Chromium-Induced Oxidative Stress: Accumulation and Catalase Response in *Clarias gariepinus* (*Arila*)

Innocent Izuchukwu Ujah
Department of Applied Biochemistry, Faculty of Applied Natural Sciences, Enugu State University of Science & Technology

* Corresponding Author: **Innocent Izuchukwu Ujah**

Article Info

P-ISSN: 3051-3405
E-ISSN: 3051-3413
Volume: 01
Issue: 02
September – October 2025
Received: 19-07-2025
Accepted: 21-08-2025
Published: 16-09-2025
Page No: 45-48

Abstract
Heavy metals such as chromium are significant aquatic pollutants due to their bioaccumulation and potential to disrupt antioxidant defense systems in fish. Catalase, a key antioxidant enzyme, protects cells from oxidative damage by decomposing hydrogen peroxide. This study evaluated chromium accumulation in *Clarias gariepinus* and its effect on catalase activity. *Clarias gariepinus* were exposed to four different concentrations of chromium (1, 2, 3, and 4 mg/L) for seven days. Chromium accumulation in fish tissues was quantified using Atomic Absorption Spectrophotometry (AAS), while catalase activity was measured using spectrophotometer. Chromium accumulation increased proportionally with exposure, from 0.059 mg/kg at 1 mg/L to 0.149 mg/kg at 4 mg/L. Catalase activity also rose with chromium exposure, reaching 0.906 µmol/min at 2 mg/L and 0.853 µmol/min at 3 mg/L. However, at the highest exposure (4 mg/L; 0.149 mg/kg accumulated), catalase activity dropped sharply to 0.097 µmol/min. The study demonstrates that chromium accumulation in *C. gariepinus* stimulates catalase activity at low to moderate concentrations as part of the oxidative stress defense mechanism. However, excessive chromium exposure suppresses catalase activity, impairing the fish's antioxidant response. These findings highlight the ecological risks of chromium contamination in freshwater ecosystems and underline the importance of monitoring heavy metal pollution to safeguard aquatic life.

DOI: https://doi.org/10.54660/IJABRN.2025.1.2.45-48

Keywords: Chromium, Accumulation, *Clarias Gariepinus*, Heavy Metals, Catalase Activity

1. Introduction

Human activities such as improper waste disposal, fuel combustion, and mining have contributed significantly to environmental contamination (Ukaogo *et al*., 2020) [38]. These activities release heavy metals that adversely affect both aquatic ecosystems and human health (Dixit *et al*., 2015) [9]. Furthermore, rapid industrialization and intensive agricultural practices, including the extensive use of fertilizers, pesticides, insecticides, and herbicides, have escalated water pollution worldwide (Sharma *et al*., 2019) [29]. Toxic heavy metals and other pollutants are often found in industrial waste, agricultural fertilizers, and pesticides (Soliman & Moustafa, 2020) [31].

Heavy metals are metallic elements with relatively high densities compared to water (Gill, 2014) [12]. Their toxic effects vary depending on factors such as species, age, sex, concentration, exposure route, and individual physiological conditions (Aslam & Yousafzai, 2017) [2]. Of particular concern are arsenic, lead, mercury, chromium, and cadmium, which pose severe health risks even at low concentrations (Leyssens *et al*., 2017; Bhat *et al*., 2019) [21, 4]. Heavy metals disrupt cellular organelles and enzyme activities involved in detoxification and tissue repair (Wang *et al*., 2001) [39]. They can damage DNA and nuclear proteins, potentially leading to carcinogenesis or apoptosis (Beyersmann *et al*., 2008) [3]. Globally, heavy metals are among the most dangerous pollutants due to their persistence, toxicity, and tendency to bioaccumulate and biomagnify through the food chain (Majed *et al*., 2016) [25].

Chromium serves as an essential micronutrient in humans but becomes toxic when present in excess (Mehri, 2020) [26]. Dietary sources include brewer's yeast, wheat germ, and kidney, with an average daily intake of about 1 mg (Al-Fartusie & Mohssan, 2017) [1]. Industrially, chromium is widely used to harden steel, manufacture stainless steel, and produce various alloys (Dewangan *et al*., 2015) [8]. It has applications in chrome plating for vehicle parts, industrial catalysts, and pigments (Kavak *et al*., 2017) [18]. Rubies owe their red color to chromium, while chromium-treated glass exhibits an emerald-green hue (Lunk, 2015) [23].

