



CADMIUM CHLORIDE INDUCED CHANGES IN GLYCOGEN CONTENT OF HEPATIC AND GILL TISSUES OF CHANNA PUNCTATUS

Diksha Bhanu^{1*}, Prof. Preety Sinha²

¹Research Scholar, P.G Department of Zoology, Patliputra University, Patna-800020, Bihar (India)

²Head, P.G. Department of Zoology, Patliputra University, Patna-800020, Bihar (India)

*Corresponding Author's Email ID: dikshabhanu2112@gmail.com

*Corresponding Author's Address: P.G. Department of Zoology, Patliputra University, Patna

ABSTRACT

Various contaminants, including toxic heavy metals such as cadmium, copper, mercury, and zinc, are widely present in rivers and reservoirs, posing a serious threat to aquatic life. Cadmium pollution in the environment is caused by the development and utilization of chemical fertilizers in agriculture, metals and minerals in industry, and the use of some industrial products containing Cd, which may be discharged into water with wastewater. Channa punctatus, commonly found across India, is highly valued commercially due to its easy cultivation, exceptional nutritional benefits, and availability throughout the year. The study revealed a significant, time-dependent reduction of total glycogen levels in both hepatic (liver) and gill tissues of Channa punctatus following exposure to sub lethal concentrations of cadmium. The decline was more pronounced at higher concentrations (Group-II) and longer exposure durations (21 days). In contrast, the control group showed stable glycogen levels throughout the experimental period, indicating that glycogen exposure disrupts glycogen metabolism in a dose- and duration-dependent manner.

Keywords: Cadmium, Glycogen, Liver, Gill, Channa

INTRODUCTION

Water, second only to air, is a fundamental component of our life support system, its quality is crucial for maintaining health. As a vital natural resource, it serves multiple purposes, including aquaculture, industry, irrigation, and domestic use. Although our country possesses abundant water resources, rapid industrialization, population growth, and the indiscriminate exploitation of natural resources have significantly escalated water pollution issues. Various contaminants, including heavy metals which are toxic in nature such as zinc, cadmium, copper, and mercury, are widely present in rivers and reservoirs, posing a serious threat to aquatic life (Olsson, 1998). These metals are generally non-biodegradable, leading to their

accumulation in fish, mussels, sediments, oysters, and other components of aquatic ecosystems, a phenomenon observed globally. Even at trace levels, heavy metals (Cu, Mn, Ni, Cd, Cr, Fe, Zn, Co, Hg, Pb) can significantly impact fish morphology, physiology, and biochemical processes. These effects include suppressed immune function (Mikryakov VR et al. 2001), behavioral alterations, stunted growth, variations in digestive enzyme activity (Golovanova IL et al. 2006), reduced food assimilation efficiency (James R et al. 1995) and disturbances in carbohydrate metabolism (Bhatkar N et al. 2004, Campbell PGC et al 2003, Zutshi BSG et al. 2010). Glycogen serves as the primary energy source for the animals, and its levels in the liver and muscles reflect overall health of the fish. (Tariang et al. 2019). Cadmium (Cd) is extremely toxic metal contaminant naturally occurring in the Earth's crust and soil at a concentration of ~0.2 ppb. It is predominantly released as a by-product of mining, smelting, and refining sulfide ores, particularly zinc, with lesser contributions from lead and copper extraction processes. Cadmium pollution arises from the use of chemical fertilizers in farming, industrial processing of metals and minerals, and certain Cd-containing products that release it into water through wastewater. Even at low levels, cadmium can build up in algae and sediments, be taken up by aquatic animals like fish and shellfish, and move through the food chain to eventually reach humans. (Satarug et al., Wang et al. 2021, Garner et al. 2016, Schaefer et al. 2020).

Fish serve as key indicators in the study of heavy metal contamination, as they freely navigate aquatic environments and absorb heavy metals through multiple pathways. These include ingesting suspended particles, ion exchange of dissolved metals through their gills, and surface adsorption onto tissues and membranes. *Channa punctatus*, commonly found across India, is highly valued commercially due to its easy cultivation, exceptional nutritional benefits, and availability throughout the year. Additionally, *Channa* is very commonly used as a model organism in toxicological research (Sharma et al., 2017). Its rich nutrient content and steady market demand further reinforce its significance as one of the most preferred freshwater fish.

