Level of Zinc and Lead in Freshwater Fishes in Balok River, Pahang, Malaysia

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Abstract — This study was to determine the level of Zinc (Zn) and Lead (Pb) in the muscle and gills of fishes in Balok River, Pahang, Malaysia. Four different species of fishes: *Megalops cyprinoides, Johnius trachycephalus, Lates calcarifer and Hexanematichthys sagor* were digested using acid digestion method and analysed by Inductive Coupled Plasma Micro Spectrometer (ICP-MS). Pb levels in muscle among all fish species occurred in the rank order: *H. sagor > L. calcarifer>M. cyprinoides> J. trachycephalus* meanwhile Pb level in gills: *Johnius trachycephalus> L. calcarifer> H. sagor > M. cyprinoides*. Pb concentration in muscle of fish was the highest in *H. sagor* (0.037±0.037mg/kg) and in gill of *J. trachycephalus* with value of 0.213±0.273mg/kg respectively. Meanwhile Zn level among all fishes were in order of: *H.sagor> J.trachycephalus> L.calcarifer> M.cyprinoides.* The mean concentration of Zn in muscle and gill of *H.sagor* was the highest (2.400±1.677 and 31.572±4.562mg/kg) compared to other species respectively. However, the concentration of Pb and Zn in all fish samples were below permissible limit according to Malaysia Food Act (MFA, 1983), US Food and Drug Administration (USFDA, 1993), European Commission (EC, 2001), Food and Agriculture Organization (FAO, 2012) and were estimated safe for human consumption.

Keywords — heavy metals, lead, zinc, freshwater fishes

I. INTRODUCTION

Among the animal species, fish are inhabitants that cannot escape the detrimental effects of these pollutants [1]. Besides that, it has high economical values, thus fish is suitable for water quality symbol and easy to be interpreted by public. Fish can response to environmental changes thus it can be used for pollution indicator study. Fish is a good bio-indicator because it is easy to be obtained in large quantity, potential to accumulate metals, long lifespan, optimum size for analysis and easy to be sampled [2].

All trace metal is toxic at some bioavailability. The natural levels of heavy metals in the environment had never been a threat to health but in the recent years increased industrial activities leading to air born emissions, auto exhausts, effluents from industries as well as solid waste dumping have become the sources of large quantities of heavy metals into the environment [3].

Metal like Pb and Zn in small amount is essential for the metabolic process and are assimilated by organisms over. These metals will stay permanently and do not degrade from the environment. It was important for the consumer to know where the fish come from to confirm whether it was grow in a healthy environment or in contaminated water. In contaminated water, there is a high risk for the fish to have absorbed the heavy metal from the environment.

Human activities such as industrial processes, domestic wastes, agricultural activities and emissions from vehicles and factory plants are the main sources of some heavy metals enters and deposited into the environment. From a recent study, it was confirmed that the river in Kuantan has been polluted with some metals including Pb, Co, Cu, Cd and As [4].

II. MATERIAL AND METHOD

Fish Collection

The fishes were caught from each sampling stations by using fishing net and placed in polyethylene bags and immediately stored in ice box with ice. The fishes were deep frozen in the cool room at -20 ˚C until ready for analysis.

Dissection

The fish samples were thawed at room temperature. Before dissection, the fish samples were rinsed with ultrapure water to remove foreign particles such as debris and other external adherent.

The length (mm) and weight (g) of each samples were measured and recorded. The fish samples were dissected for analysis using stainless steel scalpels and knife.

The dorsal part of the fish tissues was dissected and weighted at 10 g per samples. The samples were dried in an oven at 80 ˚C for overnight until completely dry. The samples were allowed to cool in desiccator. The dry weight of the fish samples were measured.

Fish Sample Analysis

Acid digestion method [5] was perform to digest the fish samples. Each dried tissues were put in a test tube. 10 mL of 70% nitric acid (HNO_3) were added into each test tube and they were left in room temperature for overnight. On the second day, the samples were digested in block thermostat at 100 ˚C for 2 hours. Then, the samples were allowed to cool. Next, 2 mL of 30% hydrogen peroxide (H_2O_2) were added to the digestion. The samples were returned to heat again at 100 ˚C for 1 hour and they were left to cool. The digested solutions were filtered through filter paper into a 25 mL volumetric flash and they were diluted to reach 25 mL with deionized water. After the digestion process was done, the samples were analysed using Inductively Coupled Plasma-Mass Spectrometer (ICP-MS).

