2021-Heavy_Metal.pdf

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Heavy metal concentrations in water, sediment and giant mudskipper (*Periophthalmodon schlosseri*) in the coastal wetlands of Kuala Lupak estuary of the Barito River, Indonesia

[1,2]Heri B. Santoso, [1]Hidayaturrahmah, [3]Eko Suhartono, [4]Rizmi Yunita, [5]Danang Biyatmoko, [6]Dewi Anggraini

[1] Department of Biology, Faculty of Mathematics and Natural Sciences, Lambung Mangkurat University, Banjarbaru, South Kalimantan, Indonesia; [2] Doctoral Program of Agricultural Science, Lambung Mangkurat University, Banjarbaru, South Kalimantan, Indonesia; [3] Department of Medical Chemistry/Biochemistry, Faculty of Medicine, Lambung Mangkurat University, Banjarbaru, South Kalimantan, Indonesia; [4] Department of Aquatic Resources Management, Faculty of Fisheries and Marine Sciences, Lambung Mangkurat University, Banjarbaru, South Kalimantan, Indonesia; [5] Department of Animal Science, Faculty of Agriculture, Lambung Mangkurat University, Banjarbaru, South Kalimantan, Indonesia; [6] Department of Statistics, Faculty of Mathematics and Natural Sciences, Lambung Mangkurat University, Banjarbaru, South Kalimantan, Indonesia. Corresponding author: H. B. Santoso, heribudisantoso@ulm.ac.id

Abstract. Pollution seriously threatens wetland habitats, one of the main pollutants coming from heavy metals. The iron (Fe), zinc (Zn), copper (Cu), mercury (Hg), cadmium (Cd) and lead (Pb) were assessed in the water, sediment and in the giant mudskipper fish, *Periophthalmodon schlosseri*. The assessment of heavy metals was carried out by using an atomic absorption reader (AA-6200 AAS Flame Emission Spectrophotometer Shimadzu). The average concentrations of these metals in water decreased as follows: Fe>Pb>Zn>Hg>Cd>Cu, while metals in the sediment samples decreased as follows: Fe>Zn>Cu>Pb>Cd>Hg. Heavy metal concentrations in fish tissue and skin mucus were higher than the concentrations found in water bodies. The highest metal concentrations were found as follows: Fe in the kidney, Zn in the skin mucus, Cu in the kidney, Hg in the skin mucus, Cd in the liver and Pb in the skin mucus. In all fish tissues and skin mucus, Fe concentration was the highest. The bio-water accumulation factors of *P. schlosseri* tissue and skin mucus were substantially higher than the bio-sediment accumulation factors, suggesting that this fish can be utilized as a bioindicator for certain heavy metals in water. Because the coefficients of variation (CV) of heavy metal accumulation in fish tissues vary, these combined three fish tissue types (liver, kidney and skin mucus) have the potential to be used as an instrument to evaluate heavy metal pollutants such as Fe, Zn, Cu, Hg, Pb and Cd. There was a correlation between the heavy metal concentrations in water, sediment, kidney, liver and skin mucus, demonstrating that giant mudskipper fish can accumulate heavy metals in these tissue types.

Key Words: bioaccumulation, bioindicator, heavy metal pollution, skin mucus.

Introduction. Heavy metal pollution in estuarine/coastal swamp ecosystems is still a major issue that threatens biota, water quality and human health globally (Sia Su et al 2013). The negative impact of heavy metal pollutions poses a serious threat to the health of organisms, biological sustainability and function of estuary ecosystems. Heavy metal pollution has a long-term influence, promoting the habitat and biodiversity loss or degradation, and changes in natural resources (Sarah et al 2019; Tabrez et al 2021). These pollutants can disrupt the fish organisms at the molecular, cellular and physiological levels, resulting in negative effects on the population and community levels. Heavy metal pollutions are mostly caused by anthropogenic activities such as industrialization, agriculture and urban development. Heavy metals such as Cu, Zn, Fe, Pb, Cd, Hg and Ag often contaminate estuaries (Marques et al 2019).

