

BIOACCUMULATION OF TOXIC METALS IN SOME TISSUES OF CAT FISH (KHAGGA/ SINGHARA) (SILURIFORMES) IN DIFFERENT PONDS OF TALUKA TANGWANI KANDHKOT DISTRICT KASHMORE

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Abstract

This study investigated the bioaccumulation of trace and toxic metals in two catfish species Khagga and Singhara collected from three different aquaculture ponds in Taluka Tangwani, District Kashmore @ Kandhkot. A total of 36 samples, comprising 1 kg and 0.5 kg individuals of each species, were analyzed for metal concentrations in gill, muscle, skin, and lung tissues. Samples were dissected and digested using a tri-acid mixture (HNO₃:HCl:H₂O₂; 3:1:2) at 80 °C, followed by filtration and dilution. Metal analysis was conducted using ICP-OES at NECAC, University of Sindh, Jamshoro.

The results showed that concentrations of most metals were within FAO/WHO permissible limits, except for aluminum (Al), which exceeded recommended thresholds. In 1 kg Khagga, Al ranged from 1.36 mg/kg (gill/liver) to 2.25 mg/kg (muscle), while in 0.5 kg samples, it reached up to 2.7 mg/kg (muscle). In Singhara, the highest Al value was 2.1 mg/kg (muscle), and the lowest was 1.0 mg/kg (gill). Nickel (Ni) levels ranged from 0.0002–0.010 mg/kg, with higher concentrations in gills and skin, particularly in smaller fish. Lead (Pb) levels remained low, ranging between 0.0022–0.0139 mg/kg, with liver and gill tissues showing the highest accumulation. Strontium (Sr) was present in all tissues, with a maximum of 0.313 mg/kg found in the gills of 1 kg Khagga and notable accumulation in the skin and lungs of smaller fish.

The study concludes that Khagga generally exhibited higher metal accumulation than Singhara, and gill and skin tissues were more prone to metal uptake. Although most values were within safe limits, the elevated aluminum levels suggest potential environmental contamination and a need for regular monitoring of fish ponds in the region.

Keywords:

Trace and Toxic Metals; Bioaccumulation; ICP-OES; Catfish; Khagga; Singhara; Kashmore@Kandhkot.

1. Introduction

In the past decade, international consumption of fish has risen immensely owing to increased knowledge of the nutritional and therapeutic values of fish. Being a good source of easily assimilated proteins, essential fatty acids, vitamins, and minerals, fish is an important component of the human diet and is well known for its contribution to heart health as well as brain development (Hosseini et al., 2015), but this increasing consumption has also raised concerns for food safety, especially in terms of the bioaccumulation of toxic pollutants in the water ecosystem.

Fish take up contaminants from the environment through natural bioaccumulation, where metals and other compounds are absorbed from water, sediment, and foodstuffs (Mahboob et al., 2014; Morina et al., 2016). Such toxins, particularly heavy metals, can accumulate in tissues over a prolonged period and find their way into the food chain, resulting in serious ecological and human health concerns (Sow et al., 2013). These pollutants, aluminum (Al), nickel (Ni), lead (Pb), and strontium (Sr) are most noteworthy. Although some of these elements are trace or are essential only in small quantities, their high concentrations are toxic, having associated them with cardiopulmonary impairment, liver and kidney failure, neurological disease, and death in severe situations (Maitlo, A. A. et al., 2023; Ahmad Kamal et al., 2011; Osman & Kloas, 2010). Contamination of freshwater sources, which have long been considered the pillar of human existence, is increasingly beset by anthropogenic stressors such as industrial effluent discharge, agricultural runoff, mining, and malpractices in waste disposal (Dahar, A. K. et al., 2021; Abiona et al., 2019; Chauhan et al., 2019).

Environmental stressors bring about persistent contaminants in water bodies, where sediments trap these and make them easily accessible to aquatic life (Noonari, N. B. et al., 2021; El-Moselhy, 2000; Santos et al., 2016). Fish, due to their position at upper trophic levels in the aquatic food web, are most exposed to such contaminants. Having metals bio-accumulate in their tissues, concentrations can exceed the values for water and sediments, posing greater risk to top predators like humans. The risk is further increased by chemicals such as methylmercury and other metal complexes, which have the potential for bio-magnification along the food web (Ray & Vashishth, 2024).

Heavy metals affect not only the physiology of fish but also act as indicators of ecosystem health. Fish are widely used as bioindicators as they are highly sensitive to environmental change and play a crucial role in human nutrition (Mazari, H. et al., 2021; Authman, 2015). Exposure to heavy metals may lead to behavioral, biochemical, and physiological changes, which can impair growth, reproduction, and survival, eventually putting both aquatic biodiversity and public health at risk (Soomro, A. H. et al. 2020; Maurya et al., 2019).

Therefore, monitoring and evaluating the concentrations of toxic metals in fish tissues, especially in organs like the gills, liver, muscle, and skin is crucial, reflecting both the pathway and magnitude of exposure. Such investigations are important not merely for defining baseline information on which to base regulatory requirements but also for informing public health policy and ensuring the sustainable use of aquatic resources (Kaçar, 2024). Through research into metal contamination of fish, we help to protect ecosystem integrity and consumer safety, as well as provide guidance for measures to avoid future environmental degradation.