Metal Calculation

The concentration of heavy metals in fish tissues were calculated by using the formula below [6]:

- $C=A\times B/M$ ……. Eqn. 1
- C= Concentration of test samples (mg/kg)
- A= Reading from ICP (mg/kg)
- B= Volume of final solution (L)
- M= Weight of sample (kg)

Statistical Analysis

The statistics calculation was performed using a computer program Minitab. One way Analysis of Variance (ANOVA) statistical test was done to determine the significance in bioaccumulation of metals in body parts. A $p\leq 0.05$ was consider statistically significant.

III. RESULT AND DISCUSSION

A total of 20 fishes were caught, comprising four species. The captured fish species were listed in Table 1.

Table 1: list of local fishes caught from Balok river

species	Length (cm)	Weight (g)	No of samples
Megalops cyprinoides	30.0-34.0	200-250	4
Johnius trachycephalus	23.0-25.0	180-200	4
Lates calcarifer	26.0-28.0	150-180	4
Hexanematichthys sagor	30.0-34.0	200-600	4

This local species was densely distributed along Balok river and each of them do have the same physical properties including feeding behavior, body length and habitats.

The findings recorded regarding levels of Pb and Zn found in local fish are presented in Table 2. It was revealed that Pb and Zn concentrations ranged as follows: 0.01-0.037mg/kg of Pb in muscle and 0.021-0.213mg/kg in gills, whereas 0.434-2.400mg/kg of Zn in muscle and 3.351-31.572 mg/kg of Zn in gills.

The mean concentration of Pb in meat was in order of *H. sagor > L. calcarifer>M. cyprinoides> J. trachycephalus.* Pb concentration in muscle of fish was the highest in *H. sagor* (0.037±0.037mg/kg) although that was the highest value, it was still below permissible limit according to [7-10]. *J. trachycephalus* had the lowest Pb level in muscle (0.008±0.004mg/kg) and *M. cyprinoides* have least concentration of Pb in gill with value of 0.021mg/kg. The accumulation of Pb in species *M. cyprinoides* in gill was low compared to other species might be due to their feeding behaviour and habitat [11]. Overall, Pb concentrations in all species were below permissible limit and safe to be consumed. Trace metals such as Pb will interfere with essential nutrients of similar characteristics such as Zn and Copper.

The concentration of Pb in gill for *M. cyprinoides* in this study was lower compared to [12] and concentration of Pb in muscle of *H.sagor* was also lower compared to [12]. However, previous study by [13] show that the highest value of Pb in *L. calcarifer* was 60.97 mg/kg. Compared to this present study, the value had big different and it had pass from permissible limit indicates that the species are dangerous to consume. Besides that, metals Pb in *H. sagor* from previous study done by [14] and [15] study was lower compared with their study. Lead is a cumulative toxicant that affects multiple body system and is particularly harmful to young children. In the aquatic environment, total dissolved lead abundance in water and pore water control primary uptake by organisms. It has been stated by [16] that lead bioaccumulation is primarily dependent on the amount of active lead compounds in the environment and the capacity of animal species to store lead. High concentration of Pb in the body can cause death or permanent damage to the central nervous system, brain and kidneys [17]. Pb occurrence in Balok River might be due to runoffs from the nearby busy road and emissions from heavy traffic from bauxite lorry.

