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HEALTH RISK ASSESSMENT OF HEAVY METAL IN FISH TO THE POPULATION IN PETAGAS RIVER, SABAH

Muhammad Nur Rashidi Rosli[*1], Madihah Jaafar Sidik[2], Syerrien Shennen Jamiol[2], Mohd Razali Shamsuddin[1], and Nurashikin Abd Azis[3]

[1] Preparatory Center for Science and Technology, Universiti Malaysia Sabah, 88400 Kota Kinabalu, Sabah, Malaysia.
[2] Borneo Marine Research Institute, Universiti Malaysia Sabah, 88400 Kota Kinabalu, Sabah, Malaysia.
[*3] Faculty of Security and Governance, North Borneo University College, Wisma Angkatan Hebat, 1 Borneo, Jalan Sulaman, 88400 Kota Kinabalu, Sabah.

***Correspondence:**
mnrashidirosli@ums.edu.my

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ABSTRACT. *The levels of heavy metals in marine environments and fish are crucial for assessing heavy metal contamination, which poses deleterious effects on communities, especially those in Petagas River. Four randomly caught fish species—Sagor Catfish, Indo-Pacific Tarpon, Spotted Catfish, and Nile Tilapia—from the river were dissected to obtain the fish flesh and prepared following the APHA (American Public Health Association) standard method (2017). The water and fish flesh samples were analyzed using Inductively Coupled Plasma Mass Spectrometry (ICP-MS) to determine the concentrations of potassium (K), arsenic (As), and lead (Pb). The highest average heavy metal concentration in the water sample was potassium (K) 10.04 µg/L, followed by arsenic (As) 0.46 µg/L and lead (Pb) 0.63 µg/L. For the fish samples, the highest average concentration was potassium (K) 14.28 µg/g, followed by arsenic (As) 1.33 µg/g and lead (Pb) 1.14 µg/g. The results were compared with the permissible limits set by the Malaysian Food Act and Regulation (MFAR 1983 & 1985), Malaysian Water Quality Standard (MWQS 2008), and FAO/WHO (1984, 2017). The study revealed that As and Pb concentrations in water exceeded the permissible limits of MWQS (2008) and FAO/WHO (2017), while K concentrations were within the acceptable range. In fish samples, As exceeded the permissible limits of MFAR (1983 & 1985) and FAO/WHO (1984), while Pb exceeded the FAO/WHO (1984) limit but not MFAR. These findings highlight potential health risks to consumers and underscore the need for continuous monitoring of aquatic ecosystems in Petagas River.*

INTRODUCTION

The contamination of fresh and marine waters with wide range of pollutants has become a major concern (Vutukuru, 2005). Rivers, including Petagas River in Sabah, are susceptible to contamination by heavy metals, primarily originating from industrial and agricultural activities, leading to detrimental effects on water, soil, and air quality. Due to their high toxicity and accumulative nature, pollutants released into the environment have a significant impact on the ecological balance of the environment, causing significant harm to aquatic organisms' lives and even mass extinction (Yasir *et al.,* 2008; Rosli *et al.,* 2018; Yamada & Inaba, 2021). According to Yunus *et al.* (2020), the concentrations of these metals in seawater are naturally low, but when organisms accumulate more than they can excrete, there is a high risk of contamination in living tissue.

Additionally, it is often necessary to examine chemical contaminants in food from aquatic sources to understand their level of hazard. The release of pollutants, especially heavy metals, into the aquatic environment is known to have detrimental effects on such an environment and on living organisms, including humans when those pollutants are allowed to enter the food chain. One of the obvious issues is regarding fish accumulate high concentrations of heavy metals beyond the standard limit or permissible level and affect humans via ingestion. Therefore, establishing a dependable database of contaminant levels in readily accessible commercial fish is essential for evaluating the current extent of heavy metal contamination.

Presently, despite the government of Sabah expressing a desire to promote river tourism development, the river remains untapped as a tourism resource. Its advantageous location near Kota Kinabalu city allows for easy market access. There are various opportunities for tourism development, including cultural tourism centred around the Bajau sea gypsy traditions of the local communities, ecotourism highlighting the region's wildlife and scenic waterfront, and adventure tourism focused on river recreation such as canoeing (Younis *et al.,* 2021). Unfortunately, rivers are also occasionally used as a convenient location for the disposal of industrial and human waste, which seriously reduces a river's usefulness for tourism.

