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Assessment of Lead and Cadmium Contamination in Water Resources of Northeastern Thailand and Its Implications for Ecological Risk Evaluation

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ABSTRACT

The accumulation of heavy metals in the environment can adversely impact both the food chain and human health. This research investigated cadmium and lead accumulation in water, sediment, and fish in Huai Kho Reservoir, Northeastern Thailand, with samples collected during both the rainy and dry seasons. Moreover, the bioconcentration factor (BCF), potential ecological risk index (PERI), and tolerable daily intake (TDI) were evaluated. Cadmium and lead levels in water and sediment exceeded the standard limits. Cadmium levels ranged from 0.02 to 0.06mg/ L and 0.31 to 0.42mg/ L, respectively, while lead levels ranged from 0.08 to 1.13mg/ kg and 3.63 to 15.28mg/ kg, respectively. Lead exceeded the limits in 18 fish species, while cadmium exceeded them in some species. In both the rainy and dry seasons, cadmium levels peaked in muscle, while lead was at its highest in the digestive tract. The BCF for lead was higher than that for cadmium, although no significant seasonal differences were observed in the accumulation. Even though the TDI assessment didn't exceed standard guidelines, the PERI indicated a high-risk level, emphasizing the need for legal measures regulating the use of heavy metal-containing substances.

INTRODUCTION

In recent decades, the presence of heavy metals in freshwater ecosystems has emerged as a significant concern, posing threats to both public water resources and the safety of fishery resources consumed by humans. Heavy metals, sourced from natural occurrences, industrial activities, sewage effluents, and agricultural inputs, continuously enter lakes, reservoirs, and rivers, posing serious risks due to their toxic nature, persistence, and tendency to bioaccumulate and biomagnify in the food chain **(Modaihsh *et al*., 2004; Mortvedt, 2004; Terra *et al*., 2008; Tanee *et al*., 2013)**. These contaminants have been linked to adverse effects on human and animal health, as well as on plants, soil, and aquatic ecosystems **(Grimm *et al*., 2008)**. Cadmium (Cd) and lead (Pb) are particularly concerning due to their potential health impacts. Cadmium exposure can lead to renal and glomerular damage, bone demineralization, increased fracture risk, reduced lung function, and emphysema, typically following prolonged exposure. Lead exposure, on the other hand, can result in cognitive impairment, mood alterations, neurosensory dysfunction, decreased kidney function, elevated blood pressure, inhibition of heme biosynthesis enzymes, reduced sperm count, and spontaneous abortion **(Sirisangarunroj *et al*., 2023)**.

Heavy metal contamination poses significant threats to the ecological equilibrium of aquatic environments and various aquatic organisms. Fish are highly susceptible to the adverse impacts of these pollutants **(Yousafzai *et al*., 2012)**, as they are unable to evade their effects. Consequently, fish serve as valuable indicators for assessing the health of aquatic ecosystems due to the bioaccumulation of pollutants in the food chain, often resulting in deleterious effects and mortality within aquatic systems **(Baby *et al*., 2010; Yousafzai *et al*., 2012)**. The toxicity of heavy metals, attributed to their accumulation in fish tissues, has spurred numerous investigations into heavy metal bioaccumulation in fish populations worldwide **(Rashed, 2001; Chi *et al*., 2007; Dural *et al*., 2007; Nesto *et al*., 2007; Terra *et al*., 2008; Vinodhini & Narayanan, 2008; Abdel-Baki *et al*., 2011; Ambedkar & Muniyan, 2011; Tanee *et al*., 2013; Saowakoon *et al*., 2021; Markmanuel *et al*., 2022; Vo *et al*., 2022; Sirisangarunroj *et al*., 2023)**. Previous reports revealed high levels of some heavy metals including Cd and Pb in Northeastern Thailand **(Tanee *et al*., 2013; Tanee *et al*., 2017; Zeng *et al*., 2020)**.

The Huai Kho Reservoir, situated in Na Chueak District, Maha Sarakham province, Northeastern Thailand, serves as a medium-sized water storage pond amidst agricultural areas and residential settlements. It acts as a recipient of untreated community wastewater discharge, potentially containing heavy metals. With a recorded presence of 22 fish species, freshwater fish from this reservoir are a significant protein source for local residents **(Khowhit *et al*., 2023)**. These fish are not only consumed fresh but are also preserved and processed into various products. Utilizing heavy metal levels in fish for environmental monitoring has proven valuable **(Rashed, 2001)**, yet there is insufficient information on heavy metal accumulation in fish habitats surrounding the Huai Kho Reservoir, particularly regarding the influence of runoff and flooding on heavy metal distribution.