Oxidative stress is another concern linked to heavy metal exposure. It occurs when the generation of reactive oxygen species (ROS) exceeds the body's antioxidant defenses. Antioxidants are compounds that prevent oxidation by neutralizing free radicals, thereby protecting cells from oxidative damage (Bhattacharya, 2015; Santos-Sánchez *et al*., 2019) [5, 28]. These may be endogenous, such as catalase and superoxide dismutase, or dietary, such as vitamins C and E (Salehi *et al*., 2018) [27]. Antioxidants may also be synthesized industrially or occur naturally in foods and tissues (Sun *et al*., 2021) [32].

Catalase is a key antioxidant enzyme found in most oxygen-exposed organisms. It breaks down hydrogen peroxide into water and oxygen, preventing oxidative damage (Chelikani, 2004; Lobo *et al*., 2010) [22]. Catalase exhibits one of the highest turnover rates among enzymes; a single molecule can convert millions of hydrogen peroxide molecules per second (Tehrani and Moosavi-Movahedi, 2018) [33]. Catalase is a tetrameric enzyme with four subunits, each containing iron-rich heme groups, It functions best at neutral pH in humans (Mahomoodally and MA-L, 2022; Ifeanyi, 2018; Kamel and Najmaddin, 2019) [15-16, 24].

The present study investigates the exposure of fish to chromium and its consequent effects on catalase activity.

2. Methods

2.1. Exposure of Fish Samples to Chromium and Enzyme Assay

Fingerlings of similar age and length were acclimatized under laboratory conditions for seven days prior to experimentation. During this period, they were fed with 0.51 mm Copen feed, which was withdrawn 12 hours before the bioassay. After acclimatization, four groups of fingerlings were exposed to different concentrations of chromium salts, while a control group was maintained in metal-free media. Water in the aquaria was renewed every two days to prevent waste and food accumulation. On the seventh day of exposure, two fish were randomly sampled from each group for heavy metal analysis and peroxidase assay.

2.2. Preparation of Chromium Stock Solution

A stock solution was prepared by dissolving 0.145 g of potassium dichromate ($K_2Cr_2O_7$) in 10 ml of distilled water. The mixture was shaken thoroughly, and deionized water was added to make up a final volume of 1 L.

2.3. Principle of Atomic Absorption Spectrophotometry (AAS)

Atomic Absorption Spectrophotometry (AAS) is an analytical technique used to determine the concentration of metals in a sample. It can measure over 70 elements and is based on Beer-Lambert's law, where atoms absorb light of specific wavelengths. In the atomizer, electrons are excited to higher energy levels by absorbing energy (light). Each element has a unique wavelength corresponding to its electron transitions, making the method highly specific. The absorbed energy is proportional to the concentration of the element present.

2.4. Determination of Chromium Concentration

Ground fish samples were analyzed for chromium using an AAS (Spectra AA, model 240 FS) set at the appropriate wavelength and detection limit. Diluted extracts of the samples were aspirated into the instrument. The chromium-specific hollow cathode lamp was fitted, and absorbance values were recorded directly from the display.

2.5. Catalase Assay

Fish tissue was homogenized in a mortar and transferred into a beaker. Thirty milliliters of 0.1 M phosphate buffer was added, followed by 40 ml of 3% hydrogen peroxide (H_2O_2). The mixture was agitated to ensure uniformity, filtered, and the clear filtrate was used for catalase activity determination.

2.6. Calculation of Catalase Activity

$$\text{Activity} = \text{Extinction coefficient} \times \text{sample volume}$$

$$\text{Change in absorbance per minute} \times \text{Total reaction volume}$$

Where:

Total reaction volume: 3 ml
Sample volume: 0.1 ml
Extinction coefficient: 40 m^{-1}
Control value: 0.093

3. Results

Table 3.1: Concentration of Chromium Accumulated in the Fish

Concentration of Chromium Introduced (mg/L)	Concentration of Chromium Accumulated (mg/Kg)
1	0.059±0.003
2	0.067±0.002
3	0.122±0.002
4	0.149 ±0.001

The result shows that the quantity accumulated increased with the increase in the concentration of chromium introduced to the Fish from 1 to 4mg/L of the chromium.