In this study, the fish species consumed by the locals are randomly selected by fishing or using gill net and water samples also will be collected at the Petagas River, Penampang, Sabah. There were four fish samples and three water samples that were analysed using the inductively coupled-plasma mass spectrometry (ICP-MS) to identify the heavy metal concentration that affecting the level of metal contaminants. As a result, this study is used to evaluate the selected heavy metal concentrations in fish flesh and spatial variation Arsenic (As), Lead (Pb) and Potassium (K) in relation to the maximum residual limit for human consumption.

MATERIALS AND METHODS

Sample Collection

The sampling site located at Petagas River (5°54'45"N 116°3'42"E) and it takes 8.8 km to the state capital of Kota Kinabalu as shown in Figure 1. It is one among several rivers in Sabah that holds potential for ecotourism. The fish samples collected from the Petagas River encompass a selection of species, including *Arius maculatus* (Spotted Catfish), popularly referred to as "Ikan Duri Tompok," *Hexanematichthys sagor* (Sagor Catfish), also known as "Ikan Belukang," *Mugil cephalus* (River Mullet), commonly recognized as "Ikan Belanak," *Megalops cyprinoides* (Indo – Pacific Tarpon), widely known as "Ikan Bulan," *Cyclichthys spilostylus* (Puffer Fish), known as "Ikan Buntal," and *Oreochromis niloticus* (Nile Tilapia), which bears the common name "Ikan Tilapia." All these fish species are abundantly found in the Petagas River. The fish samples will be catch, packed, labelled and then taken to the BMRI laboratory for cleaning and isolation of the fish flesh.

This study was conducted from March 2023 until February 2024, where the collection of fish samples and water samples was conducted in three sampling stations (Station 1: Downstream, Station 2: Midstream, Station 3: Upstream) at Petagas River. This sampling was a one-time visit to achieve the study's objectives of assessing the heavy metal concentration in the Petagas River. To prioritize safety and sample integrity, sample collection was carefully planned out and executed in favorable weather conditions and tide status. Thus, the sampling took place in August during low tides when the weather was suitable to allow easy access for the boat to the designated three sampling stations in the Petagas River.

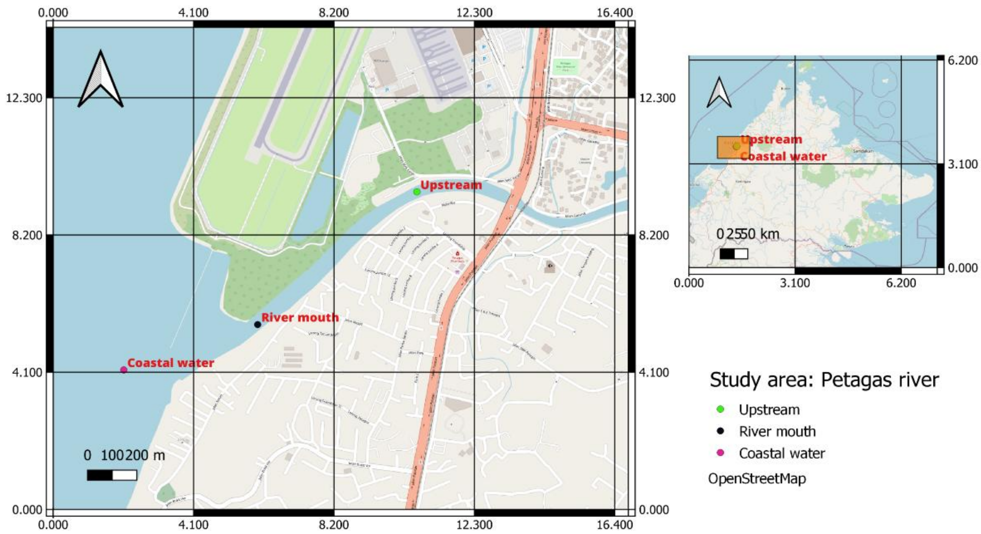

Figure 1. The map of Petagas River using QGIS 3.28.