This study seeked to assess heavy metal concentrations in ecosystem and in different fish tissues, including muscle, gills, and the digestive tract, focusing on commonly consumed fish species caught from the Huai Kho Reservoir. Moreover, the potential ecological risk index (PERI) and tolerable daily intake (TDI) were also evaluated.

MATERIALS AND METHODS

1. Ethical statement

All research procedures involving animal use were approved by the Institutional Animal Care and Use Committee of Mahasarakham University with the approval No.

IACUC-MSU-17/2023. All animal sample collection was complied with the applicable laws of Thailand.

2. Study area

The study was conducted at coordinates 15°49'35.3"N 103°02'14.3"E within the Khwao Rai subdistrict, Na Chueak district, Maha Sarakham province, the Huai Kho Reservoir was constructed in 1968 (Fig. 1). Spanning an area of approximately 2,113 square kilometers or 6,340 acres, it features three sandy islands and sits at an elevation of 150 meters above sea level. With a capacity of 31.338 million cubic meters, it can retain 208 square kilometers of rainwater. The reservoir serves multiple functions including irrigation, domestic water supply, and aquaculture, and is popular for recreational activities like swimming and boating. Fishing is a year-round activity and a vital source of income for local fishermen. The reservoir supports a thriving fishing industry, supplying fresh catch and processed products, such as fermented fish, salted fish, and pickled fish to local markets.

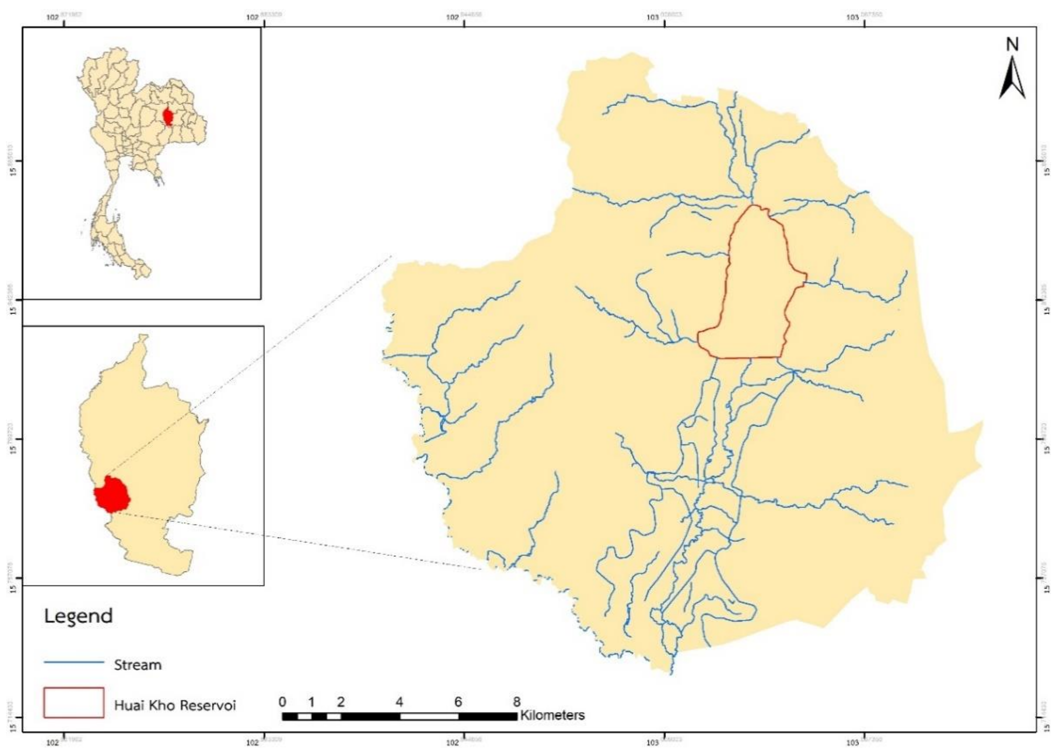

Fig. 1. Map of Thailand and location of the Huai Kho Reservoir

3. Collection of water and soil sediment samples

Sampling sites were selected based on ecological characteristics and human activities in the area, utilizing geographic information system (GIS) data. The technique for collecting water and soil sediment samples followed the protocols of **Abdel-Baki *et al.* (2011)** and **Tanee *et al.* (2013)** with modifications. Water samples were collected in plastic bottles, in triplicate from a depth of 50cm below the water surface to ensure representativeness. Nitric acid was added to the plastic bottles to preserve water samples. Sediment samples were collected in triplicate using an Eckman grab from the same locations as the water samples. The sediment samples were combined in a plastic tub, thoroughly mixed, and then transferred to a container. Both water and sediment samples were packed on ice for transportation to the laboratory to maintain sample integrity.