Table 2: concentration of Pb and Zn in mg/kg among all species

	Pb		Zn	
This study	Meat	Gill	Meat	Gill
M.cyprinoides	0.010±0.002	0.021±0.002	0.434±0.002	4.379±0.001
J.trachycephalus	0.008±0.004	0.213±0.273	1.500±1.219	3.351±0.516
L.calcarifer	0.031±0.001	0.137±0.098	1.089±0.210	4.680±2.208
H.sagor	0.037±0.030	0.054±0.001	2.400±1.677	31.572±4.562
Previous study				
Widad et al, (2013), Penang	0.23 ± 0.05	0.23 ± 0.05	1.35 ± 0.19	1.84 ± 0.27
Adawiyah, (2016) Perak River	2.42±0.83	-	36.43±3.15	-
Kamaruzzaman et al, (2010) Pahang River	2.37	-	77.74	-

Mean concentration of Zn among all fishes were in order:*H.sagor*>*J.trachycephalus*>L.calcarifer>M.cyprinoides. The mean concentration of Zn in muscle and gill of *H.sagor* was the highest (2.400±1.677 and 31.572±4.562mg/kg) compared to other species respectively. *M. cyprinoides* had the lowest concentration of Zn in meat with value 0.434 mg/kg and and the lowest of Zn in gill of *J.trachycephalus*. The variation of heavy metal concentration among fish species could be due to heavy metal intakes by fish in each different aquatic environment depends on ecological requirements, metabolisms and other factors such as salinity, water pollution level, food and sediment [18]. Zn is one of heavy metal that are needed for human consume but can cause harm when exceeded the level permitted. The concentration of Zn in muscle and gills of all fish species caught along Balok River were below permissible limit of [7] and [10] however gill of *H. sagor* exceed value 30mg/kg for Zn established safe limit of heavy metals in fish by [8]. However, gill is not organ that consumable by human, so these fish are still safe to be consumed.

H.sagor had the highest Zn level in the organs may be due to their feeding habits. *H.sagor* are known as omnivorous fish as other fish samples are carnivores. Results of this study were in agreement with report of [19] and [20] in which pelagic fish (omnivorous/herbivore) recorded higher metals concentrations than the benthic fish (carnivore) (Table 2). Although fish are mostly migratory and seldom settle in one location, metal accumulation in fish organs gives evidences of exposure to heavy metal pollution in water.

The study also showed the concentration of Zn was higher than Pb in fishes. According to [21], Zn element is always found abundant in marine organisms. Furthermore, Zn is one of the essential elements required by aquatic organisms for their metabolic processes [22].

The concentration of heavy metals in fish is related to several factors such as the food habits and foraging behaviours of the organism [23]. Physical and chemical properties of the water will affect the concentration and accumulation of heavy metals in the body of the fish [24] and the adaptation capacity of the fish to heavy metal load [25].

According to [26], compared to several other metal ions with similar chemical properties, zinc is relatively harmless. There is little information about the toxicity of zinc exposure. Chronic exposure of zinc leads to anaemia. Only exposure to high doses has toxic effects, making acute zinc intoxication a rare event. Zn also effects tissue respiration in some fishes; hypoxia; accrues structural damage affecting the growth, development and survival of fish; affects hatchability, relatively harmless; long-term and high-dose interferes with the copper uptake [27]. According to Table 2, it was confirmed that gill tend to taken up more heavy metal than edible meat. This is because gills act as water filter and had a direct contact with the polluted water and may be the main route for the contaminants to enter the fish body. Then with the blood flow they can reach other internal organs and tissues. Gill is basically main sites of deposition due to their cellular configuration [28].

IV. CONCLUSION

From this study, it can be seen that variations among concentrations of heavy metal in different species of fish and showed different affinity capabilities for accumulation. Metal concentrations were higher in omnivorous fish such as *Hexanematichthys sagor* than carnivorous fish. Pb levels among all fish species occurred in the rank order: H. sagor > L. calcarifer>M. cyprinoides> J. trachycephalus and in gills: Johnius trachycephalus> L. calcarifer> H. sagor > M. cyprinoides. Meanwhile Zn level among all fishes were in order of: H.*sagor*> *J.trachycephalus*> *L.calcarifer*>*M.cyprinoides* respectively. Metal concentration in all fish caught along the Balok River are generally below by Malaysia Food Act (1983), FAO/WHO (2009), USFDA (1993) and EC (2001) standards for essential metals (Zn) and nonessential metal (Pb),hence safe for consumption and do not pose adverse effect to human.

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