Sample Treatment

The fish specimens were subjected to a thorough washing process using distilled water to eliminate any residual water. Subsequently, the head, viscera, gills, and flesh of the fish were separated. The fish flesh was dried in an oven at 105 °C for 24 hours or until the weight is constant. This method involving drying of fish flesh using oven also ensured all the water content was completely removed and to avoid residual unreacted fats, before proceeding with wet digestion. After that, the fish samples were milled with mortar and pestle to obtain fine grains of each sample of about size around 500 µm. A total of 0.1 g was mixed with 4.0 mL nitric acid, HNO_3 (65% analytical grade) and 6.0 mL hydrochloric acid, HCl. The mixed samples underwent the ICP-MS digestion method using microwave oven for 55 minutes until the solution was clear. Subsequently, the resulting solutions were filtered, and distilled water was added to bring the sample solution volumes to 100 mL, following the methodology outlined by Yasir *et al.* (2008).

Samples Analysis

The prepared samples, in the form of solutions, were subjected to analysis using Inductive Coupled Plasma-Mass Spectrometry (ICP-MS). The sample prepared following the APHA standard method (American Public Health Association, 2017). Accuracy and precision were validated using Standard Reference Material (SRM MA-A-2 Fish Flesh Homogenate) with recoveries between 93–106%. (Yasir *et al.,* 2008; Rosli *et al.,* 2018). To ensure accurate measurements and eliminate variations arising from differences in moisture content within organisms, the heavy metal concentrations in the tissues were calculated based on their dry weights. Additionally, control measures were implemented, including the use of blanks and replicates, to monitor and validate the precision and accuracy of the analytical procedure.

RESULTS AND DISCUSSION

Heavy Metal Content of Arsenic (As), Lead (Pb), Potassium (K) in the Fish Sample

Table 1 presents the metal contents in fish flesh from various species, including Sagor Catfish (downstream), Indo-Pacific Tarpon and Spotted Catfish (midstream), and Nile Tilapia (upstream). However, many factors, including gender, age, size, reproductive cycle, swimming pattern, feeding behaviour, and geographical location, can influence metal uptake and accumulation (Abdel-Baki *et al.,* 2011). Furthermore, different metal affinities to fish tissues, as well as different uptake, deposition, and excretion rates, result in varying levels of bioaccumulation in the fish body. Food consumption is the most likely primary route of exposure to trace elements among the various modes of metal accumulation (ingestion, inhalation, and skin contact) for the great majority of people (Ahmed *et al.,* 2019). Fish are advantageous as bioindicators because they are long-lived and incorporate fluctuations in pollutants over time, allowing for continuous monitoring of the presence of pollutants while also allowing for spatial integration of pollutant data, and they are easily sampled (Ayodele *et al.,* 2020). The metal content of fish flesh was measured due to its importance in human consumption (Taweel *et al.,* 2013). The average metal concentration in fish flesh was K (14.28 µg/g), followed by As (1.33 µg/g) and Pb (1.14 µg/g).

Table 1. Heavy metal concentration in samples.

Station	Species	Heavy metal concentration in samples (µg/g)		
		As	Pb	K
Downstream	Sagor Catfish	1.38 ± 0.12	1.71± 0.49	14.68± 0.76
Midstream	Indo-Pacific Tarpon	1.25 ± 0.11	0.66± 0.33	13.45± 0.71
	Spotted Catfish	1.20 ± 0.11	0.91± 0.48	13.87± 0.71
Upstream	Nile Talapia	1.47 ± 0.13	1.29± 0.46	15.11± 0.78
Mean heavy metal concentrations by fish species ± SD		1.33 ± 0.12	1.14 ± 0.46	14.28 ± 0.76
Permissible limit MFAR (1983 & 1985)		1.0	2.0	nd

In this study, the Sagor Catfish accumulated the highest Pb concentration (1.71 µg/g) and was also high in K (14.68 µg/g). The Indo-Pacific Tarpon was found to have high concentrations of As (1.25 µg/g) and K (13.45 µg/g). The Spotted Catfish contained high concentrations of K (13.87 µg/g) and As (1.20 µg/g). The Nile Tilapia had the highest concentration of K (15.11 µg/g) and As (1.47 µg/g) compared to other fish species. According to the permissible limit established by FAO/WHO (1984), the K concentration exceeded the standard limit (1.6 - 2.0 µg/g), however Malaysia's national guidelines did not publish data on the limit for K concentration. All fish species contain high concentrations of K because it is necessary for sustaining normal body growth, enhancing fish protein, building muscle, regulating electrical conductivity, and preserving the acid-base balance (Ayodele *et al.,* 2020). On the other hand, even at relatively low concentrations, ingesting certain others, like Pb and As, can be extremely harmful to humans. Table 1 shows that Nile Tilapia had the highest As concentration (1.47 µg/g), followed by Sagor Catfish (1.38 µg/g), Indo-Pacific Tarpon (1.25 µg/g), and Spotted Catfish (1.21 µg/g), all exceeding the permissible limit set by MFAR (1983 & 1985) and FAO/WHO (1984). Inorganic arsenic is more lethal than organic arsenic and can cause cancer in humans if consumed over time (Abdel-Baki *et al.,* 2011).