4. Preparation of fish samples

Fish samples collected by our previous research **(Khowhit *et al*., 2023)** at various stations within the reservoir were utilized for the analysis. Morphometric measurements, including total length and weight, were recorded, and gill, liver, muscle, and digestive tracts were dissected using sterile instruments. The dissected tissues were desiccated in an oven set at 105°C. Whole fish specimens were preserved in 10% formalin for taxonomic identification, which was conducted using a modified comparison method with the Fish Base online database (http://www.fishbase.org/home.htm), supplemented by taxonomic keys outlined in **Rainboth (1996)** and **Vidthayanon (2008)**.

5. Heavy metals analysis

Glassware was preconditioned by immersing it in nitric acid and rinsing with deionized-distilled water before digestion. Water samples (50mL) underwent digestion with a 12mL digestion mixture ($HClO_4$: HNO_3 at a ratio of 1:3) at 80°C for 2 hours, then brought to a final volume of 50mL with deionized-distilled water **(Agunbiade *et al*., 2009)**. Sediment samples were dried in an oven at 100°C for 24 hours, then cooled and pulverized using a mortar and pestle. Approximately, 1g of each dried sample was digested in a 12mL digestion mixture and boiled at 100°C for 30 minutes **(Pyle *et al*., 2005)**.

The dried fish tissues were finely grounded, and 1g of the powdered tissues was digested with a 12mL digestion mixture at approximately 80°C until obtaining a complete dissolution of the tissues **(Agunbiade *et al*., 2009)**. The resulting clear solutions were adjusted to a final volume of 50mL using deionized-distilled water. Subsequently, the digested samples of water, sediment, and fish tissues were analyzed for heavy metal concentrations, specifically Cd and Pb, utilizing an AA 6200 Atomic Absorption Spectrophotometer (Shimadzu, Japan). All analyses were conducted in triplicate for accuracy and reliability.

6. Data analysis

The mean and standard error of the mean for metal concentrations in water, sediment, and fish tissues were computed. Descriptive analysis was conducted using a one-way ANOVA, followed by Tukey's post-hoc test ($P< 0.05$) to determine any significant differences among the groups. All statistical analyses were executed utilizing SPSS 19.0 for Windows.

6.1. Bioconcentration factor (BCF)

Bioconcentration is a crucial concept within aquatic toxicology, providing insights into whether a substance within a living organism attains a concentration surpassing that of its ambient environment. The BCF facilitates this comparison, representing the ratio of heavy metal concentrations in fish tissues to those in water. The BCF was determined using the formula suggested by **Sanou *et al*. (2021)**, as follows:

$$BCF = \frac{C_F}{C_W}$$

In which C_F is the concentration of the heavy metal in the muscle of the fish, and C_W is the concentration in the water.

6.2. Potential ecological risk index (PERI)

The PERI, originally proposed by **Hakanson (1980)**, has been widely utilized to assess the impact of heavy metals in sediments on the environment. This method has garnered significant attention internationally and has been influential in various studies **(Guo *et al*., 2010; Poopa *et al*., 2015; Liu *et al*., 2021)**. In this study, PERI was employed to quantify the potential ecological risk associated with heavy metals present in the sediments of the study sites. The calculation was performed using the following formula, as outlined by **Hakanson (1980)** with description by **Liu *et al*. (2021)**:

$$C_f^i = \frac{C_s^i}{C_n^i} \quad (1)$$

$$E_r^i = T_r^i \cdot C_f^i \quad (2)$$

$$PERI = \sum_{i=1}^{n} E_r^i \quad (3)$$

Where, C_f^i represents the individual pollution factor, C_s^i denotes the observed value of heavy metals in soil samples, C_n^i is the reference value of the metal derived from background values of soil heavy metals in the study area, E_r^i signifies the potential ecological risk index of an individual metal, T_r^i represents the toxicity response coefficient of a specific kind of metal toxicity, using standard heavy metal toxicity coefficient, defined as 30 for Cd and 5 for Pb **(Guo *et al*., 2010)**. The PERI is the summation of E_r^i, the individual ecological risk of heavy metals and integrated potential ecological risk, classified into the categories according to **Liu *et al*. (2021)**, as shown in Table (1).

Table 1. Categories for potential ecological risks[1]

Risk category	Low risk	Moderate	High	Very high	Disastrous
Grade	I	II	III	IV	V
E_r^i	< 30	30–60	60–120	120–240	> 240
PERI	< 100	100–200	200–400	> 400	-

[1]Purposed by **Liu *et al*. (2021)**.