The Barito River Estuary is a coastal swamp wetland ecosystem that is highly occupied by coal transportation, loading and unloading activities. The riverside land is used for a variety of anthropogenic activities, including the conversion of mangrove land to ponds the development of industrial areas in sectors such as wood/plywood processing, rubber processing, and the oil palm plantations, with waste disposal potentially polluting the ecosystem (Sofarini et al 2012; Sopiana et al 2018). As a result, it is critical to use a biomonitoring programs to evaluate the health of aquatic ecosystems due to pollution. Biomonitoring programs based on bioindicator instruments need to be developed for early pollution detection and to avoid the negative impacts of heavy metal pollutants on the estuary ecosystem.

There is little information on the biomonitoring program for heavy metal pollution in the Barito River Estuary. Dwiyitno & Ninoek (2008), Sofarini et al (2012) and Fahrrunnisa (2017) carried out monitoring with the water physicochemical method. According to Dwiyitno & Ninoek (2008), heavy metals such as Hg, Pb, Cd and Cu have been found in the water, sediment and fish living in the mouth of the Barito River, but have not exceeded the permissible threshold (PP. No. 82 of 2001 concerning river water quality standards), hence the water and fish are safe for consumption. Except for the chemical oxygen demand (COD) and ammonia levels at several stations, the overall quality of the waters of the Barito River Estuary is still pretty acceptable. Sofarini et al (2012) discovered in 2009-2010 that the water quality of the Barito River Estuary met the quality standard, even though the water body, sediment and shrimp have been lightly polluted by heavy metals Cu, As, Cr, Cd and Pb. Meanwhile, the sediment and the shrimp were moderately polluted by Hg.

A study of Fahrrunnisa (2017) revealed that giant mudskipper fish (*Periophthalmodon schlosseri*) in the coastal swamp area of Kuala Lupak had a Pb concentration of 0.64-0.68 mg kg^{-1} in their gills, skin and flesh, which was higher than the maximum heavy metals limit in food, of 0.03 mg kg^{-1}. The Pb concentration of 0.26-0.31 mg L^{-1} in water bodies also exceeds the quality standard of 0.03 mg L^{-1}, while in sediments it is of 14.61-29.45 mg kg^{-1}, still below the quality standard (<36 mg kg^{-1}). The water quality in Kuala Lupak's coastal swamp area shows that it has been polluted by Pb, posing a threat to *P. schlosseri*.

The biomonitoring program employing *P. schlosseri* as bioindicators, which are commonly found in the region, is a solution for monitoring and evaluating the health of the Barito River Estuary. The sensitive and rapid response of fish in assessing the quality of the aquatic environment makes it suitable as a bioindicator of heavy metal pollution (Kim et al 2017; Rajeshkumar & Li 2018; Salgado et al 2019; Sinha et al 2020; Santoso et al 2020b). The potential of *P. schlosseri* as a bioindicator of heavy metal pollution in coastal swamp environments is still unknown in Indonesia.

P. schlosseri has the potential as a bioindicator in biomonitoring programs in the Barito River Estuary because they are naturally abundant and native to the estuary (Hidayaturrahmah et al 2019). This fish species' characteristics support this point of view: has adaptability and tolerance towards various conditions, it can accumulate varying concentrations of pollutants without suffering fatal consequences, it has benthic living habits, it has a high trophic level in the aquatic food chain, it has a direct exposure to various pollutants, and it has the capability of accumulating heavy metals in their tissues (Zhou et al 2008; Ansari et al 2014; Bertrand et al 2018). *P. schlosseri* are sentinel organisms because they can accumulate heavy metals in their tissues, have a wide geographic distribution and a high sensitivity to environmental pollutants, and they are the dominant species in their habitats. Therefore, the species is suitable for identifying the effects of heavy metal pollution in water bodies and sediment and has a potential to be used for the evaluation of the ecological effects of pollutants on estuary ecosystems (Shirani et al 2012a,b). These fish are at a higher risk of exposure to heavy metals because they live in the mud of coastal intertidal zones, river mouths and mangrove forest floors in tropical and subtropical areas (Ghotbeddin & Roomiani 2020).

