, B.N., Chaudhary, H., Ujjana N.C.

Many workers have reported the manifestation of toxic effects of cadmium on various fishes. However, effects of cadmium on freshwater fishes, especially on the carp fish *Channa punctatus*, have been documented less. The present work is aimed to evaluate the LC50/96h of Cadmium chloride and to observe the variations in opercular beats, behavioural modulations and haematological indices of the freshwater fish, *Channa punctatus*, which has good nutritive value and can serve as a better indicator of freshwater streams, lakes and ponds.

MATERIALS & METHODS

The freshwater fish, *Channa punctatus*, were selected as the test organisms for the present study. They were collected from the Baraila lake with the help of local fishermen and immediately brought to the working laboratory, where they were acclimatized to laboratory conditions for 15 days prior to the experiments. A stock solution of Cadmium chloride ($\text{CdCl}_2 \cdot 2\frac{1}{2}\text{H}_2\text{O}$, MW 228.35, AR Grade) was prepared by dissolving a weighed amount of the salt in double-distilled water.

For toxicity tests, ten aquaria of 50-liter capacity were taken, each having 30 liters of dechlorinated and oxygenated tap water (Physico-chemical properties of water: pH = 7.6 ± 0.2 , Temp. $26 \pm 2^\circ\text{C}$, Alkalinity = 66 ± 4.5 mg/L, Total Hardness = 259 ± 2.5 mg/L, D.O. = 7.6 ± 0.2 mg/L). A series of concentrations of Cadmium chloride, viz., 140, 200, 280, 380, and 500 mg/L (the toxic range was predetermined by exploratory tests), was prepared by adding a calculated amount of the stock solution. One aquarium containing normal water without Cadmium chloride served as the control, in which the test animals were maintained.

Ten adult fishes of similar size and weight (average length 16 ± 1.5 cm and weight 21.5 ± 2.0 gm) were introduced into each test as well as control aquarium from a stocking tank. The fish had been acclimated to laboratory conditions in dechlorinated tap water for 2 weeks prior to the assay in 100L capacity plastic tanks. The mean values for the test water quality during this period were as follows: temperature $22 \pm 1^\circ\text{C}$, pH 5.7 ± 0.4 ; dissolved oxygen 4.7 ± 0.7 ppm; hardness 36 ± 1.24 ppm. The fish were fed *ad libitum* daily with pelletized formulated feed and maintained at a photoperiod of 12h light: 12h dark. Feeding was suspended 24 hours before the start and throughout the experiment to avoid dissolved Cadmium losses due to particulate adsorption. Proper aeration was maintained in test as well as control aquaria by air pumps and stone diffusers throughout the experimental period. Mortality was recorded at 24-hour intervals. The LC50 values at various intervals were calculated according to the method of Finney et al.

Stock solutions of $\text{CdCl}_2 \cdot \text{H}_2\text{O}$ were prepared by dissolving the toxicant in distilled water to a final volume of 10ml. The stock was then serially diluted into relevant treatment concentrations. The

toxicant in the test chamber was renewed completely with a fresh solution of the same concentration every 24 hours. A concentration of 1/10th of the 96h LC50 was taken as the sub-lethal concentration for haematological analysis over a period of 3 days, 7 days, 15 days, and 30 days. Four replicates were maintained simultaneously, with 10 fish introduced in each concentration per tank. The experimental fish were also fed with formulated feed *ad libitum* as in the control.

After the expiry of each exposure period, blood was collected from each fish by means of a hypodermic syringe at the base of the caudal peduncle and immediately transferred to EDTA containers for haematological analysis. Haemoglobin (Hb) was estimated as cyanmethemoglobin according to Ochei and Kolhatkar. RBC and WBC were determined according to the classical method using the Neubauer hemocytometer. Hematocrit was determined using microhematocrit tubes and a haematocrit centrifuge. Blood was centrifuged at 1200 x g for 5 minutes, and the haematocrit value was obtained.

Data were analyzed by one-way analysis of variance (ANOVA) followed by Duncan's Multiple Range post-hoc test using SPSS 15 Software. Statistical significance was considered at a $p \leq 0.05$ level. The LC50 value and their confidence limits of Cadmium chloride for *Channa punctatus* are summarized in the Table. The 24, 48, 72, and 96h LC50 values of Cadmium chloride were 14.59, 10.76, 8.45, and 5.36 mg/L, respectively. It was observed that an inverse relationship between exposure duration and lethal concentration was clearly evident.

RESULTS AND DISCUSSION

The experimental fish, exposed to a sublethal concentration of Cadmium chloride, exhibited various behavioral abnormalities. These included erratic and increased swimming, surfacing, hyperactivity, restlessness, abnormal swimming patterns, and secretion of mucous. This was followed by a loss of balance and succumbing of fish when initially exposed to the cadmium chloride test solution. Throughout the exposure period, the fishes showed various aggressive behavioral abnormalities such as nudging and nipping, fin flickering, partial and S-jerks, and burst swimming, which increased significantly ($P < 0.05$) compared with the control. It was also observed that the aforementioned responses increased significantly and gradually with the increasing exposure period. The behavioral abnormalities observed in fish treated with a sublethal concentration of cadmium chloride are summarized in the Table.

The LC50 values of Cadmium chloride and the upper and lower confidence limits for *Channa punctatus* are summarized in Table 2.

Table 2: LC50 values of Cadmium chloride and upper and lower confidence limits in *Channa punctatus*.

Exposure Period (hrs.)	LC ₅₀ (mg/l)	Lower confidence limit (mg/l)	Upper confidence limit (mg/l)
24	14.59	16.03	11.85
48	10.76	13.29	8.68
72	8.45	11.28	6.56
96	5.36	8.14	3.93

Cadmium chloride induced marked effects on the opercular beats per minute of *Channa punctatus*. The test fishes exhibited opercular beats that were found to be significantly higher throughout the experiment. Although a decline was noticed from the 24-hour to the 48-hour exposure period, the values were still significantly higher than the controls. The mean values of opercular beats at 72 and 96 hours were higher than the control and also higher than the other exposure periods. The differences between the mean opercular beats of test animals and control animals were highly significant at the 24, 48, 72, and 96-hour exposure periods. The overall fluctuations in the mean

opercular beats from 24 hours to 96 hours were found to be significant in both test and control animals.

The data on variations in haematological indices of *Channa punctatus* exposed to a 1/5th sublethal concentration for 96 hours is presented. It is evident that the heavy metal Cadmium inflicted a drastic and gradual reduction ($p < 0.05$) in RBC, Hb, and Hct in fish over the 30-day exposure period relative to the control, whereas the total leucocyte count (WBC) increased with the increased concentration of cadmium chloride

Table 3. Variations in Opercular beats/minute of *Channa punctatus* after exposure to Cadmium chloride

Exposure Period (hrs)	Control	Experimental
24	61.2 ± 4.74	84.3 ± 3.43
48	66.4 ± 4.78	88.3 ± 3.54
72	73.4 ± 4.26	95.4 ± 4.55
96	78.4 ± 4.55	116.5 ± 3.33

The sublethal concentration of Cadmium chloride caused changes in behavioural and haematological indices of *Channa punctatus*, including the accumulation of a bluish white mucous-like substance clogged on the vital external areas of the fish skin. These deposits were suspected to be salts of cadmium. They were noticed only in the Cd-treated fishes, which indicated contamination through water.

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