



Lead Contamination in Water and Fish from Tanda Dam, Kohat: Environmental and Public Health Concerns

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Abstract

HEAVY metal contamination in aquatic ecosystems poses significant ecological and public health concerns. This study investigated lead (Pb) concentrations in water and muscle tissues of four cyprinid species—*Hypophthalmichthys molitrix*, *Cirrhinus mrigala*, *Cyprinus carpio*, and *Labeo rohita*—from Tanda Dam, Kohat, Pakistan, a major source of irrigation and fish production. Water and fish samples were collected from seven sites and analyzed using Flame Atomic Absorption Spectroscopy. Pb concentrations in water ranged from 0.046 to 1.03 mg/L, while fish muscle samples contained 0.258 to 0.378 mg/kg. Statistical analysis (ANOVA, $p < 0.05$) revealed significant spatial variation among sites and interspecific differences in fish accumulation. *H. molitrix* showed the highest Pb concentration, whereas *L. rohita* had the lowest. Although measured concentrations were within international safety thresholds, localized hotspots in the south and west suggest potential contamination risks. This study provides a systematic assessment of Pb contamination in Tanda Dam, highlighting the need for continuous monitoring of heavy metals in both aquatic environments and fish species consumed by local communities. The findings underscore the importance of implementing proper waste management and effluent treatment to prevent future ecological degradation and safeguard public health.

Keywords: Heavy metals, Lead, Aquatic toxicology, Civic pollution, Public health.

Introduction

Aquaculture has made a significant contribution to global fish production, reaching 223.2 million tons in 2022 [1]. The use of fisheries resources under environmentally sustainable conditions has enhanced the significance of the fisheries and aquaculture sector [2]. The combined production of capture and aquaculture fisheries contributes to food security, livelihoods, employment, and national economies [3,4], as fish are widely consumed and provide essential nutrients, including minerals, vitamins, amino acids, and polyunsaturated fatty acids [5–7]. In addition to their antiarrhythmic and cardioprotective roles [8,9], fish oils benefit pregnant women and neonates by reducing complications

[10,11]. They are also applied in heart transplant surgery to minimize rejection, regulate blood pressure, and prevent kidney damage [12,13].

One of the key environmental issues is the release of toxic metals into the aquatic environment. Humans may be exposed to these metals through the food chain, posing a significant threat to human health. Among these toxic metals, lead (Pb) is particularly harmful and biologically non-essential [14]. Various anthropogenic activities, such as battery production, paint manufacturing, and leaded gasoline processing [15], along with cement mining and smelting, are significant sources of pollution [16]. Fish accumulate these non-essential metals primarily through contaminated water, food, and

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sediments [17]. This accumulation occurs in fish tissues [18] and depends on both the concentration and duration of metal exposure, as well as species-specific traits (e.g., ecological needs, sex) and environmental conditions such as water pH, salinity, temperature, and hardness [19]. Fish occupy higher trophic levels in the food chain in most aquatic environments and, therefore, are most susceptible to Pb toxicity [20].

The buildup of toxic heavy metals in aquatic organisms can lead to various diseases and physiological disorders [19]. Exposure to Pb in animals leads to a decline in physiological, biochemical, and behavioral functions. This exposure has detrimental effects on various systems, including the central and peripheral nervous systems, the hematopoietic system, and the cardiovascular system, as well as vital organs such as the kidney and liver [21]. Additionally, the accumulation of lead can cause hypocalcemia and negatively impact immune response, as exposure to this metal is directly linked to changes in the immune system of fish [22]. Lead is also a potent immunotoxin, disrupting immune function in animals [23]. Therefore, Pb exposure could be lethal to aquatic fauna besides the bioaccumulation dilemma [19].

The study area, Tanda Dam, is located in Kohat District, Khyber Pakhtunkhwa, Pakistan [24]. It provides irrigation water to different regions of Kohat and contributes to a substantial annual fish production [25]. We hypothesize that Pb accumulation in aquatic species may pose serious health risks to humans through the consumption of contaminated fish and the irrigation of crops. Therefore, this study aimed to quantify Pb accumulation in various cyprinid fish species and water from Tanda Dam, Kohat.