The highest Pb concentration was found in Sagor Catfish (1.71 µg/g), followed by Nile Tilapia (1.29 µg/g), Spotted Catfish (0.91 µg/g), and Indo-Pacific Tarpon (0.66 µg/g). Based on the findings, the range of Pb concentrations in each species did not pose a threat to human consumption within the permissible limits set by MFAR (1983 & 1985). However, it is safe to be aware of the risk of metal contamination when eating these fish because the Pb concentration has exceeded the permissible limit set by FAO/WHO (1984) of 0.2 µg/g. This is also because lead is one of the most common pollutants in the environment, occurring naturally in rocks, soils, and the hydrosphere. Pb is quickly absorbed by fish after it is released into the marine environment and builds up in their body tissues, bones, gills, kidneys, liver, and scales (Md Yunus *et al.,* 2014). The findings of this study revealed that different fish species accumulated varying Pb concentrations.

According to Praveena and Lin (2015), heavy metal residues in contaminated environments typically build up in microorganisms, aquatic fauna, and flora. Heavy metals enter rivers through a variety of natural and anthropogenic sources (Saher & Kanwal, 2019). Fish bioaccumulation of heavy metals is influenced by a variety of factors, the most important of which are feeding behavior, growth rate, temperature, hardness, salinity, age, sex, and metal interactions (Sivaperumal *et al.,* 2007; Sow *et al.,* 2019). As a result, heavy metals' uptake by long-term contaminated organisms causes severe diseases such as food poisoning, liver damage, cardiovascular disorders, and even death (Tair & Eduin*,* 2018;

Zulkipli *et al.,* 2021). Furthermore, because fish are abundant and susceptible to accumulating trace elements, they serve as bioindicators for assessing the status of the aquatic ecosystem.

Heavy Metal Content of Arsenic (As), Lead (Pb), Potassium (K) in the Water Sample

Results of heavy metal concentration in water were compared with the permissible limit that have been set by the Malaysian Interim Water Quality Standard (NWQS) (2008) and World Health Organization (WHO) and the Food and Agriculture Organization (FAO) for arsenic (As), lead (Pb), and potassium (K) in microgram per gram (µg/L) in Table 2. Based on the permissible limit set by NWQS (2008) and FAO/WHO (2017) in milligrams per litre (mg/L) unit for water sample, the As concentration in downstream (0.39 µg/L), midstream (0.56 µg/L) and upstream (0.42 µg/L) and Pb concentration in downstream (1.10 µg/L), midstream (0.32 µg/L) and upstream (0.48 µg/L) exceeded the permissible limit (0.05 µg/L) and (0.01 µg/L) for both heavy metal elements. The K concentration of all three sampling sites did not exceed the permissible limit set by FAO/WHO (12.0 µg/L) as shown for downstream (8.10 µg/L), midstream (11.49 µg/L), and upstream (10.52 µg/L), respectively.

Table 2. Heavy metal sample concentration detected by ICP-MS from a water sample.

Heavy Metal	Heavy Metal concentration (µg/L)			Mean Heavy Metal concentrations by site (µg/L)	Permissible limit NWQS (2008)	Permissible limit FAO/WHO (2017)
	Downstream	Midstream	Upstream			
Arsenic (As)	0.39	0.56	0.42	0.46	0.05	0.01
Lead (Pb)	1.10	0.32	0.48	0.63	0.01	0.01
Potassium (K)	8.10	11.49	10.52	10.04	nd	12.0

Water-Fish Transfer Factor

The water-fish transfer factor was calculated for As, Pb, and K, and if the result is < 1, it means that bioaccumulation of metal (As, Pb, and K) in the fish flesh is not from the water, and otherwise it means that the bioaccumulation of metal in fish is from water (Ayodele *et al.,* 2020). Based on Table 3, 4, and 5 show that all elements were > 1, suggesting bioaccumulation of metal in fish is from the water and pose risk of metal contaminant of humans consuming the fish.

Table 3. Transfer factor of Arsenic (As) between fish and water.