Estuary ecosystems are considered as the most degraded habitat types in the world because they are constantly subjected to heavy metal pollution from anthropogenic activities (Ferreira et al 2019; Marques et al 2019), as well as naturally occurring processes such as sedimentation and floods (Barbee et al 2014; Zhang et al 2019). This work aimed at studying the heavy metal content in water, sediment and *P. schlosseri* tissues in the coastal swamp waters, as well as at discovering their potential as heavy metal pollution bioindicators for the biomonitoring programs of the water health in the Barito River Estuary ecosystem. This study is critical because biomonitoring studies on heavy metal pollution in the Barito River Estuary area, South Kalimantan, Indonesia are still limited, although the increasing industrialization and urbanization may have harmed aquatic ecosystems.

Material and Method

Description of the study sites. The Barito River is the largest and longest river in South Kalimantan. The Barito River originates Schwaner Mountains, stretching approximately 1,000 km from the Central Kalimantan region, in the northern part of the island of Borneo, to the river's estuary in the Java Sea. The river has an average width of 650 to 800 m and a depth of 8 m. The river's width in the funnel-shaped estuary reaches 1,000 m, making the Barito River the widest river in Indonesia. The longest section of the Barito River, starting from the upper reaches of the river, is in Central Kalimantan, while the remainder, up to the river mouth, is in South Kalimantan. This river joins the estuary of Negara River before reaching the mouth of the Barito River in the Barito Kuala region. This study was conducted in the estuary waters (station 1) and the coastal waters of Kuala Lupak (station 2), both of which are located along the west coast of the Barito River estuary (figure 1). The description of the research locations at each station is presented in Table 1.

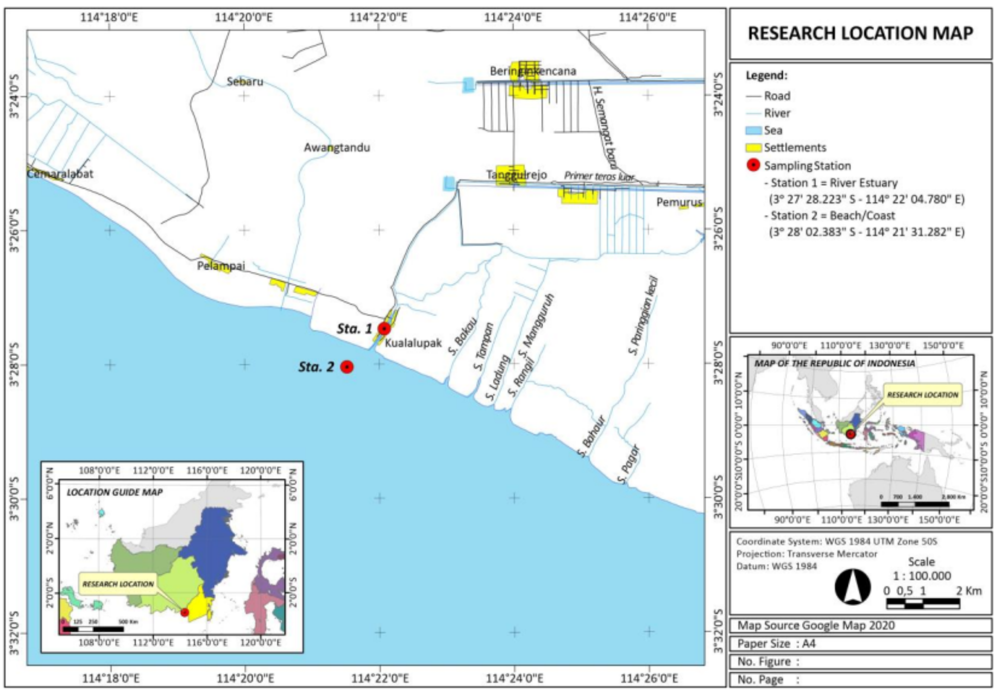

Figure 1. Map of research locations and stations.