6.3. Tolerable daily intake of metals (TDI)

Fish muscles constitute a primary source of food consumption for the human population. Consequently, in assessing human health risks associated with metal exposure, we focused on evaluating fish muscles using the tolerable daily intake (TDI) method, which serves as a quantitative health risk assessment tool. The TDI was determined using the following equation, as proposed by **Markmanuel *et al*. (2022)**:

$$TDI = C \times \frac{IR}{BW}$$

Where, C is the mean heavy metals concentration in fish muscle (mg/g) of dry weight basis. IR is the daily ingestion rate of fish products for adults (0.3mg/kg-bw/day). BW is the average body weight, which in this study was set at 70kg for adults.

RESULTS AND DISCUSSION

1. General site conditions

Common conditions at the sampling stations were measured during the rainy and dry seasons. The pH ranged from 5.0 to 9.0, the temperature was 28.4 to 29.7°C, and the dissolved oxygen (DO) levels were 6.49 to 7.24mg/ L, as shown in Table (2). These values are at the standard levels **(Duangsawasdi, 1988)**, hence they are appropriate for the living of aquatic organisms.

Table 2. General properties of water at the collection sites include pH, temperature, and dis-solved oxygen (DO)

Collection period		Water condition in the sampling sites (mean)		
Season	**Month**	**pH**	**Temperature (°C)**	**DO (mg/L)**
Rainy	September	8.41	33.6	7.52
	Early-October	6.85	30.2	6.07
	Mid-October	6.91	25.3	5.89
	average	7.39	29.7	6.49
Dry	November	8.43	31.8	7.61
	December	7.36	30.0	6.64
	January	8.35	23.4	7.46
	average	8.05	28.4	7.24
Surface water quality standards[1]			6.0 - 9.0	23 - 32

[1]Surface water quality standards referred to **Duangsawasdi (1988)**.

2. Seasonal heavy metal concentration in water and soil sediments

The heavy metals, cadmium (Cd) and lead (Pb) accumulation in water from the sampling sites in rainy and dry season were averaging 0.02- 0.06mg/ L and 0.31- 0.42mg/

L, respectively (Table 3). These average values exceeded the standard level, which in fact, must not exceed 0.005 and 0.05mg/ L, respectively. The accumulation levels are insignificantly different ($P > 0.05$) between rainy and dry seasons collections.

Additionally, in all the sampling sites, the accumulation levels exceeded the standard levels, with no significant difference between rainy and dry season samplings, regardless of the water's utilization of each site. This refers to the fact that heavy metals can contaminate every water entry site. This is likely to take place since there are agricultural areas around the water reservoir, chemical fertilizers, and other agricultural chemicals are heavy metal contaminated, hence the metals were washed from the lands according to **Modaihsh *et al*. (2004)**.

Table 3. Concentration of cadmium and lead in water and sediment samples collected during rainy and dry seasons

Collection period		Concentration in surface water (mean ± SD, mg/L)		Concentration in sediment (mean ± SD, mg/kg)	
Season	**Month**	**Cadmium**	**Lead**	**Cadmium**	**Lead**
Rainy	September	0.02 ± 0.01	0.31 ± 0.02	1.07 ± 0.35	4.42 ± 2.59
	Early-October	0.02 ± 0.01	0.35 ± 0.02	0.08 ± 0.02	4.81 ± 1.32
	Mid-October	0.02 ± 0.01	0.37 ± 0.01	0.94 ± 0.39	4.08 ± 1.04
	Average	0.02 ± 0.00	0.34 ± 0.03	0.70 ± 0.54	4.44 ± 0.37
Dry	November	0.04 ± 0.01	0.39 ± 0.02	1.05 ± 1.11	3.63 ± 1.63
	December	0.05 ± 0.00	0.41 ± 0.02	0.84 ± 0.15	8.06 ± 7.42
	January	0.06 ± 0.00	0.42 ± 0.01	1.13 ± 0.39	15.28 ± 1.07
	Average	0.05 ± 0.01	0.41 ± 0.02	1.01 ± 0.15	8.99 ± 5.88
P-value		0.116	0.283	0.060	0.076
Surface water quality standards[1]		0.005-0.05	0.05	-	-
Sediment quality standards[2]		-	-	1	36

[1]Surface water quality standards referred to **Ministry of Natural Resources and Environment, Thailand (1994)**.

[2]Sediment quality standards referred to **Ministry of Natural Resources and Environment, Thailand (2022)**.

