THE INTERNATIONAL JOURNAL OF SCIENCE & TECHNOLEDGE

Studies on Toxicity and Behavioural Responses under Cadmium Stress in *Channa punctatus* (Bloch.)

Sujata Kawade
Department of Zoology, Dr. Babasaheb Ambedkar Marathwada University, Aurangabad, India
Yashwant Khillare
Department of Zoology, Dr. Babasaheb Ambedkar Marathwada University, Aurangabad, India

Abstract:
Contamination of aquatic environment with heavy metals has become a matter of great concern because of their toxicity, persistence and bioaccumulation. The present study deals with the toxicity of Cadmium to fresh water fish, Channa punctatus. Static bioassay tests were carried out to evaluate LC_{50} values of Cadmium (Cd) as $CdCl_2$ for fish, Channa punctatus as well as to observe the behavioural alterations imposed by Cadmium. The LC_{50} values for 24, 48, 72 and 96 hrs were found at 630, 538, 452 and 360 ppm respectively. The major behavioural alterations observed during the experiment were erratic swimming, gulping of air at the surface, opercular movements, loss of equilibrium, restlessness, sluggishness and jumping in exposed animals. The observed data shows that Channa punctatus can be used as a good biomarker to detect pollution status of fresh water bodies.

Keywords: *Channa punctatus, Cadmium, LC_{50}, Behavioural changes*

1. Introduction

Discharge of heavy metals into the aquatic environment has created severe health hazards in the aquatic organisms including man. Although their normal level is not dangerous to aquatic life and are known to be essential to sustain life activities, recent advances in industrialization and agricultural development has been considered as the main source of metal in aquatic ecosystem [20]. Accumulation of industrial effluents and agricultural runoff in water bodies has become a topic of major concern [14]. The recent developments have brought forth scientists to take a remedial action to this problem. The harmful effects of the pollutants depend upon the functions of concentrations of chemical pollutant and its duration of exposure. Though industrialization and development in agriculture are necessary to meet the basic requirement of people, at the same time it is necessary to preserve the environment. In India, due to lack of sewage treatment plants, generally untreated effluents are released either on agricultural land or disposed off in the nearby water bodies [36, 5]. The effluents containing toxic heavy metals like Mercury, Copper, Cadmium, Nickel, Zinc, Chromium, etc are non-biodegradable and bioaccumulate in the aquatic organisms, espically fishes through food chain [4, 19].

Fish population is genereally considered sensitive to all kinds of environmental stressors [9, 6]. Effect of different heavy metals on aquatic organisms has been studied by many workers. Alterations in protein level were reported in the flesh and liver of *Cyprinus carpio* on exposure to mixture of Nickel and Chromium [32]. Mortality and growth alterations were recorded in rainbow trout on exposure to Copper [12]. Alterations were also reported in the biochemical constituents of muscles of *Cirrhinus mrigala* following exposure to heavy metals [33]. Changes in the biochemical profiles of *Labeo rohita* were observed on Chromium exposure [34]. Effect of Chromium on the survival, growth, oxygen consumption and biochemical profiles of *Labeo rohita* (Ham.) were reported on Chromium exposure [35]. Histopathological lesions were observed in the gills and kidney of *Cirrhinus mrigala* (Ham.) fingerlings on exposure to Mercury [11].

Among heavy metals, Cadmium has been blacklisted by European community [21]. It is a non corrosive and highly toxic metal. It is a nonessential element with no known biological function. It is used in batteries, plastics, metal alloys, dye and metal plating industries. Effluents from such activities are sources of Cadmium in aquatic environment. A higher concentration of Cd in the aquatic environment is lethal to many organisms [37, 7]. Cadmium has been considered as an important xenobitic, persistent and nonbiodegradable chemical pollutant in the aquatic environment [18].

Toxicity testing is an essential tool for accessing the action and fate of toxicants in aquatic ecosystems, to derive water quality standards for chemicals and to identify suitable organisms as bioindicators. The purpose of toxicity test is to access various abnormalities cused due to administration of a chemical or pollutant and to determine the order of lethality of the chemical [2]. Toxicity in fish is a result of a series of events involving various physical, chemical and biological processes. Estimation of median lethal concentration or dosage (LC_{50} or LD_{50}) is valuable as it can be used as an indicator to the level of resistance of population response to metals [25].

Fishes store, concentrate, metabolize toxicants and exhibit behavioural changes. They also act as biomarkers to detect the pollution status of water bodies. Behaviour allows an organism to adjust to the external and internal stimulus in order to meet the challenges of survival in a changing environment. Effects of heavy metals on the behavioural changes have been reported by many workers. Individual and combined effect of heavy metals on behaviour and respiratory responses of *Oreochromis*

mossambicus were recorded [15]. Cadmium accumulation and physiological alterations were recorded in Rainbow trout on Cadmium exposure [13]. Toxicity of Cadmium on behavioural and morphological aspects in *Labeo rohita was* recorded [22].
Study of behavioural changes due to the effect of toxicants in fishes is the best suitable method to check the pollution in water bodies [29]. Thus it is a promising tool in ecotoxicology and act as a diagnostic endpoint for screening, differentiating the effluents and their effects on aquatic organisms.