Material and Methods

Study area

Tanda Dam is a small reservoir located in Kohat District, Khyber Pakhtunkhwa, Pakistan (33°35'13" N, 71°26'29" E) at an elevation of 489 m above sea level (Figure 1). The dam receives its primary inflow from the Kohat Toi River, which originates in the hills of the Kohat and Hangu regions. Its catchment area includes predominantly semi-arid hills and valleys, with seasonal streams and monsoon rainfall contributing to runoff. No major industrial zones lie within the upstream catchment or aquifers feeding Tanda Dam. The surrounding landscape is largely rural and agricultural with dispersed settlements. Consequently, anthropogenic influences on the inflows are limited to small-scale agriculture and domestic effluents rather than industrial discharges. The study area is characterized by mountainous topography, with mean maximum and minimum temperatures of approximately 40 °C and 6 °C during summer and winter, respectively. Rainfall ranges

between 24 and 321 mm per month, with the highest precipitation occurring in July and August. June represents the hottest month of the year, whereas January is the coldest, while relative humidity peaks in July-August during summer and in December during winter [26].

Water sample collection

For water sample collection, pre-rinsed sterile plastic bottles were used, each rinsed three times before sampling. Water samples were obtained in triplicate by immersing the bottles at least 10 cm below the surface, during three consecutive months from December 2022 to February 2023. Samples were collected from seven designated locations representing different parts of the reservoir: east, west, north, south, center, upstream, and downstream. Collected water was stored in PET (polyethylene terephthalate) bottles that had been pre-acid-washed with 10% concentrated nitric acid (HNO₃, v/v) and thoroughly rinsed with distilled deionized water. A total volume of 500 mL per sample was taken, preserved in ice boxes, and immediately transported to the laboratory for further analysis [27].

Fish sample collection

Fish sampled from the dam mainly comprised cyprinid species; therefore, *Hypophthalmichthys molitrix* (Silver Carp), *Labeo rohita* (Rohu), *Cirrhinus mrigala* (Mori), and *Cyprinus carpio* (Common Carp) were taken for analysis. Fish were collected using gill nets and cast nets with the assistance of local fishermen. All possible efforts were made to reduce the risk of injury and stress during handling and capture. Morphometric and meristic data were recorded for each specimen. The morphological measurements of the fish samples were obtained using a digital balance and a measuring scale, i.e., the body weight of each specimen was determined using a digital balance, and total length, standard length, and girth were measured using a measuring scale. The results, including mean values with their standard errors of the mean (SE), are presented in Table 1. The specimens were transported in sterile polyethylene bags with fresh water to the Fisheries and Aquaculture Lab Department of Zoology, Kohat University of Science and Technology (KUST), Khyber Pakhtunkhwa, Pakistan. The samples were kept at -20°C until further analysis.

Dissection of specimens and isolation of samples

In the laboratory, the fish were thoroughly cleaned with deionized water. Fish were anesthetized with MS-222 (100-150 mg/L) until they reached a surgical level of anesthesia, characterized by complete loss of response to stimuli [28,29]. Safe conditions for fish dissection and tissue collection were ensured. Dissection was performed with the

help of sterilized surgical scissors. The operculum was gently removed first, followed by fin removal and ventral dissection for muscle isolation. The tissues were then air-dried to remove any residual moisture. Approximately 200 g of muscle tissue per specimen was homogenized and stored in labelled polyethylene bags at -20°C to prevent deterioration until further analysis [25].

Dehydration and digestion of muscle tissue samples

Muscle tissue samples were dehydrated in a water bath after being placed in 250 mL conical flasks. Samples were incubated at 90°C for 45 minutes and subsequently maintained at 65°C for 48 hours to ensure complete dehydration. The digestion protocol was followed with slight modifications [30]. Briefly, one gram of muscle tissue was placed in a 100 mL digestion tube, followed by the addition of 25 mL of concentrated nitric acid (HNO_3). The tube was then kept on a hot plate in a fume hood. Samples were first heated to 90°C for 45 minutes and then increased to 150°C for eight hours. After cooling to room temperature, 15 mL of perchloric acid was added, followed by gentle boiling of the solution for one hour. The sample started boiling, and a clear colorless solution was obtained. After digestion, the samples were diluted with deionized water filtered with Whatman No. 40 filter papers (Sigma-Aldrich USA) and transferred into a 15 mL Falcon tube. The Falcon tubes were labeled and stored at -4°C before atomic absorption spectroscopic analysis. To analyze the water sample, 450 mL of the water sample was first poured into a flask. Next, 6 mL of nitric acid was added, and the mixture was heated to 100°C . After heating, H_2O_2 was introduced, and the solution was then filtered before proceeding with Flame Atomic Absorption Spectroscopy (FAAS) [31].

Atomic absorption spectroscopy analysis

After sample digestion, Pb concentration was determined using FAAS (Model Z-2000 Hitachi). The instrument was calibrated with a standard solution, and a calibration curve was derived using various standard concentrations of Pb. A lamp current of 7.5 mA and a burner height of 7.5 mm were employed. An air-acetylene flame was used as the excitation source, and the instrument was set to a wavelength of 247.6 nm. Pb concentrations were estimated through correlation of the sample absorbance data with the calibration curve derived from known Pb standard solutions.