2. Materials and Methods

2.1. Study area

The current research work was undertaken in Taluka Tangwani, Kashmore District of Sindh Province, Pakistan (Fig. 1.). Geographically, the site is situated at 28.3557° N latitude and 69.8918° E longitude. With an approximate population of about 289,259, Tangwani is a rural but economically active area, being majorly dependent on fishing and agriculture. Tangwani is located near the Indus River, one of the country's largest sources of freshwater. The river is important for sustaining the area's economy through irrigation purposes as well as being a main location for fishing. There are multiple fishing points along the river, such as the famous Qadirpur Loop Fishing Point, that sustain the livelihoods of fisherman in the area. The most commonly caught species are catfish, Rohu (*Labeo rohita*), and other freshwater fish species.

Apart from fishing, agriculture forms the backbone of Tangwani's economy. The region's warm climate, fertile alluvial soil, and abundant irrigation resources make it ideal for the cultivation of major crops such as cotton, wheat, and sugarcane. These agricultural and aquatic resources significantly contribute to the food security and economic stability of the area. Tangwani is a small, barely developed town with basic amenities like primary schools, health centers, and small local markets. The one clear feature of the town's cultural makeup is its age-old pottery craftsmanship. The town hosts a yearly pottery fair to which artisans and visitors are invited from across the region, highlighting the rich heritage and artisanal abilities of the people.



Fig. 1. Map of Tangwani

With regards to transportation, Tangwani is relatively linked to other areas of the district and province through road networks. Sukkur is the closest airport, some 60 kilometers away, and offers limited but vital air links for people and business. The fact that the region is dependent on river water, together with its economic reliance on agricultural output and fish production, makes Tangwani a site of paramount importance for environmental and ecological research, especially of water quality and bioaccumulation of contaminants in aquatic life.

2.2. Sampling

Sampling for this study was conducted with the assistance of local fishermen using Malian nets, which were deployed overnight in three prominent fish ponds located in Taluka Tangwani, District Kashmir. The selected sampling sites included Seth Abdul Majeed Sarki Fish Farm, Mir Saif-u-Rehman Khan Rind Pond, and Mir Manzoor Khan Bukhari Pond. Fish specimens were carefully selected based on size and age, with two weight categories considered 500 g (0.5 kg) and 1000 g (1 kg). These categories were chosen to observe variations in heavy metal accumulation based on fish biomass.

Immediately after capture, fish samples were transported in acid-washed plastic ice boxes, maintained at 20°C, and pre-acidified with nitric acid (HNO₃) to prevent metal leaching or degradation. In the laboratory, fish were dissected to isolate gills, muscles, liver, and skin tissues for further analysis. Tissue samples were dissolved using a combination of nitric acid (HNO₃) and hydrogen peroxide (H₂O₂) under controlled circumstances to completely break down organic content and mobilize trace metals into solution. Concentrations of chosen toxic and trace metals Aluminum (Al), Nickel (Ni), Lead (Pb), and Strontium (Sr) were measured employing an Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES). This technique was chosen due to its sensitivity and precision for analyzing trace amounts of metal impurities in biological materials.

2.3. Procedure

Fish samples were chosen according to the objective of the study and divided into two groups according to weight 500 g (0.5 kg) and 1000 g (1 kg). Homogenization of the fish tissues was done by a dry and clean pestle and mortar until homogenous consistency was obtained. Homogenized tissues were transferred into clean containers for further processing. Prior to dissection, each fish was thoroughly rinsed with ultrapure water to eliminate surface contaminants. Teflon forceps and stainless-steel scalpels were used to dissect fish under contamination-free conditions. Four tissues gills, muscles, liver, and skin were carefully extracted. These tissues were oven-dried at 70°C to a constant weight.

Once dried, the tissues were ground into fine powders using a clean, contamination-free grinding apparatus. Approximately 0.1 g of the powdered sample was transferred into a clean digestion vessel or tube for chemical digestion. To each sample, 6 mL of 65% nitric acid (HNO₃) and 2 mL of 35% hydrogen peroxide (H₂O₂) were added. The mixture was allowed to fully cover the powdered tissue and placed on a hot plate under well-ventilated conditions. The temperature was gradually increased to near boiling and maintained for 1 to 4 hours, until the solution became transparent, indicating complete digestion. After digestion, the samples were cooled to room temperature and filtered using Whatman No. 42 filter paper. The clear filtrate was collected and diluted with ultrapure water in a 25 mL volumetric flask to the required final volume.

All samples were prepared and analyzed in triplicates, and blank samples were also processed under identical conditions to eliminate any contamination bias. For quantitative metal analysis, an Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES) was used to determine the concentrations of aluminum (Al), nickel (Ni), lead (Pb), and strontium (Sr). Instrument calibration was carried out employing 1000 mg/L stock standard solutions of each element to generate calibration graphs. All analyses followed instrument-specific procedure for calibration, quality control, and quality assurance.

In order to provide safety and reliability in the digestion and analytical processes, the right personal protective equipment (PPE) such as gloves, lab coats, and eye protection was used at all times. Each step was conducted according to laboratory guidelines and government regulations to ensure data accuracy and analytical integrity.

2.4. Preparation of reagents

2.4.1. Preparation of 1000ppm solution of Al

For the analysis of elements by Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES), a 1000 ppm (mg/L) aluminum (Al) standard solution was prepared from aluminum nitrate $[\text{Al}(\text{NO}_3)_3]$, a readily available and highly soluble aluminum salt.