METHODS AND METHODOLOGY

Test Animal

Channa punctatus (12 – 15 cm length and 22 ± 3 gm weight) was used in this study were sourced from a local fish market in Patna, Bihar, India. The fish were acclimatized in the laboratory for approximately one week in 70-liter glass aquaria at Pisciculture Lab of A.N. College, Patna. Before acclimatization, they were treated with 0.1% KMnO_4 solution for two minutes to prevent dermal infections. The water used for maintaining the fish in the aquaria had a pH of 7.2 ± 0.1 , dissolved oxygen levels of 8.0 ± 0.3 mg/L, and bicarbonate concentrations of 95.0 ± 5.0 mg/L. Fishes were provided with a regular diet of commercial feed (Taiyo), and the aquatic medium was refreshed on a daily basis to eliminate waste and uneaten food.

Determination of LC50

Cadmium chloride ($\text{CdCl}_2 \cdot \text{H}_2\text{O}$) was obtained from Naina Enterprises, located on Ashok Rajpath, Patna, India (GSTIN: 10AQKPP2064M1ZR), and utilized in the current study. The median lethal concentration (LC_{50}) of cadmium chloride over a 96-hour exposure period was determined to be 60.53 mg/l using the probit analysis method outlined by Finney (1971). Based on the 96-hour LC_{50} value, fish were subjected to

sublethal concentrations equivalent to 1/10th (Group – I, 8.053mg/l) and 1/5th (Group – II, 16.106mg/l) of the LC₅₀ for experimental durations of seven, fourteen and twenty one days. A control group was maintained under identical environmental conditions to ensure consistency and comparability. At the end of each exposure period (7th, 14th, and 21st days), specimens from both control and treated groups were sacrificed for biochemical assessment.

Estimation of Glycogen

Fishes were sacrificed for the determination of glycogen level. Glycogen was estimated using the method of Nicholas et al. (1956). Liver and gill samples were digested with hot 30% KOH, cooled, and mixed with ethanol, then refrigerated overnight. After centrifugation, the supernatant was removed, and the residue was dissolved in warm distilled water. A portion of the solution was mixed with anthrone reagent in sulfuric acid, heated for 10 minutes, cooled, and the absorbance was measured at 620 nm using a spectrophotometer.

RESULTS AND DISCUSSION

Fish are effective bioindicators of aquatic pollution due to their continuous interaction with the water for respiration and nutrition. Cadmium, a non-degradable and persistent heavy metal, can accumulate in fish tissues over time. Glycogen serves as a readily available energy reserve that is broken down into glucose through glycogenolysis during periods of stress, such as exposure to environmental pollutants. A concentration-dependent reduction in glycogen levels was observed in both liver and gill tissues of *Channa punctatus*.

Table 1: Impact of sublethal cadmium concentrations on glycogen content (mg/g) in the liver tissue of *Channa punctatus* across various exposure durations (N = 6).

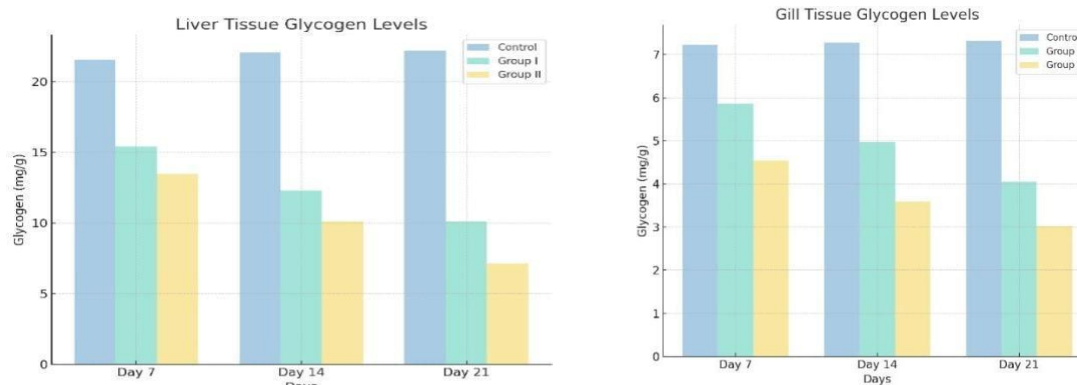
Duration of exposure (Days)	Control group	Group- I	Group- II
7	7.23±0.26	5.86±0.21	4.54±0.14
14	7.28±0.21	4.98±0.31	3.59±0.25
21	7.32±0.31	4.05±0.12	3.02±0.31
F. value	0.031**	29.17*	33.16*

**=Non Significant; *=Significant at 5% level of F test (p<0.05)

Table 2: Impact of sublethal cadmium concentrations on glycogen content (mg/g) in the gill tissue of *Channa punctatus* across various exposure durations (N = 6).

Duration of exposure (Days)	Control group	Group- I	Group- II
7	21.56±0.19	15.43±0.21	13.48±0.24
14	22.05±0.18	12.32±0.12	10.13±0.24
21	22.21±0.20	10.09±0.31	7.14±0.28
F. value	0.039**	37.58*	42.38*

** = Non Significant ; * = Significant at 5% level of F test (p<0.05)



Graph showing decline in the glycogen levels of liver tissue and gill tissue with increase in exposure durations respectively (X axis – duration of exposure, Y axis- concentrations in tissues in mg/g).