The observed levels of heavy metals in the sediment collected during rainy and dry seasons revealed that Cd levels ranged from 0.08- 1.13mg/ kg, while Pb levels ranged from 3.63- 15.28mg/ kg (Table 3). The Cd levels exceeded the standard allowed level; however in early October, the levels were at the standard level. Comparing the levels in the sediment between rainy and dry seasons, theyshowed no significant difference ($P>$ 0.05). The highest level was found in January (Table 3), which is in the dry season. Moreover, **Modaihsh *et al*. (2004)** reported that inorganic fertilizers are contaminated by Cd and Pb. Additionally, the geographical location of the Huai Kho Reservoir, which is surrounded by agricultural farms that extensively use organic fertilizers and other agricultural chemicals, and the water which does not easily run off, both factors make this area vulnerable to pollution. This geography is suitable for the accumulation of heavy metals, in contrast to easily run-off areas like the Chi River in Muang district, Mahasarakham province. **Tanee *et al.* (2013)** reported that the cadmium (Cd) levels in those areas do not exceed the standard level. Accordingly, in the soil sediment from all the sampling sites, the Cd level did not go over the standard level, and the Pb level was also within the standard level. The levels of Cd and Pb in the influx and run off sampling sites between the seasons are insignificantly different ($P > 0.05$).

The concentrations of Cd and Pb in water and sediment during the dry season are higher compared to those during the rainy season, despite the higher agricultural activities in the rainy season. A possible explanation for this discrepancy could be the concentration of metals due to the reduced volume associated with the higher evaporation rate induced by the elevated water temperature during the dry season. Similar results were observed in the studies of **Obasohan and Eguavoen (2008)** and **Rasheed *et al*. (2024)**, who attributed the phenomenon to an increased evaporation rate during the dry season.

3. Heavy metals concentration in fish species

The 18 fish species collected are listed in Table (4). Cd and Pb levels were detected in different tissues of the fish including muscle, digestive tract, and gill. Considering the levels in all studied tissues, in the rainy season, Cd levels ranged from 0.02 to 0.27mg/ kg, whereas Pb levels ranged from 3.67 to 454.08mg/ kg. In the dry season, Cd levels ranged from 0.04 to 0.13mg/ kg, whereas Pb levels ranged from 0.49 to 53.42mg/ kg.

These results indicate that the Pb levels in the fish species exceed the standard limit, set at 0.3mg/ kg, while the Cd levels in 10 species during the rainy season and one species during the dry season exceeded the standard limit, set at 1.0mg/ kg **(Food & Drug Administration, Ministry of Public Health, Thailand, 2020)**. These findings align with previous reports by **Tanee *et al*. (2013, 2017)**, which showed that Cd levels in fish species caught from the Chi River, Moon River, and Huai Kaeng reservoir in northeastern Thailand exceeded the standard limits. Additionally, our study found that Pb levels also exceeded the standard limits. These high levels related to the high levels found in the water and soil sediment samples, which can be affected by agricultural areas around the reservoir according to the reports by **Rashed (2001)** and **Rajeshkumar and Li (2018)**. Furthermore, the levels of heavy metal accumulations are influenced by various factors related to the fish, including age, size, weight, sex, species, and food and feeding habitat **(Dural *et al*., 2007; Yousafzai *et al*., 2012)**.

Within the fish tissues, cadmium accumulation levels follow the order of muscle > digestive tract > gill, while lead accumulation levels follow the order of digestive tract > gill > muscle in both seasons (Table 4). Importantly, in the muscle, which is commonly consumed as food, the levels of cadmium in some species exceed the standard level, and the lead levels in all species exceed the standard level according to the **World Health Organization (2003)** and the **Food and Drug Administration, Ministry of Public Health, Thailand (2020)**. These sources state that for food safety, cadmium levels should not exceed 0.5- 1.0mg/ kg, and lead levels should not exceed 0.3- 0.5mg/ kg. These results are crucial data for raising public awareness and managing environmental protection to safeguard human health. Heavy metals can pose significant risks to human health by adversely affecting the liver, kidneys, and brain through the inhibition of metabolic enzymes. Additionally, they can induce mutations, cancers, and other diseases **(Manahan, 1992)**.

Table 4. Concentration of cadmium and lead in different tissues of fish species collected during rainy and dry seasons