To make 100 mL of 1000 ppm aluminum solution 0.789 g of aluminum nitrate $[\text{Al}(\text{NO}_3)_3]$ was weighed accurately using a calibrated electronic balance. The weighed salt was dissolved in a minimal amount of deionized water in a clean volumetric flask. The mixture was shaken until the aluminum nitrate dissolved. Following the dissolution process, the solution was diluted to a final volume of 100 mL using deionized water. The solution was used as the stock standard (1000 ppm) for calibrating aluminum during ICP-OES analysis. Appropriate laboratory protocols, such as the use of acid-cleaned glassware and personal protective equipment (PPE), were adhered to in order to prevent contamination and for accuracy.

2.4.2. Preparation of 1000ppm solution of Ni

In order to perform elemental quantitation with Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES), a nickel (Ni) standard solution of 1000 ppm (mg/L) was prepared from nickel(II) chloride (NiCl_2), which is water-soluble and readily available salt used for nickel calibration.

In order to prepare 100 mL of 1000 ppm nickel solution, 0.221 g of nickel (II) chloride (NiCl_2) was weighed accurately with a calibrated digital balance. The salt was weighed and put into a clean beaker and dissolved in little deionized water. Once dissolved completely, the solution was poured into a 100 mL volumetric flask, and volume was completed to the mark with deionized water. This solution was applied as the 1000 ppm nickel stock standard for calibration in ICP-OES analysis. The glassware was well cleaned and washed with deionized water, and proper laboratory safety precautions were followed during the preparation.

2.4.3. Preparation of 1000ppm solution of Pb

For measuring lead (Pb) by Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES), a 1000 ppm (mg/L) standard solution of lead was made using lead(II) nitrate $[\text{Pb}(\text{NO}_3)_2]$, a very soluble and widely used salt for lead calibration. To make 100 mL of 1000 ppm lead solution 0.160 g of lead (II) nitrate $[\text{Pb}(\text{NO}_3)_2]$ was weighed accurately using a calibrated digital balance.

Weighed salt was dissolved in a small amount of deionized water in a clean beaker. Following dissolution, the solution was placed in a 100 mL volumetric flask. The volume was then made up to the mark with deionized water. The resulting solution served as the stock standard solution (1000 ppm) for calibration during ICP-OES analysis. All preparations were carried out using acid-cleaned glassware, and strict precautions were taken to avoid contamination. Personal protective equipment (PPE) was worn during the procedure due to the toxic nature of lead compounds.

2.4.4. Preparation of 1000ppm solution of Sr

For the quantification of strontium (Sr) using Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES), a 1000 ppm (mg/L) stock standard solution was prepared using strontium nitrate $[\text{Sr}(\text{NO}_3)_2]$, a water-soluble and commonly utilized salt in elemental analysis. To prepare 100 mL of 1000 ppm strontium solution 0.242 g of strontium nitrate $[\text{Sr}(\text{NO}_3)_2]$ was accurately weighed using a calibrated

digital balance. The measured quantity was dissolved in a small volume of deionized water in a clean beaker. After ensuring complete dissolution, the solution was transferred to a 100 mL volumetric flask. The volume was then made up to the mark using deionized water. The prepared solution was used as the 1000 ppm stock standard for Sr calibration during ICP-OES analysis. For accuracy and reliability, acid-cleaned glassware was employed, and all the preparation procedures were done with suitable personal protective equipment (PPE) in order to prevent contamination and exposure.

2.5. Quality assurance

In order to prevent analytical accuracy and potential contamination, prior to sample preparation and analysis, all of the glassware was soaked overnight in 10% nitric acid (HNO_3). Following acid treatment, glassware was rinsed extensively with ultra-pure water and air dried before use. Blank samples and quality control (QC) standards made from certified standard solutions were run with the test samples for verification of precision and reliability of measurement. Incorporation of blanks ensured the identification of background contamination, whereas the QC samples provided assurance of the accuracy and reproducibility of the instrument performance (Ansari, S., et al. 2021). Recovery tests were done through spiking known amounts of metals into a chosen sample for the determination of the efficiency and effectiveness of the digestion and analytical process. The recovery tests established the accuracy and reproducibility of the method for the determination of aluminum (Al), nickel (Ni), lead (Pb), and strontium (Sr).

The calibration curves of all four metals Aluminum (Al), Nickel (Ni), Lead (Pb), and Strontium (Sr) were prepared from standard solutions in different concentrations. Absorbance was determined by Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES). Calibration equations and correlation coefficients indicate outstanding linearity, ensuring the trust and authenticity of the method for trace metal determination. For Aluminum (Al) y is $0.0201x$, and Coefficient of Determination R^2 is 0.9999 (Fig. 2.).