Table 3.2: Concentration of Chromium introduced and the catalase activity

Concentration of Chromium Introduced (mg/L)	Catalase Activity (µmol/min)
1	0.415±0.005
2	0.906±0.004
3	0.853±0.513
4	0.097±0.005

The results show an increase in the catalase activity with increase in concentrations except for the 4mg/L concentration.

Table 3.3: Chromium accumulation and the corresponding catalase activity

Concentration of Chromium Accumulated (mg/Kg)	Catalase Activity (µmol/min)
0.059±0.003	0.415±0.005
0.067±0.002	0.906±0.004
0.122±0.002	0.853±0.513
0.149 ±0.001	0.097±0.005

The results indicate that as the accumulation increased, there was a corresponding increase in the catalase activity except for the 0.149 mg/kg where the catalase activity decreased.

4. Discussion

This study investigated the effect of chromium accumulation on catalase activity in *Clarias gariepinus*. Catalase (CAT) is a key antioxidant enzyme that protects cells by eliminating reactive oxygen species (ROS) (Yang *et al*., 2015) [40]. Aquatic ecosystems are the ultimate sinks for heavy metals released from natural and anthropogenic sources (Dixit *et al*., 2015) [9]. Fish organs such as the liver, kidney, and muscle contain abundant antioxidant enzymes, including CAT, superoxide dismutase (SOD), glutathione peroxidase (GPX), glutathione S-transferase (GST), and glutathione reductase (GR), which safeguard against oxidative stress (Kanak *et al*., 2015) [17].

Chromium is an essential trace mineral found in foods like broccoli, potatoes, whole grains, lean meats, nuts, seafood, brewer's yeast, and spices (Konikowska & Mandecka, 2018) [20]. In contrast, refined grains, processed foods, and high-sugar foods are low in chromium and may even increase its loss from the body (Guleria, 2021) [13]. Chromium plays a vital role in insulin function and glucose metabolism, and individuals with poor diets or certain health conditions are at risk of deficiency (Genchi *et al*., 2021) [11]. Chromium accumulation rose with increasing concentration after 7 days of exposure. Fish exposed to 1 mg/L chromium showed an accumulation of 0.059 mg/kg with a corresponding catalase activity of 0.415 µmol/min. At higher exposures (2–3 mg/L), both accumulation and catalase activity increased significantly. However, at 4 mg/L, although chromium accumulation reached 0.149 mg/kg, catalase activity dropped sharply, suggesting enzyme inhibition or oxidative damage at high chromium levels. Elevated catalase activity at moderate concentrations reflects increased ROS production and a compensatory antioxidant response (Ho *et al*., 2004; Shilpi *et al*., 2015) [14, 30]. The lower activity in control fish agrees with Tyokumbour *et al*. (2014), who noted minimal enzyme response under low heavy metal burden.

Similar findings have been reported in fish exposed to other metals such as mercury, nickel, cadmium, and zinc (Bozcaarmtlu *et al*., 2017), as well as in earthworms, where bioaccumulation affected protein, carbohydrate, and glycogen levels (Ujah *et al*., 2017) [37]. Chromium toxicity also impacts growth, survival, physiology, and DNA integrity in fish, as shown in Chinook salmon exposed for prolonged periods (Farag *et al*., 2006) [10]. Thus, while moderate chromium exposure stimulates catalase activity, excessive accumulation impairs antioxidant defenses and may cause broader physiological damage (Zhou *et al*., 2020; Koner *et al*., 2021) [41, 19].

5. Conclusion

The findings revealed that chromium accumulation in the fish corresponded with elevated catalase activity.