Fish species	Tissue	Concentration in rainy season (mg/kg)		Concentration in dry season (mg/kg)	
		Cadmium	Lead	Cadmium	Lead
Puntius leiacanthus	M	0.02 ± 0.00	0.70 ± 0.42	0.04 ± 0.01	0.70 ± 0.71
	D	0.12 ± 0.13	1.45 ± 1.45	0.05 ± 0.02	2.56 ± 2.11
	G	0.13 ± 0.08	3.76 ± 0.80	nd.	6.42 ± 2.85
Oxyeleotris marmorata	M	0.06 ± 0.01	1.00 ± 0.00	0.08 ± 0.01	1.07 ± 1.16
	D	0.02 ± 0.00	2.67 ± 0.00	0.04 ± 0.00	3.45 ± 2.54
	G	0.04 ± 0.04	nd.	0.02 ± 0.01	5.33 ± 3.04
Anabas testudineus	M	0.07 ± 0.01	2.00 ± 0.00	0.03 ± 0.01	2.14 ± 1.22
	D	0.01 ± 0.01	8.59 ± 0.00	0.03 ± 0.02	1.95 ± 0.29
	G	0.02 ± 0.01	6.68 ± 0.00	0.01 ± 0.00	11.11 ± 1.54
Trichopodus trichopterus	M	0.05	4.21	0.05 ± 0.00	1.74 ± 1.34
	D	nd.	nd.	nd.	53.42 ± 0.00
	G	nd.	3.34	0.01 ± 0.01	8.95 ± 2.80
Hampala dispar	M	0.09 ± 0.07	2.67 ± 0.61	0.02 ± 0.00	0.87 ± 0.42
	D	0.05 ± 0.05	6.04 ± 2.36	0.03 ± 0.01	3.28 ± 0.46
	G	0.06 ± 0.03	5.01 ± 2.65	0.03 ± 0.01	2.51 ± 2.74
Hemibagrus filamentus	M	0.06	3.00	No sample	No sample
	D	0.01	5.01		
	G	0.10	4.41		
Dermogenys siamensis	M	0.02	4.01	No sample	No sample
	D	nd.	100.16		
	G	nd.	13.36		
Channa striata	M	No sample	No sample	0.02	3.21
	D			0.02	1.69
	G			0.03	2.05
Labiobarbus siamensis	M	0.03 ± 0.01	2.60 ± 0.53	0.03 ± 0.00	1.55 ± 1.30
	D	0.01 ± 0.01	3.94 ± 0.30	0.03 ± 0.01	nd.
	G	0.01 ± 0.00	15.31 ± 14.23	0.01 ± 0.00	nd.
Clarias batrachus	M	0.03	0.60	0.04 ± 0.00	1.60 ± 0.00
	D	0.20	2.17	0.02 ± 0.01	2.82 ± 1.11
	G	0.07	2.40	0.02 ± 0.00	1.15 ± 0.95
Oreochromis niloticus	M	No sample	No sample	0.03	1.40
	D			0.01	2.40
	G			0.02	5.46
Tetraodon leiurus	M	0.05 ± 0.00	nd.	0.04 ± 0.01	0.49 ± 0.95
	D	nd.	53.42 ± 0.00	0.04 ± 0.02	nd.
	G	nd.	400.66 ± 5.78	nd.	nd.
Parambassis siamensis	M	0.03 ± 1.06	5.87 ± 2.70	0.05 ± 0.00	2.90
	D	No tissue	No tissue	No tissue	No tissue
	G	No tissue	No tissue	No tissue	No tissue
Anematichthys apogon	M	0.01 ± 0.00	0.80 ± 0.69	0.03 ± 0.01	0.20 ± 0.00
	D	0.02 ± 0.02	126.21 ± 0.00	0.01 ± 0.01	3.34 ± 1.46
	G	0.01 ± 0.00	19.08 ± 6.88	nd.	12.19 ± 0.00
Henicorhynchus siamensis	M	0.02 ± 0.01	2.24 ± 0.98	0.03 ± 0.01	1.30 ± 0.71
	D	nd.	40.81 ± 17.29	0.02 ± 0.01	3.12 ± 0.00
	G	0.01 ± 0.00	14.97 ± 4.98	nd.	36.73 ± 4.72

Henicorhynchus lobatus	M	0.02 ± 0.00	1.62 ± 0.00	0.04 ± 0.01	0.45 ± 3.15
	D	nd.	54.76 ± 0.00	nd.	15.74 ± 6.07
	G	nd.	nd.	0.01 ± 0.00	4.01 ± 0.00
Notopterus notopterus	M	0.11 ± 0.00	2.50 ± 0.43	0.02 ± 0.00	0.60 ± 0.28
	D	0.01 ± 0.01	20.27 ± 23.95	0.01 ± 0.01	3.00 ± 2.39
	G	0.02 ± 0.00	6.68 ± 2.36	0.01 ± 0.00	nd.
Macrognathus siamensis	M	0.12 ± 0.01	1.94 ± 1.17	No sample	No sample
	D	0.04 ± 0.01	2.36 ± 1.05		
	G	0.03 ± 0.02	2.65 ± 0.51		
Thailand standards[1]			1.00	0.30	
WHO standards[2]			0.50	0.50	

M – muscle, D – digestive tract, G – gill; nd. – non-detected; No sample – the fish species are not observed and caught during the specified season; No tissue – the specified tissue cannot be dissected due to the fish being too thin.

[1]Thailand standards referred to the **Food and Drug Administration, Ministry of Public Health, Thailand, 2020)**.

[2]WHO standards referred to the **World Health Organization (2003)**.