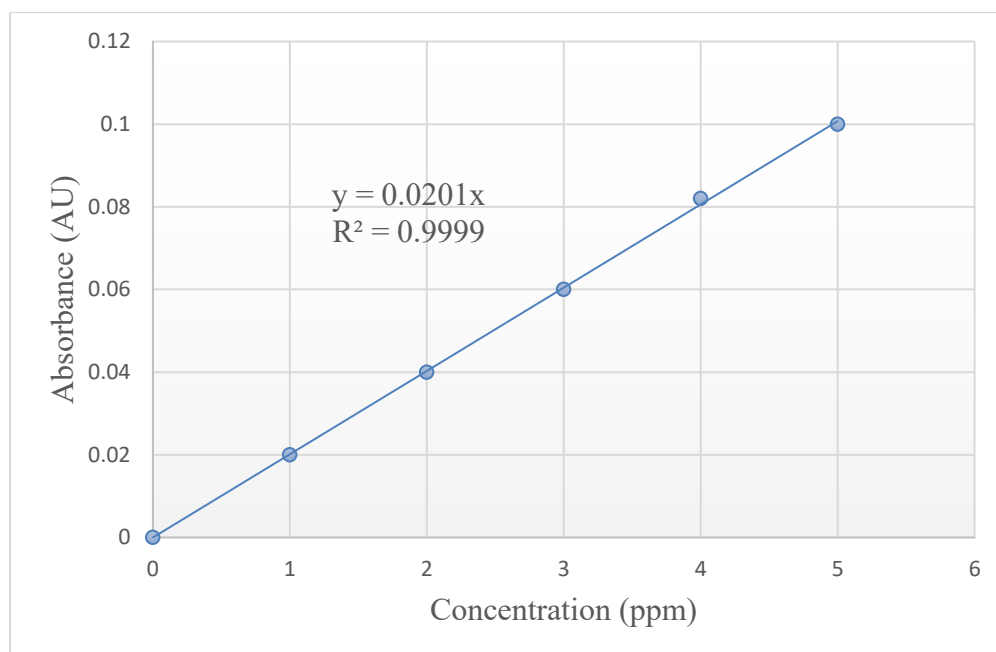


Fig. 2. Calibration Graph of Aluminum

For Nickel (Ni) y is $0.1691x + 0.0186$, and Coefficient of Determination R^2 is 0.998 (Fig. 3.).

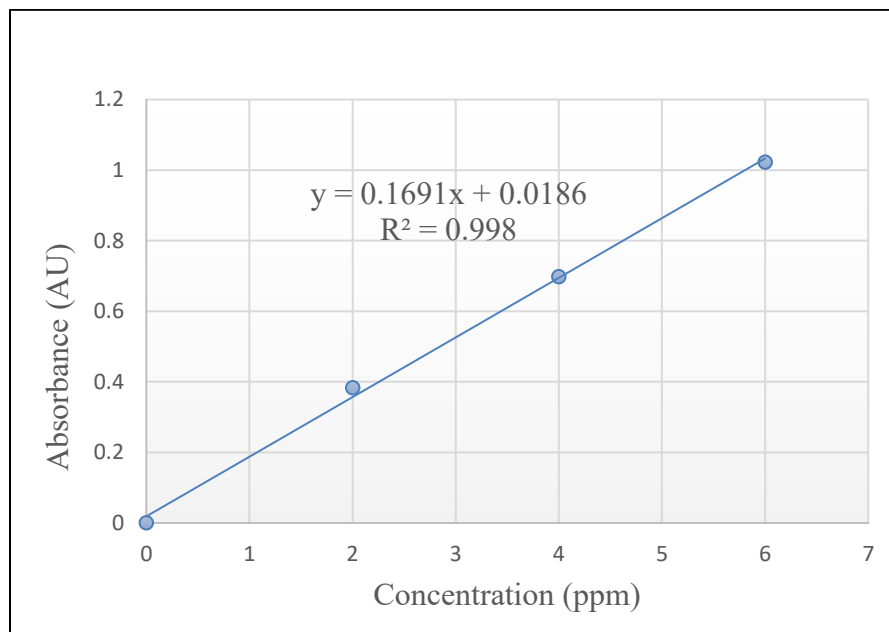


Fig. 3. Calibration Graph of Nickel

For Lead (Pb) y is $0.1223x - 0.001$, and Coefficient of Determination R^2 is 0.9997 (Fig. 4.).