6. References

1. Al-Fartusie FS, Mohssan SN. Essential trace elements and their vital roles in human body. Indian J Adv Chem Sci. 2017;5(3):127-36.
2. Aslam S, Yousafzai AM. Chromium toxicity in fish: a review article. J Entomol Zool Stud. 2017;5(3):1483-8.
3. Beyersmann D, Hartwig A. Carcinogenic metal compounds: recent insight into molecular and cellular mechanisms. Arch Toxicol. 2008;82(8):493-512.
4. Bhat SA, Hassan T, Majid S. Heavy metal toxicity and their harmful effects on living organisms–a review. Int J Med Sci Diagn Res. 2019;3(1):106-22.
5. Bhattacharya S. Reactive oxygen species and cellular defense system. In: Free radicals in human health and disease. New Delhi: Springer; 2015. p. 17-29.
6. Bozcaarmutlu A, Arinc E. Effect of mercury, cadmium, nickel, chromium and zinc on kinetic properties of NADPH-cytochrome P450 reductase purified from leaping mullet (Liza saliens). Toxicol *In Vitro*. 2007;21(3):408-16.
7. Chelekani P, Fita I, Loewen PC. Diversity of structures and properties among catalases. Cell Mol Life Sci. 2004;61(2):192-208.
8. Dewangan AK, Patel AD, Bhadania AG. Stainless steel for dairy and food industry: a review. J Mater Sci Eng. 2015;4(5):1-4.
9. Dixit R, Wasiullah, Malaviya D, Pandiyan K, Singh UB, Sahu A, *et al*. Bioremediation of heavy metals from soil and aquatic environment: an overview of principles and criteria of fundamental processes. Sustainability. 2015;7(2):2189-212.
10. Farag AM, May T, Marty GD, Easton M, Harper DD, Little EE, *et al*. The effect of chronic chromium exposure on the health of Chinook salmon (Oncorhynchus tshawytscha). Aquat Toxicol. 2006;76(3-4):246-57.
11. Genchi G, Lauria G, Catalano A, Carocci A, Sinicropi MS. The double face of metals: the intriguing case of chromium. Appl Sci. 2021;11(2):638. doi:10.3390/app11020638.
12. Gill M. Heavy metal stress in plants: a review. Int J Adv Res. 2014;2(6):1043-55.
13. Guleria S. Food, obesity, and noncommunicable diseases. J Postgrad Med Educ Res. 2021;55(1):8-11.
14. Ho YS, Xiong Y, Ma W, Spector A, Ho DS. Mice lacking catalase develop normally but show differential sensitivity to oxidant tissue injury. J Biol Chem. 2004;279(31):32804-12.
15. Ifeanyi OE. A review on free radicals and antioxidants. Int J Curr Res Med Sci. 2018;4(2):123-33.
16. Kamel FH, Najmaddin C. A novel extraction of plant enzyme has more activity compare to traditional techniques. J Res Lepidoptera. 2019;50(2):203-8.
17. Kanak EG, Dogan Z, Eroglu A, Atli G, Canli M. Effects of fish size on the response of antioxidant systems of Oreochromis niloticus following metal exposures. Fish Physiol Biochem. 2014;40(4):1083-91.