Table 1

Description of stations 1 and 2

No	*Station*	*Description*
1	Estuary/River estuary	Kuala Lupak is a Wildlife Reserve Area of the mangrove (swamp forest) ecosystem type. Around the mouth of the Kuala Lupak River, there are residential areas. The Kuala Lupak River flows into the Java Sea. Samples were collected in the estuary waters at the coordinates of 3°27'28.223"S 114°22'04.780"E.
2	Coastal Waters	Kuala Lupak's coastal waters are about 8 km west of the mouth of the Barito River, located on the coast of the Java Sea with a coastline of about 30 km. Mangrove swamp forests may be found around Kuala Lupak's coastline waters. Samples were collected in coastal waters near the river's mouth at the coordinate point of 3°28'02.383"S 114°21'31.282"E.

In the period of 2-3 February, 2021, water, sediment and *P. schlosseri* were sampled using the "purposive sampling" approach, which entails obtaining samples based on various researchers' concerns. The researcher's consideration is based on the source of activities suspected of generating pollutant loads. Collecting samples in estuaries and coastal waters is justified by the residential areas in the region: the water flow of the Barito River is suspected to be polluted by anthropogenic activities. Natural causes, such as landslides, sedimentation, flooding and other natural phenomena are also considered to contribute to the occurrence of pollution. Furthermore, sampling assumes that estuaries and coastal regions are the most sensitive to biodegradation because of anthropogenic activities in the river's upstream and intermediate watersheds, which will have an impact on the estuary area.

Sampling. Water samples were collected from the surface layer at each station using a Ruttner Water Sampler and preserved with 5 mL of 70% nitric acid, before being frozen in the freezer for heavy metal analysis. The heavy metals that were analyzed in water, sediment and fish organs were Pb, Cd, Hg, Fe, Cu and Zn. Sediment samples were collected from each station using the Ekman-Grab Sampler and stored in plastic bags to be dried and analyzed for heavy metal concentration. From each site, 10 *P. schlosseri* of similar size (varying from 17 to 20 cm and weighing 150 to 160 g) were collected. Experienced fishermen captured the fish by hand, by inserting their hands into the fish burrows. Fish from each station were cleaned with water, in order to remove contaminants.

The skin mucus preparation of *P. schlosseri* followed the technique of Fernández-Alacid et al (2018), in which clean fish were placed on a tray, held and the skin mucus was carefully collected with a sterile glass slide by rubbing from the area behind the gills down to the caudal. Two or three times, a sterile slide was gently slid down on both sides of the animal and the skin mucus was carefully pushed and collected in a sterile tube (2 mL). It is not recommended to collect mucus by continually contacting the body's surface, which would provide the greatest volume of mucus, because epidermal lesions may occur and blood or other cells may contaminate the samples. To avoid mucus dilution by the water, this procedure must be followed precisely, without re-wetting the animal and without any contact with the operculum, ventral anal, and caudal fins. The mucus was promptly frozen in liquid nitrogen and kept at -80°C for further analysis.

According to animal welfare standards, the typical treatment for fish before surgery is to place the fish in a container filled with cold/ice water, then decapitate or puncture the pond/brain stem so that the fish's body does not experience pain during incision. The fish were dissected on a polyethylene work surface with stainless instruments, taking care not to contaminate the samples. Each fish's liver and kidney tissues were removed and frozen until metal analyses could be performed. After being dissected to retrieve the skin mucus, liver and kidney organs, the fish are trimmed while undesirable fish leftover is burned in an incinerator.