Quality control and quality assurance (QC/QA)

Quality control and quality assurance (QC/QA) play a crucial role in the lab's operations. To prevent potential contamination, all flasks, tubes, and bottles were pre-rinsed with a 6% HCl solution for 24 hours and then thoroughly cleaned three times with deionized water. Additionally, during the FAAS procedure, we conducted analyses of reagent blanks

and certified reference materials to ensure analytical accuracy and precision.

Statistical analysis

Before statistical analysis, variance, homogeneity, and normality were assessed using Bartlett's test and the Shapiro-Wilk test to measure the spread of values relative to the mean. The data were analyzed statistically with GraphPad Prism (version 10) and MS Excel. Data are reported as mean $\pm$ standard deviation (SD), with one-way ANOVA and subsequent Tukey test was performed to detect differences among means. Significance was set at $p < 0.05$.

Results

Pb concentration in fish bodies

Pb concentrations (mg/kg) in the muscle tissues of four cyprinid species were measured. Six samples of each species were analyzed. *H. molitrix* showed the highest Pb concentration (0.378 ± 0.037 mg/kg), whereas *L. rohita* had the lowest (0.258 ± 0.036 mg/kg). Analysis showed that all species differed significantly ($p < 0.05$), as determined by one-way ANOVA followed by Tukey's test ($n = 6$, $F_{3, 20} = 14.51$, Figure 2). Low variance indicates consistent accumulation within species. However, *H. molitrix*, *C. mrigala*, and *L. rohita* exhibited higher variances (0.0014, 0.0013, and 0.0013, respectively) compared to *C. carpio* (0.0005). This suggests that Pb accumulation was more variable in *H. molitrix*, *C. mrigala*, and *L. rohita* compared to *C. carpio*, indicating less uniform exposure among individuals. These variations in variance may indicate differences in individual exposure size or bioaccumulation efficiency.

Pb concentration in water samples

The water samples collected from seven sites were analyzed to detect Pb concentrations (mg/L). The highest Pb concentration was recorded at the southern site (1.01 ± 0.026 mg/L), followed by West (0.44 ± 0.023 mg/L), while the lowest concentrations were observed at the upstream and downstream sites (0.04 ± 0.02 and 0.04 ± 0.01 mg/L, respectively). Analysis using one-way ANOVA revealed significant differences among all water samples ($p < 0.05$), which was further confirmed through Tukey's test ($n = 3$, $F_{6, 14} = 333.68$, Figure 3). The variance analysis in Pb concentration among water samples ranged from 0.0004 to 0.00163. Sampling points North and Downstream exhibited the lowest variance values (0.0004 to 0.00043), demonstrating highly consistent Pb concentrations. Conversely, the West Centre and Upstream samples showed the highest variance values (0.00160 to 0.00163), reflecting greater fluctuation in Pb concentrations across the replicates. These differences suggest variability in local contamination sources or water mixing dynamics at these locations.

Discussion

Among the major global concerns, environmental pollution persists as one of the most serious challenges of our time [32-34]. Heavy metals are among the most harmful contaminants due to their persistence, non-biodegradability, and toxic effects on both humans and aquatic organisms [35]. Among heavy metals, Pb has particularly hazardous effects on the aquatic ecosystem. Fish are more susceptible to heavy metal pollution and can accumulate these substances in their tissues, resulting in toxic effects that can become lethal with long-term exposure and bioaccumulation [36]. Through trophic pathways, these fish transfer heavy metals to humans, posing serious risks to health and well-being [37].

Since the study site serves as a major source of commercially and nutritionally important fish species, the local population relies heavily on cyprinids for their inexpensive and accessible supply of fish meat. The assessment of heavy metals, particularly Pb, in fish and water from Tanda Dam was therefore necessary to evaluate contamination risks and to promote the safety of water and aquatic food sources for community and agricultural purposes. Furthermore, Pb was chosen because of its significant ecotoxicological relevance. It is among the most toxic heavy metals, posing severe threats to ecosystem health [38].