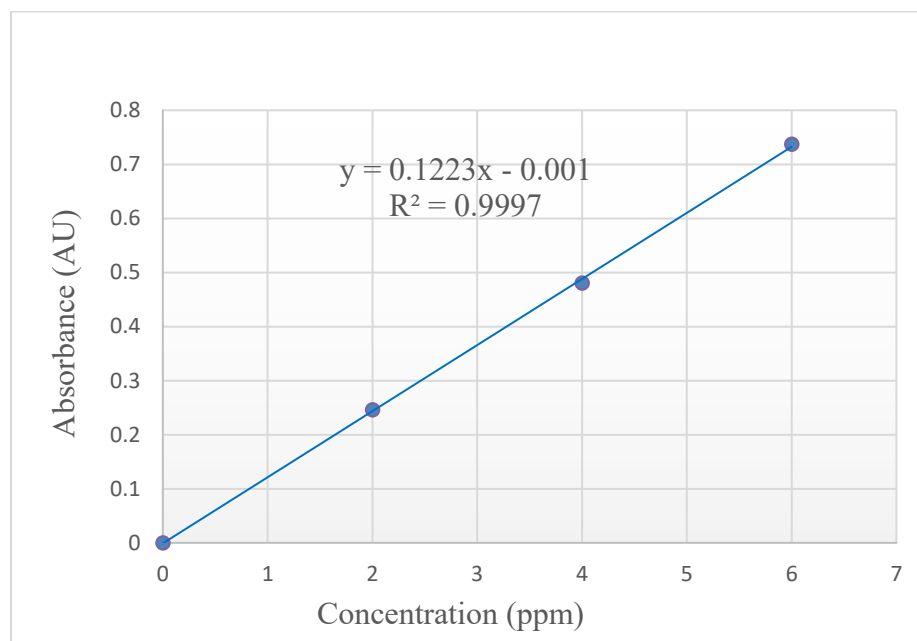


Fig. 4. Calibration Graph of Lead

Strontium (Sr) y is $0.01x$, and Coefficient of Determination R^2 is 0.9996 (Fig. 5.) The high R^2 values (≥ 0.998) indicate excellent linearity and sensitivity for all calibration curves, ensuring the suitability of the analytical method for quantitative trace metal analysis in biological tissues.

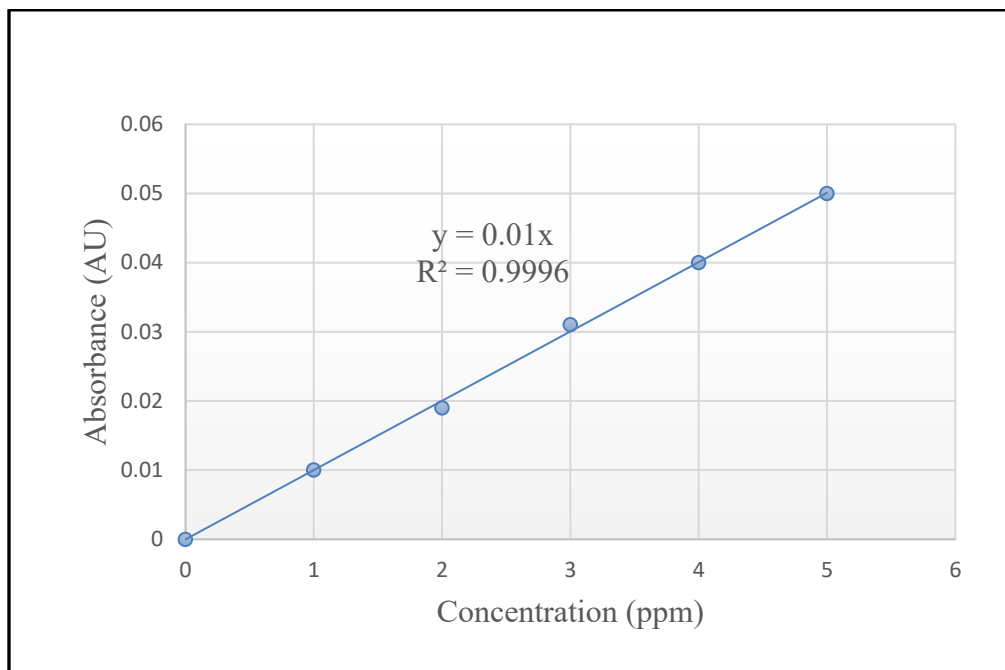


Fig. 5. Calibration Graph Strontium

3. Results and Discussion

Aluminum concentration (Table 1.) in Khagga and Singhara catfish from three different ponds (P1, P2, P3) in Taluka Tangwani, District Kashmir. The data represents aluminum levels (in mg/kg) in different tissues Gills (G), Muscles (M), Skin (S), and Lungs (L). The concentration of aluminum (Al) was assessed in various tissues (gills, muscles, skin, and lungs) of two types of catfish Khagga and Singhara at different body weights (1 kg and 0.5 kg) across three pond sites (P1, P2, P3). The data reveals several notable patterns, Muscles (M) consistently showed the highest aluminum accumulation across most samples, particularly in Khagga, with values reaching up to 2.71 mg/kg. Gills (G) and skin (S) also showed significant aluminum levels, indicating these tissues are also active sites for metal uptake. Lungs (L) generally exhibited lower aluminum concentrations, especially in Singhara at 0.5 kg body weight.

In most cases, Khagga fish had higher aluminum levels than Singhara in the same tissues and pond conditions, especially for 1 kg body weight. For example, in P1 Muscle tissue in 1 kg Khagga found to be 2.52 mg/kg while in Singhara 1.27 mg/kg. Similarly Gills of 1 kg Khagga was 1.38 mg/kg and in Singhara 2.08 mg/kg. This suggests species-specific differences in metal uptake or metabolism. The aluminum content in 0.5 kg fish was often higher than in 1 kg fish in certain tissues, particularly in Khagga. This could suggest that smaller fish are more vulnerable to aluminum exposure, possibly due to higher surface area-to-body mass ratios or faster metabolic activity. Pond 1 (P1) had slightly higher average aluminum concentrations in most tissues compared to P2 and P3, indicating it may be more contaminated. Ponds 2 and 3 showed similar but slightly lower accumulation, with some variation in Singhara fish. High aluminum levels in edible tissues like muscle and skin may pose risks to human health, especially with frequent consumption. The elevated metal concentrations also indicate potential contamination of local water bodies, possibly due to agricultural runoff, industrial effluents, or geogenic sources. Regular monitoring and mitigation measures are necessary to control metal pollution in aquaculture systems.