Station	Fish species	$M_{fish\ flesh}$	M_{water}	Transfer Factor (TF)
Downstream	Sagor Catfish	1.38	0.392	3.520
Midstream	Indo-Pacific Tarpon	1.247	0.559	2.230
Midstream	Spotted Catfish	1.204	0.559	2.153
Upstream	Nile Tilapia	1.467	0.423	3.466

Table 4. Transfer factor of Lead (Pb) between fish and water.

Station	Fish species	$M_{fish\ flesh}$	M_{water}	Transfer Factor (TF)
Downstream	Sagor Catfish	1.705	1.095	1.557
Midstream	Indo-Pacific Tarpon	0.659	0.321	2.052
Midstream	Spotted Catfish	0.912	0.321	2.840
Upstream	Nile Tilapia	1.289	0.482	2.674

Table 5. Transfer factor of Potassium (K) between fish and water.

Station	Fish species	$M_{fish\ flesh}$	M_{water}	Transfer Factor (TF)
Downstream	Sagor Catfish	14.679	8.096	1.813
Midstream	Indo-Pacific Tarpon	13.446	11.491	1.170
Midstream	Spotted Catfish	13.869	11.491	1.207
Upstream	Nile Tilapia	15.109	10.523	1.436

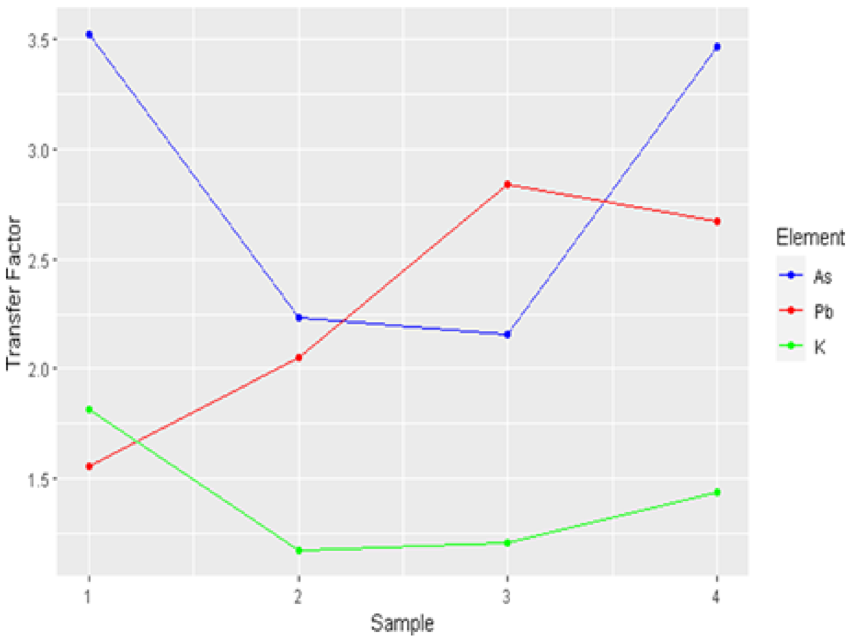

Figure 2. Transfer factor for As, Pb, and K between fish flesh and water.

Figure 2 depicts the amount of transfer factor of three elements (As, Pb, and K) in fish flesh versus water across four samples. The transfer factor determines how much of an element is transferred from the water to the fish tissue. A higher transfer factor indicated that more of the element was accumulated in the fish. Figure 2 shows that As follows, a fluctuating pattern, beginning with a high transfer factor, dropping in the second sample, rising again in the third, and peaking in the fourth. This implies that As uptake by fish is influenced by factors other than water concentration, such as fish species, size, diet, or metabolism. The first sample of Pb had a moderate transfer factor; the second sample showed a slight increase; the third sample showed a significant drop; and the fourth sample showed a sharp rise. This suggested that Pb was readily absorbed by fish from the water and accumulated in their tissue over time. K has a low transfer factor that remains stable across all samples. This suggested that fish regulate K and that there was less As and Pb buildup in their tissue.

According to this study, station 2 (midstream) had the highest concentration of heavy metal K in the water sample. Stations 3 (upstream) and 1 (downstream) had the next-highest concentrations, at 10.52 µg/L, 8.10 µg/L, and 11.49 µg/L, respectively. Pb levels were highest downstream (1.10 µg/L), followed by upstream (0.48 µg/L) and midstream (0.32 µg/L), while As levels were highest in midstream (0.56 µg/L), followed by upstream (0.42 µg/L) and downstream (0.39 µg/L). Overall, all heavy metal elements have exceeded the national and international guideline values established by NWQS (2008) and FAO/WHO (2017), as shown in Table 2, except for K concentration, which remains within the FAO/WHO (2017) standard limit.