Our findings revealed significant differences ($p < 0.05$) in Pb concentrations among the four cyprinid species, with *H. molitrix* showing the highest and *L. rohita* the lowest concentrations (Figure 2). Although all values remained within the permissible range (1 µg/g) established by UK food safety guidelines. These values contrast sharply with those reported for *C. carpio* in the River Kabul (53.3 µg/g) by Ahmad *et al.* [39], likely due to higher urban and industrial activity in that region. Likewise, the reported results of the current study were not aligned with those of Iqbal *et al.* [40], who reported that concentrations of heavy metals exceeded acceptable limits. Furthermore, previous literature, such as Ali and Khan [37], Pandey and Singh [41], Ahmad *et al.* [42], and Dwivedi *et al.* [43], has indicated varying concentrations of Pb in the muscle tissues of fish species. In comparison, our results indicated significantly lower concentrations of Pb than those reported in these studies, likely due to the low industrial activity and population density. The variations in the Pb concentration among different species may arise from a range of intrinsic and extrinsic factors, including species type, age, size, feeding habits, physiological regulation of heavy metals within the body, habitat type, and climatic conditions [44]. Currently, this dam bears the burden of sewage and agricultural runoff from the surrounding area. However, if the inflow of these wastes continues into the dam, it will pose a serious threat in the future.

Pb concentrations in water samples showed significant spatial variation among sampling sites ($p < 0.05$) (Figure 3). The concentration trend from highest to lowest was: South > West > North > Center > East > Upstream $\approx$ Downstream. In some sites, the Pb concentrations didn't align with the World Health Organization (WHO) guideline limit for Pb in drinking water, which is 0.01 mg/L [45]. The documented levels of other heavy metals in Tanda Dam's fish and water were consistent with the findings of the present research [25]. Similarly, Pb concentrations in the Shah Alam River, Peshawar, were higher in range compared to the current results [46]. The high levels can be attributed to Peshawar's dense population and industrial activity, which results in significant discharges of domestic wastewater and industrial effluents. The disposal of these sewage and effluents has been recognized as a key source of global heavy metals in surface waters [47]. Additionally, our results aligned with the reported Pb concentrations in Ghol Dam Karak by Azeem *et al.* [48]. Kohat and Karak are adjacent areas with Tanda Dam and Ghol Dam fed by streams and seasonal rains. Therefore, we can conclude that the similar results could stem from their comparable rock structures and water sources.

Pb concentrations in fish tissues and water that exceed permissible limits pose serious health risks to humans. Exposure to heavy metals can harm vital organs like the kidneys, liver, and brain, and also disrupt major systems such as the reproductive, metabolic, and endocrine systems [49]. Pb concentrations measured in the muscle of fish species and in the water of Tanda Dam complied with WHO standards, reflecting no significant health hazards. The study recorded very low concentrations of Pb, possibly originating from natural sources, such as rocks, as well as contributions from surface runoff, agricultural runoff, and wastewater disposal. These sources are recognized as major contributors to elevated heavy metal concentrations due to seasonal streams and anthropogenic activities in Tanda Dam, as indicated by elevated heavy metal contents at the north, south, and west sites, where seasonal streams and agricultural and sewage runoff enter the dam. Furthermore, the dam's role as a tourist attraction makes it susceptible to anthropogenic contributions of heavy metals. Although the current levels in the water do not exceed permissible standards, continuous disposal may pose future environmental concerns. Therefore, regular monitoring and proper treatment of waste effluent discharge are recommended to protect this dam from further chemical pollutants.

Conclusion

The contamination of Tanda Dam by heavy metals arises primarily from geological origins and human activities, notably agricultural effluents and sewage discharge. Significant spatial and

interspecific variations in Pb concentrations were observed, despite overall concentrations being within acceptable limits. To mitigate future risks, continuous monitoring of Pb in water and fish is essential, as ongoing waste disposal could elevate concentrations to unsafe levels. Furthermore, effluent treatment and proper waste management practices should be strictly enforced to protect aquatic environments and safeguard public health. Future research should examine seasonal variations in Pb accumulation and extend monitoring to other aquatic organisms to better understand food web dynamics and long-term ecological risks. In addition, dietary exposure assessments such as estimated daily intake (EDI) should be incorporated in future studies to better evaluate potential human health risks.

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Declaration of Conflict of Interest

No potential conflict of interest exists concerning the preparation or publication of this article.

Ethical approval

Samples were handled in accordance with the Quaid-I-Azam University's animal welfare committee in compliance with local legislation (BEC-FBS-QAU2021-72)

TABLE 1. Morphological measurements of family Cyprinidae fish specimens

No	Species	Mean Weight (g)± SE	Mean Standard Length (cm)± SE	Mean Total Length (cm)± SE	Mean Body Depth (cm)± SE
1	<i>Labeo rohita</i>	460±3.0	28.2±1.3	32.1±1.0	7.6±0.1
2	<i>Cirrhinus mrigala</i>	466±2.0	29.5±0.7	35±1.0	8.6±0.2
3	<i>Hypophthalmichthys molitrix</i>	458±2.0	27±1.1	32.5±1.0	8.8±0.2
4	<i>Cyprinus carpio</i>	445±4.0	26.3±2.0	28.3±0.7	7.8±0.1