Table. 1. Aluminum in Khagga and Singhara Fish

Pond	Organ	1Kg (mg/kg)	Khagga 1Kg (mg/kg)	Singhara 1/2 Kg (mg/kg)	Khagga 1/2 Kg (mg/kg)	Singhara (mg/kg)
P1	G	1.38	2.08	2.25	1.28	
	M	2.52	1.27	2.71	1.79	
	S	2.43	1.29	1.79	0.68	
	L	1.36	0.99	1.75	0.67	
P2	G	1.38	0.99	1.78	0.66	
	M	2.51	1.29	2.26	0.68	
	S	2.41	1.27	2.69	1.76	
	L	1.37	2.10	0.84	1.28	
P3	G	1.32	0.99	1.78	0.68	
	M	2.51	1.29	2.26	0.68	
	S	2.41	1.28	2.70	1.77	
	L	1.35	2.11	0.83	1.28	
P1 = Pond no. 1; P2 = Pond no.2; and P3 = Pond no. 3						
G = Gills; M = Muscles; S = Skin; L = Lungs						

Table. 2. represents Nickel (Ni) concentrations (mg/kg) in different tissues of Khagga and Singhara catfish from ponds in Taluka Tangwani, District Kashmir. Nickel (Ni), a potentially toxic trace metal, was analyzed in various tissues of Khagga and Singhara catfish at two body weights (1 kg and 0.5 kg) collected from three pond sites (P1, P2, P3). The findings indicate that Ni accumulates differently depending on fish species, body weight, tissue type, and environmental exposure.

In gills (G) the highest Ni concentrations were observed in the gills of 1 kg Khagga across all ponds (0.0097–0.0098 mg/kg). Gills serve as the primary interface with water, and thus, show maximum Ni uptake due to their direct exposure to metal-contaminated water. Singhara gill values were significantly lower (0.00086–0.00093 mg/kg). Muscles (M) Showed very low levels of Ni, e.g., 0.00076–0.00084 mg/kg in 1 kg Khagga. This suggests that muscle tissue is less prone to Ni accumulation, which is positive from a food safety perspective since muscles are the most commonly consumed part. Singhara skin showed slightly higher Ni levels (up to 0.00095 mg/kg) than Khagga (as low as 0.00030 mg/kg), possibly due to differences in dermal absorption or mucus composition. Still, overall skin accumulation remained relatively low. Lung tissues showed moderate levels of Ni across both species. In 0.5 kg Khagga, lung Ni levels peaked at 0.0011 mg/kg, indicating some bioaccumulation even in internal organs. Khagga fish

exhibited significantly higher Ni levels in gills compared to Singhara, implying greater exposure or higher respiratory absorption. Singhara, however, showed slightly higher levels in other tissues like skin and muscles (e.g., muscle Ni in 1 kg Singhara = 0.00127–0.0013 mg/kg vs. Khagga = 0.00076–0.00084 mg/kg). These differences may reflect species-specific metabolism, behavior, or feeding habits. 1 kg fish (both species) generally showed higher Ni concentrations in gills and muscles compared to 0.5 kg fish, likely due to longer exposure duration, larger surface area for accumulation, Bioaccumulation over time. Lungs of 0.5 kg Khagga showed the same Ni level (0.0011 mg/kg) across all ponds, suggesting some tissue-specific retention regardless of age or size. Across all ponds, Ni concentration patterns remained consistent Gills in Khagga always had the highest Ni content. Minor variations in muscle and skin concentrations suggest stable environmental conditions or similar pollutant sources across ponds. Although Ni levels in edible tissues (muscle) are relatively low and within safe limits, the presence of Ni, especially in gills and lungs, signals ongoing environmental contamination. Chronic exposure could pose long-term risks to aquatic life and human health, especially if consumption rates are high. Continued Ni pollution in aquatic systems can lead to bioaccumulation and biomagnification, affecting entire food chains.

Table. 2. Nickel in Khagga and Singhara Fish

Pond	Organ	1Kg (mg/kg)	Khagga 1Kg (mg/kg)	Singhara 1/2 Kg (mg/kg)	Khagga 1/2 Kg (mg/kg)	Singhara 1/2 Kg (mg/kg)
P1	G	0.00980	0.00091	0.00047	0.00024	
	M	0.00076	0.00127	0.00017	0.00020	
	S	0.00039	0.00094	0.00035	0.00068	
	L	0.00041	0.00116	0.00110	0.00084	
P2	G	0.00970	0.00086	0.00047	0.00024	
	M	0.00082	0.00120	0.00016	0.00021	
	S	0.00030	0.00093	0.00034	0.00069	
	L	0.00039	0.00110	0.00110	0.00083	
P3	G	0.00970	0.00093	0.00049	0.00026	
	M	0.00084	0.00130	0.00018	0.00022	
	S	0.00032	0.00095	0.00036	0.00070	
	L	0.00041	0.00110	0.00110	0.00086	

P1 = Pond no. 1; P2 = Pond no.2; and P3 = Pond no. 3

G = Gills; M = Muscles; S = Skin; L = Lungs

Table. 3. shows the bioaccumulation of Lead (Pb) in Khagga and Singhara catfish based on your data from three pond sites (P1, P2, P3) in Taluka Tangwani, District Kashmore. Lead (Pb) is a non-essential and highly toxic heavy metal that poses serious ecological and health risks. This study analyzed Pb concentrations in various tissues — Gills (G), Muscles (M), Skin (S), and Lungs (L) — of two common freshwater catfish species: Khagga and Singhara, at two body weights (1 kg and 0.5 kg) from three separate pond sites.