Liver Tissues (Table 1)

In the control group, hepatic glycogen levels were recorded at 21.56 ± 0.19 mg/g on day 7, 22.05 ± 0.18 mg/g on day 14, and 22.21 ± 0.20 mg/g on day 21, indicating no statistically significant variation over time ($F = 0.039$, not significant). In Group-I, exposed to a sublethal concentration of cadmium, a steady reduction was noted with values of 15.43 ± 0.21 mg/g at day 7, 12.32 ± 0.12 mg/g at day 14, and 10.09 ± 0.31 mg/g at day 21 ($F = 37.58$, $p < 0.05$). A more marked depletion occurred in Group-II, where glycogen concentrations declined from 13.48 ± 0.24 mg/g on day 7 to 10.13 ± 0.24 mg/g on day 14, and further to 7.14 ± 0.28 mg/g by day 21 ($F = 42.38$, $p < 0.05$).

Gill Tissues (Table 2)

In the gill tissues of the control group, glycogen content remained relatively consistent, measured at 7.23 ± 0.26 mg/g on day 7, 7.28 ± 0.21 mg/g on day 14, and 7.32 ± 0.31 mg/g on day 21 ($F = 0.031$, not significant). Conversely, Group-I exhibited a gradual reduction over time, with levels decreasing to 5.86 ± 0.21 mg/g on day 7, 4.98 ± 0.31 mg/g on day 14, and 4.05 ± 0.12 mg/g on day 21 ($F = 29.17$, $p < 0.05$). Group-II demonstrated an even steeper decline, with glycogen levels falling from 4.54 ± 0.14 mg/g at day 7 to 3.59 ± 0.25 mg/g at day 14, and reaching 3.02 ± 0.31 mg/g by day 21 ($F = 33.16$, $p < 0.05$).

The study revealed a significant, time-dependent reduction in total glycogen levels in both liver and gill tissues of *Channa punctatus* following exposure to sublethal concentrations of cadmium. According to Sujata (2015), the reproductive tissues of *Channa punctatus* showed a significant drop in both glycogen and protein content following a 21-day exposure to sublethal concentrations of cadmium. In present study the decline was more pronounced at higher concentrations (Group-II) and longer exposure durations (21 days). In contrast, the control group showed stable glycogen levels throughout the experimental period, indicating that glycogen exposure disrupts glycogen metabolism in a dose- and duration-dependent manner. Supporting these findings, Suryakant et al. (2021) observed a reduction in tissue glycogen levels in *Tilapia mossambica* when exposed to methyl parathion. Similarly, in 2021 Bharti and Rasool reported decrease in glycogen content accompanied by a rise in blood glucose levels in *Labeo rohita* subjected to cypermethrin-induced stress. Previous studies in aquatic toxicology have highlighted that cadmium distribution within fish is tissue-specific and strongly influenced by the route of exposure—whether through waterborne exposure or dietary intake (Guinot et al., 2012; Nasser et al., 2015).

CONCLUSION

Cadmium chloride, a hazardous heavy metal released into water bodies from agricultural and industrial sources, presents serious environmental threats. This study examined the impact of different cadmium chloride concentrations on glycogen levels in *Channa punctatus*. Glycogen serves as a readily available energy reserve that is broken down into glucose through glycogenolysis during periods of stress, such as exposure to environmental pollutants. A concentration-dependent reduction in glycogen levels was observed in both liver and gill tissues, indicating metabolic disruption. These results highlight glycogen content as a potential biomarker for cadmium-induced stress in fish. Considering the dietary importance of fish, understanding heavy metal impacts on aquatic biochemistry is crucial for environmental and public health.

ACKNOWLEDGEMENT

The authors sincerely appreciate the financial support from the University Grants Commission, New Delhi. They also extend their gratitude to P.G. Department of Zoology, Patliputra University, Patna and also Central Research Laboratory, Patna Women's College for offering essential laboratory and infrastructural facilities.

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Cite this Article

Diksha Bhanu, Prof. Preety Sinha, “Cadmium Chloride Induced Changes in Glycogen Content of Hepatic and Gill Tissues of *Channa Punctatus*”, *International Journal of Scientific Research in Modern Science and Technology (IJSRMST)*, ISSN: 2583-7605 (Online), Volume 4, Issue3, pp. 33-38, March 2025.

Journal URL: <https://ijrmst.com/>

DOI: <https://doi.org/10.59828/ijrmst.v4i3.309>.



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