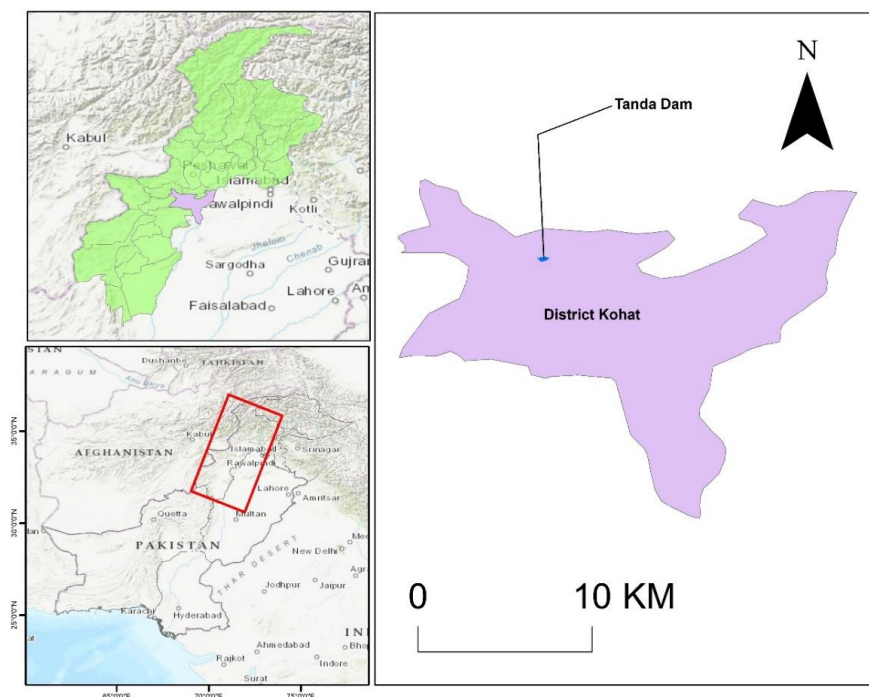


Fig.1. Map of the study area, Tanda Dam, Kohat, Khyber Pakhtunkhwa, Pakistan

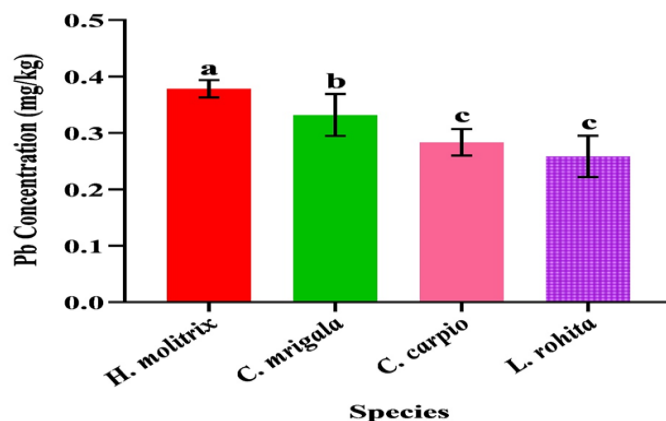


Fig.2. Pb Concentration in four family Cyprinidae species. Results are shown as mean $\pm$ SD and are significantly different ($p < 0.05$) using one-way ANOVA.

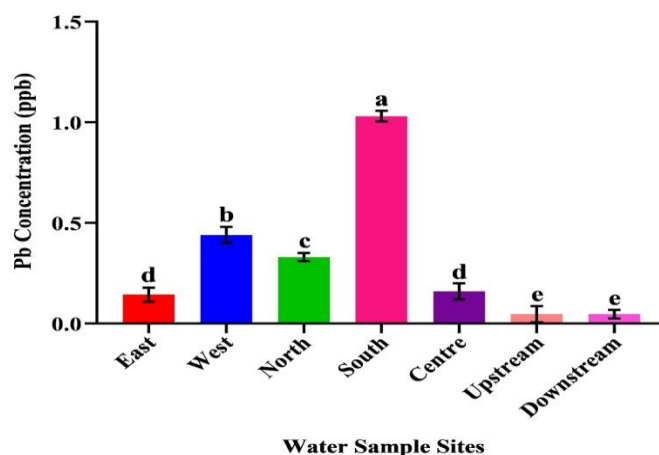


Fig.3. Pb Concentration in water samples from seven sites. Results are shown as mean $\pm$ SD and are significantly different ($p < 0.05$) using one-way ANOVA.

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