All procedures were carried out in line with the ethical clearance of the Health Research Ethics Committee No. 549/KPEK-FK ULM/EC/III/2021 Faculty of Medicine, Lambung Mangkurat University, Banjarmasin, Indonesia, which was formed to control and supervise the animal experiments.

Determination of heavy metals. The samples were thawed in the laboratory and dried for 48 hours in a 60°C oven. The dry and wet mass of each sample was recorded to measure the moisture content. In a 100 mL Erlenmeyer flask, 20 mL of concentrated nitric acid (55%) and 10 mL of perchloric acid (70%) were added to roughly 1 g of tissue (dry mass). Digestion was carried out on a hotplate (200-250°C) until the solutions were clear. The solutions were then filtered through 0.45 µm acid-resistant filter paper and diluted to 50 mL each with doubly distilled water. The samples were kept in clean glass bottles until the metal concentration could be analyzed with an atomic absorption reader (AA-6200 AAS Flame Emission Spectrophotometer Shimadzu) (Kotze et al 1999).

Water samples were placed on a hot plate and mixed well with 10 mL of concentrated nitric acid (HNO_3), before being slowly boiled and evaporated till the lowest volume of around 200–100 mL. Before precipitation, another 10 mL of HNO_3 was added, followed by continuous heating until the volume was decreased to 80–100 mL. The samples were then transferred into a 100 mL volumetric flask with 10 mL distilled water and diluted to the mark and filters; the metals were then analyzed (Shaaban et al 2017).

Sediment samples were dried to a constant weight at 80°C (Waykar & Petare 2016) then crushed in Agate mortar. Subsamples of 0.5 g were weighed and 2 mL of concentrated hydrofluoric acid, 2 mL of concentrated HNO_3 and 1 mL of H_2O_2 were mixed in. A Milestone microwave digester was used for digestion. After cooling, the solutions were transferred to 25 mL calibrated flasks and diluted to the mark with deionized water. Following that the latter step, metal concentrations were filtered and analyzed (Shaaban et al 2017).

Pb, Cd, Hg, Fe, Zn and Cu in surface water, sediment, fish tissue and skin mucus were analyzed using an atomic absorption reader (AA-6200 AAS Flame Emission Spectrophotometer Shimadzu).

Analysis methods. Bioaccumulation factor (BF) was calculated to measure the degree of metal accumulation in organism tissues as follows: Bio-water accumulation factor (BWAF) = metals' concentration in fish tissues (mg/kg dry weight)/metals' concentration in water (mg L^{-1}). Bio-sediment accumulation factor (BSAF) = metals' concentration in fish tissues (mg kg^{-1} dry weight)/metals' concentration in sediment (mg kg^{-1} dry weight) (Usero et al 2005; Gawad 2018). The degree of variability of metal concentration in fish tissues is calculated by the Coefficient of variation: CV (%) = Standard deviation x 100/Mean (Yap et al 2003). Descriptive statistics were used to determine the center and spread of data. Meanwhile, Pearson correlation was utilized to calculate the correlation between the metal concentrations in water, sediment and in the liver, kidney and skin mucus of giant mudskipper fish.

Results and Discussion

The concentration of heavy metals in water and sediment. Water and sediment samples from the estuary waters of Barito River (station 1) and coastal waters of Kuala Lupak (station 2) were analyzed for heavy metal concentrations of Fe, Zn, Cu, Hg, Cd and Pb. The average concentrations of heavy metal found in water bodies across all research sites decreased as follows Fe>Pb>Zn>Hg>Cd>Cu. Fe has the highest average concentration, of 7.929 mg L^{-1} (Table 2). The average heavy metal concentrations in the sediment decreased as follows: Fe>Zn>Cu>Pb>Cd>Hg.