2. Material and Methods

Adult and live *Channa punctatus* were collected from the local market and brought to laboratory. Only healthy fishes (Length: 15-20 cm, Weight: 55-60 g) were taken for experiment. Fishes were acclimatized in glass aquaria for 15 days and were fed with fish food (earthworms) and water in the aquaria was replaced at every 24 hrs. Stock solution of Cadmium Chloride was prepared by dissolving appropriate amount of $CdCl_2$ as Cd salt in distilled water. The test fish were exposed to Cd as $CdCl_2$ to know the acute toxicity at 24, 48, 72 and 96 hrs and the behavioural changes were recorded simultaneously at the exposure periods. For selection of test concentration, some pilot tests were carried out. The range of concentration was selected between 0 to 100% mortality. In order to maintain the concentration of Cadmium, the water in the aquaria was changed every 24 hrs during the exposure. The percentage for corrected mortality was given by [1]:

$$\text{Corrected mortality (\%)} = \frac{\text{Percentage living in control - percentage living in treatment}}{\text{Percentage living in control}} \times 100$$

The LC_{50} value for 24, 48, 72 and 96 hrs was determined by static renewal bioassay following probit analysis [10]. For studying the behavioural changes, fishes were divided in two groups as control and experimental.

3. Results

3.1. Toxicity

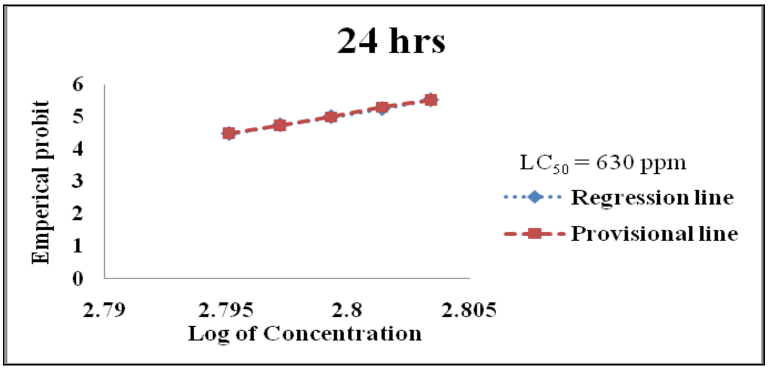
The test fish *Channa punctatus* were exposed to heavy metal Cadmium (Cd) as ($CdCl_2$) for 24, 48, 72 and 96 hrs and the LC_{50} values were recorded at 630, 538, 452 and 360 ppm respectively. [Table 1, Fig1:1(A), 1(B), 1(C), 1(D)]

3.2. Behavioural Responses

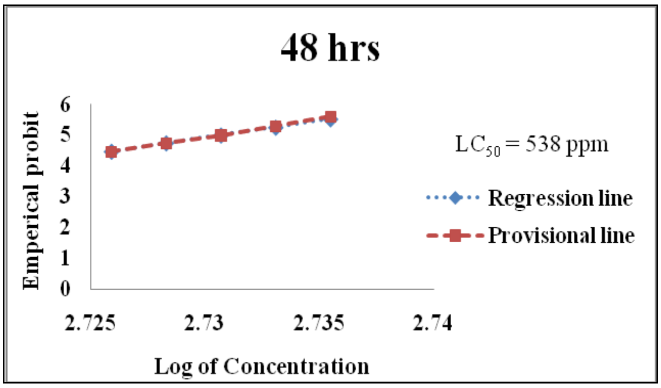
- Control fish: Fishes were active with well synchronized movements. They mostly settled to the base of the aquarium tub, sometimes came to the surface of water and actively responded to slight disturbance.
- Treated fish: The treated fishes showed erratic swimming, loss of equilibrium, restlessness, sluggishness, jumping, opercular movements and gulping of air at surface on acute exposure to Cadmium. Lastly they remained in a vertical position for a few minutes with the anterior side or terminal mouth up near the surface of water, trying to gulp the air and the tail in a downward direction. Soon they settled at the bottom of tank and after some time their bellies turned upward and the fish died. (Table 2)

Exposure period in hrs.	**LC_{50} Values (ppm)**	**Regression equation $Y = y + b (x – x)$**	**Variance**
24	630	Y = 5.8552 X – 11.389	0.000963
48	538	Y = 5.6444 X - 10.4135	0.001048
72	452	Y = 7.2368 X – 14.2138	0.000636
96	360	Y = 12.9782 X – 28.1749	0.000197

Table 1: LC_{50} values for heavy metal Cadmium (Cd) as $CdCl_2$, for a period of 24, 48, 72 and 96 hrs in freshwater fish, Channa punctatus



1(A)