4. Bioconcentration factor (BCF) of cadmium and lead in fish species

The BCF exhibits the accumulation of the chemical in organisms living in the chemical-contaminated area, which refers to the concentration of the chemical in the organism to the concentration in the habitat. This research calculates the BCF based on the heavy metal level in the tissues and the level in water, as shown in Table (5). The calculated BCF values for cadmium ranged from 0.03 to 6.11 and for lead ranged from 1.77 to 17.26 in rainy season. Whereas in dry season, the values for cadmium ranged from 2.00 to 8.00 and for lead ranged from 0.67 to 10.68. The BCF of lead tends to be higher than that of cadmium, referring that fish can accumulate more lead than cadmium. This finding is consistent with the report by **Tanee *et al.* (2017)**. However, to date, there is no set of standard level for BCF values.

Table 5. Bioconcentration factor (BCF) of cadmium and lead in fish species collected during rainy and dry seasons calculated from the data in Table (4)

Fish species	**Bioconcentration factor (BCF)**			
	Rainy		**Dry**	
	Cadmium	**Lead**	**Cadmium**	**Lead**
Puntius leiacanthus	0.97[a]	2.06[a]	4.00[b]	2.34[a]
Oxyeleotris marmorata	2.87[a]	2.95[a]	8.00[b]	3.56[a]
Anabas testudineus	3.63[a]	5.89[a]	3.00[a]	7.12[a]
Trichopodus trichopterus	2.70[a]	12.37[a]	5.00[b]	5.79[b]
Hampala dispar	4.55[a]	7.86[a]	2.00[a]	2.89[b]
Hemibagrus filamentus	3.04	8.84	No sample	No sample
Dermogenys siamensis	1.08	11.78	No sample	No sample
Channa striata	No sample	No sample	2.00	10.68
Labiobarbus siamensis	1.56[a]	7.66[a]	3.00[b]	5.17[a]
Clarias batrachus	1.52[a]	1.77[a]	4.00[b]	5.34[b]
Oreochromis niloticus	No sample	No sample	3.00	4.67
Tetraodon leiurus	2.55[a]	Nd.	4.00[b]	1.65

Parambassis siamensis	1.74[a]	17.26[a]	5.00[b]	9.68[a]
Anematichthys apogon	0.30[a]	2.36[a]	3.00[b]	0.67[a]
Henicorhynchus siamensis	0.76[a]	6.59[a]	3.00[b]	4.34[a]
Henicorhynchus lobatus	1.56[a]	4.78[a]	4.00[b]	1.51[a]
Notopterus notopterus	5.52[a]	7.35[a]	2.00[b]	2.00[b]
Macrognathus siamensis	6.11	5.70	No sample	No sample
Average	2.53	6.58	3.67	4.49

a,b – Different letters refer to significantly different values ($P< 0.05$); nd. – non-detected; No sample – the fish species is not observed and caught during the specified season

5. Potential ecological risk index (PERI)

The PERI of cadmium ranged from 177.83 to 425.53 and 154.55 to 563.14 in rainy and dry season, respectively, which are in high to very high risk. Contrarily, the PERI of lead ranged from 0.01 to 0.05 in rainy and dry season, which are in low risk. PERI of the metals in sediment collected in rainy season from 7 sites ranged from 150.33 to 425.55 and averaged at 299.88, which is considered at a high risk level. Accordingly, the PERI for dry season samples ranged from 154.57 to 563.16, and averaged at 311.59, which is also considered at a high risk level, as exhibited in Table (6). The PERI was successfully applied to evaluate the ecological risk in various areas such as harbor **(Guo *et al*., 2010)** and rivers **(Poopa *et al*., 2015; Liu *et al*., 2021)**.

Table 6. Risk assessment of environment in the Huai Kho Reservoir calculated from the data in Table (4)

Site	Rainy						Dry					
	Cadmium		Lead		PERI	Level	Cadmium		Lead		PERI	Level
	E_r^i	Level	E_r^i	Level			E_r^i	Level	E_r^i	Level		
1	365.19	Disastrous	0.01	Low	365.20	High	154.55	Very high	0.02	Low	154.57	Moderate
2	357.78	Disastrous	0.01	Low	357.80	High	563.14	Very high	0.02	Low	563.16	Very high
3	425.53	Disastrous	0.02	Low	425.55	Very high	208.53	Very high	0.02	Low	208.55	High
4	327.09	Disastrous	0.02	Low	327.11	High	256.87	Disastrous	0.03	Low	256.90	High
5	177.83	Very high	0.02	Low	177.85	Moderate	358.49	Disastrous	0.03	Low	358.52	High
6	150.31	Very high	0.02	Low	150.33	Moderate	316.15	Disastrous	0.04	Low	316.19	High
7	295.33	Disastrous	0.03	Low	295.36	High	323.21	Disastrous	0.05	Low	323.25	High
Average					299.88	High	Average				311.59	High

E_r^i is potential ecological risk index of an individual metal; PERI refers to potential ecological risk index, calculated based on the formula by **Hakanson (1980)** and **Liu *et al*. (2021)**.