Like water bodies, Fe concentration in sediment also has the highest average concentration of 13,704.410 mg kg^{-1} (Table 3). The high Fe concentration is consistent with the findings Supriyantini & Endrawati (2015) who studied water bodies, sediment and green mussel (*Perna viridis*) tissues in the coastal waters of Tanjung Emas Semarang. Haeruddin et al (2020) added that the highest concentration of heavy metals in sediment was detected near the sites of pollution. Estuary and coastal waters polluted with Fe may originate from natural phenomena, such as geological processes like weathering and decomposition of rocks, ore materials and volcanic eruptions that are released into water bodies through runoff, erosion and flooding. Furthermore, Fe is also released by anthropogenic activities such as loading and unloading coal transportation and mining operations (Dalu et al 2020). Anthropogenic activities on land also contribute to the Fe concentration, including: domestic waste, reservoir water, industrial waste deposits and water pipes corrosion carried by rivers to the estuary. The oxidation of iron pyrite (FeS_2) from coal seams produces sulfuric acid and releases iron (Fe^{2+}) (Jaishankar et al 2014).

Table 2

Heavy metals concentrations in water (mg L^{-1})

Heavy metals	*Station 1*			*Average (SD)*	*Station 2*			*Average (SD)*	*Grand average (Grand SD)*
	1	*2*	*3*		*1*	*2*	*3*		
Fe	5.600	5.428	5.900	5.643 (0.239)	11.070	9.505	10.070	10.215 (0.793)	7.929 (2.559)
Zn	0.101	0.073	0.072	0.082 (0.016)	0.122	0.082	0.105	0.103 (0.020)	0.093 (0.020)
Cu	0.027	0.028	0.040	0.032 (0.007)	0.027	0.027	0.030	0.028 (0.002)	0.030 (0.005)
Hg	0.075	0.075	0.075	0.075 (0.000)	0.075	0.075	0.075	0.075 (0.000)	0.075 (0.000)
Cd	0.029	0.035	0.037	0.034 (0.004	0.031	0.033	0.035	0.033 (0.002)	0.033 (0.003)
Pb	0.246	0.275	0.341	0.287 (0.049)	0.197	0.200	0.230	0.209 (0.018)	0.248 (0.054)

Table 3

Heavy metals concentrations in sediment (mg kg^{-1} dry weight)

Heavy metals	*Station 1*			*Average (SD)*	*Station 2*			*Average (SD)*	*Grand average (Grand SD)*
	1	2	3		1	2	3		
Fe	12,978.702	14,818.518	14,696.486	14,164.569 (1028.802)	12,479.876	14,586.872	12,666.006	13,244.251 (116.462)	13,704.410 (1105.315)
Zn	88.481	80.892	79.673	83.015 (4.772)	87.412	80.428	74.291	80.710 (6.565)	81.863 (5.286)
Cu	25.866	44.296	49.521	39.894 (12.427)	25.874	41.463	38.787	35.375 (8.336)	37.635 (9.782)
Hg	0.066	0.017	0.018	0.034 (0.028)	0.065	0.030	0.023	0.039 (0.023)	0.037 (0.023)
Cd	1.464	0.899	0.900	1.088 (0.326)	1.598	0.999	0.991	1.196 (0.348)	1.142 (0.307)
Pb	22.743	25.397	25.359	24.500 (1.521)	21.578	25.277	22.277	23.044 (1.965)	23.772 (1.762)

Heavy metals such as Fe, Pb, Zn, Al and Cu dissolve easily and have a high mobility at pH<5. Fe at low pH will be toxic. Fe concentration >0.3 mg L^{-1} endanger aquatic organisms' lives (WHO 1993). Small amounts of heavy metal contamination can occur naturally and enter aquatic systems through ore-bearing rocks, windblown dust, forest fires and plants. Because heavy metals cannot be degraded, they are deposited, absorbed or incorporated into water, sediment and fish tissues, resulting in heavy metal pollution in water bodies (Malik et al 2010). According to the quality standard of PP RI No. 82 (2001) on the Water Quality Management and Water Pollution Control, the concentration of Fe, Zn, Cu, Hg, Cd and Pb found in water bodies in the present study have exceeded the permissible values (maximum concentrations of Zn, Cu, Hg, Cd and

Pb should not exceed 0.05 mg L^{-1}, 0.02 mg L^{-1}, 0.002 mg L^{-1}, 0.001 mg L^{-1} and 0.03 mg L^{-1}, respectively). Fe concentration limits in water bodies are not required.