18. Kavak O, Aydin F, Fidan C, Aydin I. Chromium and thallium in Turkish asphaltite sample. In: International Multidisciplinary Scientific GeoConference: SGEM; 2017; Vienna, Austria. Sofia: SGEM; 2017. p. 797-802.
19. Koner D, Banerjee B, Kumari A, Lanong AS, Snaitang R, Saha N. Molecular characterization of superoxide dismutase and catalase genes, and the induction of antioxidant genes under the zinc oxide nanoparticle-induced oxidative stress in air-breathing magur catfish (Clarias magur). Fish Physiol Biochem. 2021;47(6):1909-32.
20. Konikowska K, Mandecka A. Trace elements in human nutrition. In: Recent advances in trace elements. Chichester: Wiley; 2018. p. 339-72.
21. Leyssens L, Vinck B, Van Der Straeten C, Wuyts F, Maes L. Cobalt toxicity in humans—a review of the potential sources and systemic health effects. Toxicology. 2017;387:43-56.
22. Lobo V, Patil A, Phatak A, Chandra N. Free radicals, antioxidants and functional foods: impact on human health. Pharmacogn Rev. 2010;4(8):118-26.
23. Lunk HJ. Discovery, properties and applications of chromium and its compounds. ChemTexts. 2015;1(1):6. doi:10.1007/s40828-015-0006-z.
24. Mahomoodally MF, MA-L ER. Catalase. In: Antioxidants effects in health. Amsterdam: Elsevier; 2022. p. 81-90.
25. Majed N, Real MIH, Akter M, Azam HM. Food adulteration and bio-magnification of environmental contaminants: a comprehensive risk framework for Bangladesh. Front Environ Sci. 2016;4:34. doi:10.3389/fenvs.2016.00034.
26. Mehri A. Trace elements in human nutrition (II)—an update. Int J Prev Med. 2020;11:2. doi:10.4103/ijpvm.IJPVM_48_19.
27. Salehi B, Martorell M, Arbiser JL, Sureda A, Martins N, Maurya PK, *et al*. Antioxidants: positive or negative actors? Biomolecules. 2018;8(4):124. doi:10.3390/biom8040124.
28. Santos-Sánchez NF, Salas-Coronado R, Villanueva-Cañongo C, Hernández-Carlos B. Antioxidant compounds and their antioxidant mechanism. In: Antioxidants. Basel: MDPI; 2019. p. 1-29.
29. Sharma A, Kumar V, Shahzad B, Tanveer M, Sidhu GPS, Handa N, *et al*. Worldwide pesticide usage and its impacts on ecosystem. SN Appl Sci. 2019;1(11):1446. doi:10.1007/s42452-019-1485-1.
30. Shilpi G, Shilpi S, Sunita S. Tolerance against heavy metal toxicity in cyanobacteria: role of antioxidant defense system. Int J Pharm Pharm Sci. 2015;7(2):9-14.
31. Soliman NK, Moustafa AF. Industrial solid waste for heavy metals adsorption features and challenges; a review. J Mater Res Technol. 2020;9(5):10235-53.
32. Sun N, Li M, Liu G, Jing M, He F, Cao Z, *et al*. Toxic mechanism of pyrene to catalase and protective effects of vitamin C: studies at the molecular and cell levels. Int J Biol Macromol. 2021;171:225-33.
33. Tehrani HS, Moosavi-Movahedi AA. Catalase and its mysteries. Prog Biophys Mol Biol. 2018;140:5-12.
34. Ujah II, Nsude CA, Okpako IO, Ugwu CM. Induction of catalase response in Clarias exposed to graded concentration of iron as fish's body defence mechanism against environmental pollutants. Acta Sci Microbiol. 2019;2(10):49-52.
35. Ujah II, Chikezie NJ. Arsenic accumulation in Clarias gariepinus and effects on catalase activity. Acta Sci Microbiol. 2019;2(10):155-8.
36. Ujah II, Nsude CA, Odo GE. An insight into effects of pollution in aquatic environment through effects of nickel accumulation on catalase activity of Clarias gariepinus. Ann Clin Toxicol. 2018;1(1):1007.
37. Ujah II, Onwurah INE, Ubani SC, Okeke DO, Okpashi VE. Assessing bioaccumulation in earthworms. J Environ Sci Public Health. 2017;1(4):224-7.
38. Ukaogo PO, Ewuzie U, Onwuka CV. Environmental pollution: causes, effects, and the remedies. In: Microorganisms for sustainable environment and health. Amsterdam: Elsevier; 2020. p. 419-29.
39. Wang S, Shi X. Molecular mechanisms of metal toxicity and carcinogenesis. Mol Cell Biochem. 2001;222(1-2):3-9.
40. Yang HT, Yang MC, Sun JJ, Guo F, Lan JF, Wang XW, *et al*. Catalase eliminates reactive oxygen species and influences the intestinal microbiota of shrimp. Fish Shellfish Immunol. 2015;47(1):63-73.
41. Zhou Q, Yang N, Li Y, Ren B, Ding X, Bian H, *et al*. Total concentrations and sources of heavy metal pollution in global river and lake water bodies from 1972 to 2017. Glob Ecol Conserv. 2020;22:e00925. doi:10.1016/j.gecco.2020.e00925.