Downstream had obvious dense riverside settlements along the Petagas River, providing communities with direct access to the river for economic benefits. Humans primarily contribute to the increase of pollutants that disrupt natural balance, such as organic substances, heavy metals, artificial agricultural fertilizers, detergents, radioactivity, pesticides, inorganic salts, artificially organic chemicals, and wastewater. For example, the houses in the riverside settlements of downstream do not have proper waste system and it will directly discharge in the river along with other solid waste that will increase metal contamination. According to the results, downstream has the highest concentration of Pb (1.10 µg/L), while midstream has the lowest concentration (0.32 µg/L). Based on this value, the Pb concentration exceeded the safe limits for human consumption established by the FAO/WHO (2017) and the NWQS (2008), both of which were set at 0.01 mg/L. According to Kamaruzzaman *et al.* (2011), this could be attributed to heavy metal accumulation at downstream, which was closer to the river mouth.

It is also possible that the presence of industrial and agricultural areas in the upstream contributes to the high Pb concentration at the downstream.

Meanwhile, there were agricultural and residential areas nearby at both stations 2 (midstream) and 3 (upstream). Metal contamination in river water occurs from a variety of sources, including metal-based pesticides, inorganic and organic fertilizers, industrial emissions, and transportation. Inorganic phosphate fertilizers may contain traces of As, Cd, Ni, and Pb (Liu *et al.,* 2015; Mat Amin *et al.,* 2018; Piddocke *et al.,* 2021). Midstream and upstream had the highest As concentrations (0.56 µg/L and 0.42 µg/L) due to their agricultural areas. The As concentration results show that it had exceeded the allowable limits set by FAO/WHO (2017) and NWQS (2008), which were 0.01mg/L and 0.05 mg/L, respectively. Stations 2 and 3 have K concentrations of 11.49 µg/L and 10.053 µg/L, respectively, which fall within the FAO/WHO (2017) standard limit of 12.0 µg/L. The environment is stressed by widespread and unplanned development for hotels, residential areas, deforestation, urbanization, and agriculture, particularly in river systems and water bodies (Pourang, 1995; Kamaruzzaman *et al.,* 2011).

CONCLUSION

The different values of heavy metal concentration in each of the stations correlated to the surrounding environment and human activities. It was found that the mean heavy metal concentration in water sample were higher for K (10.04 µg/L), followed by Pb (0.63 µg/L) and As detected lower (0.46 µg/L). This finding supported by other researchers which K naturally has higher concentration in all the station compared to As and Pb because it is required for fish growth and metabolism. However, the As and Pb concentration are influence by discharge of waste from riverside settlements (downstream) and agricultural area in midstream and upstream which uses arsenic or phosphate-based fertilizer and pesticide. The mean concentrations of heavy metals in randomly collected fish species were investigated, revealing higher levels of K (14.28 µg/g), followed by As (1.33 µg/g) and Pb (1.14 µg/g). Intrinsic factors like species, age, size, metabolic demand, and extrinsic factors such as heavy metal type, location, and environmental conditions were noted as influencing factors on the heavy metal content in various fish species. Overall, the results for heavy metals in water samples were compared to the permissible limits established by the NWQS (2008) and WHO/FAO. It shows that only the K concentration did not exceed the permissible limit of 12.0 mg/L. Meanwhile, the fish sample results were compared to the permissible limits set by MFAR (1983 & 1985) and FAO/WHO (1984). The As concentration has exceeded the permissible limit under both national and international guidelines. It is advised that the public exercise caution when consuming fish from the Petagas River because the Pb concentration was still below the FAO/WHO (1984) limit but above the MFAR (1983 & 1985) permissible limit of 2.0 µg/g. Although the permissible limit set by MFAR (1983 & 1985) did not include a limit for K concentration, caution should still be employed. As a result, there is a need to closely monitor industrial effluent and the wastewater system in the coastal area, as well as develop various strategies to prevent heavy metal accumulation in seafood, which may eventually reduce the chronic health risk to the exposed consumer. Nonetheless, additional research is necessary to guarantee that the same conclusions are made.

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