In gills (G) highest Pb accumulation was observed in the gills of 0.5 kg Khagga from P2 (0.0245 mg/kg) and P3 (0.021 mg/kg). Gills, being directly exposed to water, act as primary sites for metal uptake, especially for waterborne Pb. Across all ponds, 1 kg Khagga gills consistently had higher Pb levels (~0.0095–0.0098 mg/kg) compared to Singhara (~0.0063–0.0071 mg/kg). Muscle tissues showed moderate Pb concentrations, with higher levels in 0.5 kg Khagga (up to 0.0177 mg/kg in P2) than Singhara. Since muscles are the main edible tissue, these levels are relevant for assessing human health risks. Generally, Singhara muscle Pb levels were lower than in Khagga. Pb concentrations in skin ranged from 0.00504–0.0122 mg/kg, with higher levels found in Khagga, particularly in P2 and P3. Singhara skin Pb levels were relatively low and stable, with values between 0.0036–0.0103 mg/kg. Lung tissues in 1 kg Khagga contained higher Pb levels (up to 0.0139 mg/kg in P1) compared to other tissues, suggesting Pb is also being internalized systemically. In 0.5 kg Singhara, lung Pb values were quite high (up to 0.0117 mg/kg in P3), indicating this tissue may also serve as a Pb storage site in smaller fish. Khagga consistently showed higher Pb accumulation in all tissues across ponds and sizes. Generally, Singhara had lower Pb levels, except in lungs of 0.5 kg fish, where Pb reached comparable values to Khagga. Khagga appears to be more susceptible to Pb bioaccumulation, especially in gills, muscles, and skin. In most cases, 0.5 kg Khagga showed higher Pb concentrations than 1 kg individuals, especially in gills and muscles. P2 Muscle (0.5 kg Khagga) = 0.0177 mg/kg vs. (1 kg) = 0.0056 mg/kg. This may be due to higher metabolic rate in smaller fish, greater surface area-to-body mass ratio, leading to more metal uptake, or shorter exposure durations before detoxification begins in larger fish. Pb levels were relatively consistent across ponds, with P2 showing slightly higher concentrations in some tissues (e.g., gills and muscles of 0.5 kg fish). This suggests similar contamination sources — possibly from agriculture runoff, industrial effluents, or soil erosion. Though Pb levels in edible tissues like muscle are within international safety limits, the presence of Pb — especially in multiple tissues — indicates bioavailability and chronic exposure in these fish species. Continuous Pb presence in fish indicates pollution of pond ecosystems, potentially harmful to aquatic biodiversity and unsafe for regular human consumption if unmonitored.

Table. 3. Lead in Khagga and Singhara Fish

Pond	Organ	1Kg (mg/kg)	Khagga 1Kg (mg/kg)	Singhara 1/2 Kg (mg/kg)	Khagga 1/2 Kg (mg/kg)	Singhara 1/2 Kg (mg/kg)
P1	G	0.0095	0.0063	0.0199	0.0039	
	M	0.0057	0.0071	0.0127	0.0025	
	S	0.0062	0.0051	0.0109	0.0103	
	L	0.0139	0.0072	0.0042	0.0107	
P2	G	0.0098	0.0071	0.0245	0.0047	
	M	0.0056	0.0081	0.0177	0.0033	

P3	S	0.0112	0.0052	0.0122	0.0038
	L	0.0127	0.0092	0.0036	0.0113
	G	0.0098	0.0070	0.0210	0.0042
	M	0.0056	0.0077	0.0170	0.0034
	S	0.0112	0.0054	0.0110	0.0036
	L	0.0125	0.0077	0.0041	0.0117
P1 = Pond no. 1; P2 = Pond no.2; and P3 = Pond no. 3					
G = Gills; M = Muscles; S = Skin; L = Lungs					

Table. 4. shows discussion on the bioaccumulation of Strontium (Sr) in Khagga and Singhara catfish from Ponds P1, P2, and P3 in Taluka Tangwani, District Kashmore. The data represents Sr concentrations (in mg/kg) in four tissues Gills (G), Muscles (M), Skin (S), and Lungs (L) for fish of two different weights (1 kg and 0.5 kg). Strontium (Sr) is an alkaline earth metal naturally present in water bodies but can accumulate in aquatic organisms when present in excess due to environmental contamination (e.g., industrial discharge, fertilizers). While less toxic than heavy metals like Pb or Ni, elevated Sr levels can still affect bone formation, organ function, and ecological balance.