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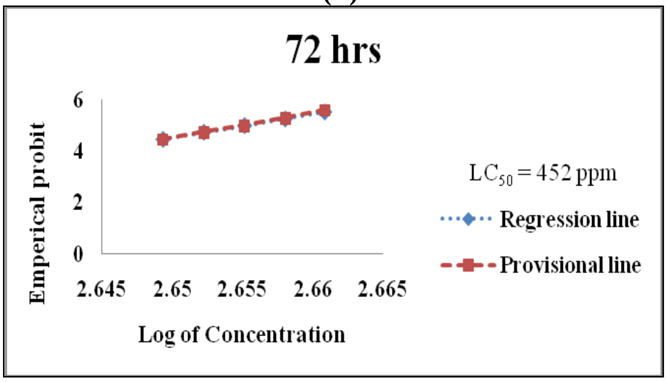
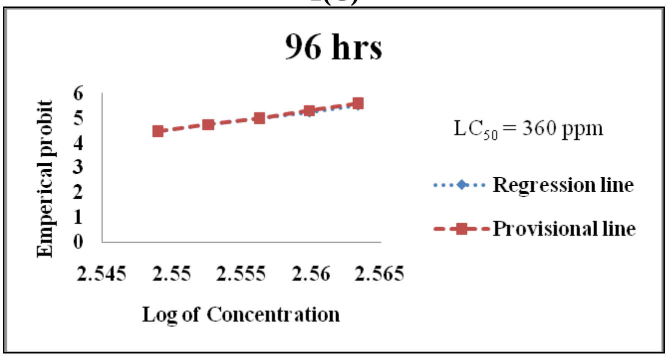


1(C)



1(D)

Figure 1: Average probit analysis of heavy metal Cadmium (Cd) as CdCl2 on exposure to fresh water fish, Channa punctatus for a period of 24, 48, 72 and 96 hrs [1(A), 1(B), 1(C) and 1(D) respectively]

Behavioural changes	Control	Cadmium (Cd) as $CdCl_2$			
		24 hrs	48 hrs	72 hrs	96 hrs
1.Erratic Swimming	-	+++	+++	+++	++
2.Loss of Equilibrium	-	+	++	+++	+++
3.Restlessness	-	+++	+++	+++	++
4.Sluggishness	-	+	++	++	+++
5.Jumping	-	+++	+++	+++	++
6.Opercular movements	-	++	++	+++	+++
7.Gulping air at surface	-	+++	+++	+++	++

Table 2: Comparative behavioural responses given by fish Channa punctatus after exposure to 24, 48, 72 and 96 hrs to heavy metal Cadmium (Cd) as $CdCl_2$

Note:
Symbol:
(-) Normal response
(+) Mild increase response
(++) Moderate increase response
(+++) Maximum increase response

4. Discussion

The fresh water ecosystem is under increasing threat due to rapidly expanding population and the subsequent modernization processes resulted in inconspicuous exploitation of nature leading to pollution crises. At present environmental protection is the main need of the society.

Toxcological studies of the pollutants upon aquatic organisms are very important from the view piont of environmental consequences. Fishes are often forced to encounter highly contaminated waters especially in areas where dilution rate of waste waters is low. Estimation of LC_{50} of heavy metals is important with reference to a particular fish species. According to Brongs and Mount, the application of the LC_{50} value is the most highly reliable test for accessing the adverse effects of a chemical contaminant to aquatic life. In the present study, acute toxicity of Cadmium on the test fish, *Channa punctatus* has been analysed and behavioural responses were also recorded. Cadmium poses toxic effect on fish *Channa punctatus* which is evident from the findings of present investigations [31, 28, 8].

Behavioural alterations have been established as sensitive indicators of chemically induced stress in aquatic organisms [27]. Behavioural changes in animals are indicative of internal disturbances of the body functions such as inhibition of enzyme functions, impairment in neural transmission nervous impairment due to blockage of nervous transmission between the nervous system and various effectors sites and disturbance in metabolic pathways. Behavioural alterations like erratic swimming, restlessness and jumping observed in the present study may be an avoiding reaction to the heavy metals. The avoidance reaction may be related to change in sensitivity of chemoreceptors. Similar results were reported by [30, 3]. Sluggishness observed at the end of exposure periods may be due to loss of energy as a result of erratic swimming, jumping and restlessness. Loss of balance during swimming, observed during this study, might be due to some neurological impairment in the Central nervous system. Similar results were reported by [17, 24]. In the present study, increased opercular activity may be due to stressful toxic environment along with sensory stimulus to increase the opercular movement for proper ventilation of gills to cope with the hypoxia [17]. Surfacing and gulping of air might be an attempt to cope with the oxygen deficiency. The increased opercular movement and gulping activity by the treated fish may be an attempt to extract more oxygen to meet the increased energy demand to withstand the Cadmium toxicity [26]. It might also be due to accumulation of mucous on the gill epithelium producing hypoxic condition to the fish. Similar findings were reported by [16, 23].

Thus acute toxicity studies can help to detect and evaluate the degree of pollution by providing reliable estimates of safe concentration from which water quality criteria can be derived. The knowledge obtained from dose response studies in animals is used to set standards for human exposure and the amount of chemical residue that is allowed in the environment. Behavioural toxicology is a rewarding tool for hazard assessment of water pollution. The behavioural alterations in fish can be considered as biomarkers to access the health status of the fishes as well as aquatic bodies polluted by toxicants. Thus environmental protection is the major requirement of the society.

5. References

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