6. Quantitative health risk assessment

Fish muscles are a primary food source for the human population. Hence, we utilized metal concentrations in fish muscles (Table 4) to assess human health risks via the tolerable daily intake (TDI) of metals. In this study, the TDI represents the estimated maximum intake of Pb that an individual is expected to receive, considering the potential ingestion rate of fish muscles per day. Fortunately, the TDI levels (Table 7) are below the standard guidelines proposed by **JECFA/WHO (2001)** and **EVM (2003)**. This suggests that fish from the reservoir are currently safe for daily consumption. However, frequent intake may pose health risk; therefore, further observational studies are needed.

Table 7. Tolerable daily intake (TDI) of metals for adults with an average body weight of 70kg, calculated from the data in Table (4)

Fish species	TDI (mg/kg-bw/day)			
	Rainy		Dry	
	Cadmium	Lead	Cadmium	Lead
Puntius leiacanthus	$8.57E^{-05}$	$3.00E^{-03}$	$1.71E^{-04}$	$3.00E^{-03}$
Oxyeleotris marmorata	$2.61E^{-04}$	$4.29E^{-03}$	$3.43E^{-04}$	$4.59E^{-03}$
Anabas testudineus	$3.00E^{-04}$	$8.57E^{-03}$	$1.29E^{-04}$	$9.17E^{-03}$
Trichopodus trichopterus	$2.14E^{-04}$	$1.80E^{-02}$	$2.14E^{-04}$	$7.46E^{-03}$
Hampala dispar	$3.86E^{-04}$	$1.14E^{-02}$	$8.57E^{-05}$	$3.73E^{-03}$
Hemibagrus filamentus	$2.57E^{-04}$	$1.29E^{-02}$	No sample	No sample
Dermogenys siamensis	$8.57E^{-05}$	$1.72E^{-02}$	No sample	No sample
Channa striata	No sample	No sample	$8.57E^{-05}$	$1.38E^{-02}$
Labiobarbus siamensis	$1.29E^{-04}$	$1.11E^{-02}$	$1.29E^{-04}$	$6.64E^{-03}$
Clarias batrachus	$1.29E^{-04}$	$2.57E^{-03}$	$1.71E^{-04}$	$6.86E^{-03}$
Oreochromis niloticus	No sample	No sample	$1.29E^{-04}$	$6.00E^{-03}$
Tetraodon leiurus	$2.14E^{-04}$	nd.	$1.71E^{-04}$	$2.10E^{-03}$
Parambassis siamensis	$1.29E^{-04}$	$2.52E^{-02}$	$2.14E^{-04}$	$1.24E^{-02}$
Anematichthys apogon	$4.29E^{-05}$	$3.43E^{-03}$	$1.29E^{-04}$	$8.57E^{-04}$
Henicorhynchus siamensis	$8.57E^{-05}$	$9.60E^{-03}$	$1.29E^{-04}$	$5.57E^{-03}$
Henicorhynchus lobatus	$8.57E^{-05}$	$6.94E^{-03}$	$1.71E^{-04}$	$1.93E^{-03}$
Notopterus notopterus	$4.71E^{-04}$	$1.07E^{-02}$	$8.57E^{-05}$	$2.57E^{-03}$
Macrognathus siamensis	$5.14E^{-04}$	$8.31E^{-03}$	No sample	No sample
Standard guidelines				
Safe level of food contaminants[1]	2.00			
Safe level for minerals[2]	0.22			

No sample – the fish species are not observed and caught during the specified season; nd. – metals were not detected in the fish muscle.

[1]Safe level of food contaminants referred to **JECFA/WHO (2001)**.

[2]Safe level for minerals referred to **EVM (2003)**.

CONCLUSION

The findings of this research underscore the significant threats posed by heavy metal contamination to both the ecological equilibrium of aquatic environments and human health. Cadmium and lead, particularly, are of priority concern due to their toxicity and adverse effects on human health. The study revealed that cadmium and lead levels in water and sediment of Huai Kho Reservoir exceeded standard limits, indicating the severity of contamination in the area. Additionally, accumulation of lead exceeded limits in 18 fish species, highlighting the potential risks associated with consuming fish from the reservoir. The distribution of cadmium and lead in fish tissues varied between rainy and dry seasons, suggesting the influence of environmental factors on metal accumulation patterns. Although the tolerable daily intake assessment did not exceed standard guidelines, the potential ecological risk index indicated a high-risk level, necessitating urgent regulatory measures to mitigate the impact of heavy metal contamination.

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