In gills (G) highest Sr concentration detected was 0.313 mg/kg in 1 kg Khagga (P3). Gills of Khagga consistently showed much higher Sr levels (0.278–0.313 mg/kg) compared to Singhara (0.0227–0.033 mg/kg). Sr accumulation in gills confirms direct absorption from water. In 1 kg fish, muscle Sr levels were low for both species (~0.0233–0.0483 mg/kg). However, in 0.5 kg fish, muscle Sr levels increased significantly, especially in Singhara (0.156 mg/kg, P2) and Khagga (0.094 mg/kg, P2). Muscles are critical for human consumption, so even moderate accumulation warrants attention for food safety. Skin tissues of 0.5 kg Khagga showed remarkably high Sr levels, with a peak of 0.195 mg/kg in P2 and 0.19 mg/kg in P3. This could be due to dermal contact with contaminated water and Sr's affinity for keratinous tissues. Lungs showed moderate to high Sr accumulation, especially in 1 kg Khagga (up to 0.193 mg/kg in P2). Notably, 0.5 kg Singhara lungs in P2 contained 0.151 mg/kg, suggesting systemic Sr absorption into internal organs. Khagga significantly higher Sr in gills, skin, and lungs, especially in 1 kg fish. Singhara exhibited higher Sr in muscles and lungs in 0.5 kg fish, particularly in P2. Khagga tends to bioaccumulate more Sr overall, while Singhara shows variable tissue-specific uptake. 0.5 kg fish often showed higher Sr accumulation in muscle and skin compared to 1 kg fish e.g., Khagga muscle in P1: 0.0807 mg/kg (0.5 kg) vs. 0.0233 mg/kg (1 kg). Possible reasons are higher metabolic rate in smaller fish, greater surface-to-volume ratio leading to more efficient absorption, less developed excretory or detox systems in juvenile fish. In pond 3 (P3) highest Sr in 1 kg Khagga gills (0.313 mg/kg) and 0.5 kg Singhara skin (0.16 mg/kg), pond 2 (P2) showed widespread high Sr values across all tissues and fish types suggesting it may be the most contaminated pond. Pond 1 (P1) had moderate Sr accumulation, but notable peaks in lung and muscle tissues of Khagga. While Sr is not as acutely toxic as Pb or Ni, elevated levels in edible tissues (muscle, skin) can affect bone health and mineral metabolism in humans over time. Ecologically, continuous Sr accumulation may disrupt fish organ functions, especially in gills and lungs. The trend suggests localized environmental contamination, likely from fertilizers, groundwater leaching, or industrial inputs.

Table. 4. Strontium in Khagga and Singhara Fish

Pond	Organ	1Kg (mg/kg)	Khagga	1Kg (mg/kg)	Singhara	1/2 Kg (mg/kg)	Khagga	1/2 Kg (mg/kg)	Singhara
P1	G	0.2780		0.0227		0.0762		0.0597	
	M	0.0233		0.0252		0.0807		0.0563	
	S	0.0372		0.0335		0.1450		0.0645	
	L	0.0936		0.0159		0.0750		0.1010	
P2	G	0.3090		0.0280		0.0960		0.1090	
	M	0.0483		0.0350		0.0940		0.1560	
	S	0.0220		0.0480		0.1950		0.1140	
	L	0.1930		0.0220		0.1250		0.1510	
P3	G	0.3130		0.0330		0.0840		0.0610	
	M	0.0260		0.0350		0.0890		0.1100	
	S	0.0220		0.0380		0.1900		0.1600	
	L	0.1370		0.0170		0.1180		0.1000	
P1 = Pond no. 1; P2 = Pond no.2; and P3 = Pond no. 3									
G = Gills; M = Muscles; S = Skin; L = Lungs									

4. Conclusion

The findings of this study revealed that concentrations of trace and toxic elements including aluminum (Al), nickel (Ni), lead (Pb), and strontium (Sr) were detected in various tissues (gills, liver, muscles, and skin) of two catfish species (Khagga and Singhara) collected from fish ponds in Taluka Tangwani, District Kashmore@Kandhkot. Most metal concentrations remained within the permissible limits set by FAO and WHO, with the exception of aluminum (Al), which exceeded recommended levels and raises potential health concerns.

For aluminum, the maximum concentration recorded was 2.7 mg/kg in the muscle of a 0.5 kg Khagga, and 2.1 mg/kg in the muscle of a 1 kg Singhara, significantly exceeding the WHO safety limit of 0.4 mg/kg. This indicates a notable bioaccumulation of aluminum, which could pose health risks to regular fish consumers in the region.

Nickel (Ni) concentrations ranged from 0.0002 to 0.010 mg/kg, with the highest found in the gills of 1 kg Khagga and the lowest in liver and skin samples. All observed Ni levels were within international safety

standards, although prolonged exposure even at low concentrations has been associated with dermatological, respiratory, and cardiovascular issues.

Lead (Pb), a known neurotoxin, was detected in concentrations between 0.0022 to 0.013 mg/kg. The highest Pb level was observed in the liver of 1 kg Khagga fish, and although these values were within permissible limits, continuous exposure should be monitored due to the metal's cumulative toxicity.

Strontium (Sr) levels ranged from 0.012 to 0.3 mg/kg, with the highest value recorded in the gill tissues of Khagga and the lowest in Singhara muscle. While Sr is not as toxic as other heavy metals, its bioaccumulation still warrants monitoring.

The study highlights that while Ni, Pb, and Sr levels were within the safe limits, aluminum concentrations were consistently elevated up to five times higher than the WHO limit especially in the muscles and skin of Khagga and Singhara. This poses a potential health hazard for local fish consumers. The findings underscore the urgent need for regular monitoring, pollution control, and public awareness initiatives to mitigate heavy metal exposure through fish consumption in Taluka Tangwani.

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5. Conflicts

The authors declare no conflict of interest.

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