In this study, sediment accumulates more heavy metals than water, making it a potential source of heavy metals and the primary storage location for all pollutants (Weber et al 2013). Sediments can hold a wide range of heavy metals in significant and consistent quantities, so they can be used as a reference to determine the state of water pollution (Haeruddin et al 2020). Sedimentation in estuaries retains a considerable amount of metals which are adsorbed on sediment particles and deposited at the bottom. Estuarine sedimentation retains a considerable amount of metals that are adsorbed on sediment particles and deposited at the bottom. Due to the diminishing environmental conditions of the waterways, low dissolved oxygen and high pH, waste containing heavy metals is deposited in the sediment when it enters the estuary, resulting in increasing concentration or enrichment of metals in the sediment (Carvalho Neta et al 2019). The findings of this study are consistent with the findings of Dalu et al (2020), which states that the heavy metal concentration of sediment is substantially higher than that of the water bodies because heavy metals entering the water bodies are adsorbed by suspended particles.

Metals accumulate in sediment from leaded gasoline dumps, chemical manufacturing industries, motorized transportation activities, corrosion of underground pipes, coal-fired thermal power plants and municipal sewage (Jayaprakash et al 2015). The result is also consistent with the findings of Kumar et al (2019), who discovered that heavy metals concentrations in water are always lower than their concentrations in sediment in polluted water bodies because suspended sediment particles adsorb metals from water. Sediment particles containing heavy metals tend to settle to the bottom of water bodies and fish that dwell in mud such as giant mudskipper fish will accumulate heavy metals that enter with food and water.

The concentration of heavy metals in Periophthalmodon schlosseri. Many studies have discovered that indicator species accumulate pollutants in their tissues at a higher rate than their habitats' components, such as water and sediments, and hence have the potential to be employed as a bioindicator instrument in the aquatic ecosystem health biomonitoring programs. The heavy metal concentrations in *P. schlosseri* tissues were higher than the heavy metal concentrations found in water bodies, as shown in Table 4. Table 4 shows the highest concentration of Fe was found in the kidney, the highest Zn in skin mucus, the highest Cu in the kidney, the highest Hg in skin mucus, the highest Cd in the liver and the highest Pb in skin mucus.

In this study, heavy metal concentrations in the liver decrease in the following order: Fe>Zn>Cu>Pb>Hg>Cd. In kidney, heavy metal concentrations decrease as follows: Fe>Zn>Cu>Pb>Hg>Cd, while in skin mucus they decrease as follows: Zn>Fe>Cu>Pb>Hg>Cd. Fe concentration appears to be the highest in all fish tissues. The findings of this study add to the findings of others who have used *P. schlosseri* as a bioindicator instrument of biomonitoring the quality of estuarine and coastal swamp waters at risk for heavy metal and polycyclic aromatic hydrocarbons (PHAs) pollution, by studying heavy metals bioaccumulation in their tissues (Sinaei & Mashinchian 2014; Buhari & Ismail 2016; Moslen & Miebaka 2016; Looi et al 2016; Sarkar & Subrata 2016; Acquavita & Bettoso 2018; Liu et al 2019; Looi et al 2021; Sangur et al 2021).

The high concentration of Fe in fish tissues was produced by the high Fe concentration in sediment and water bodies. These findings are in line with a study of Gawad (2018), which states that high concentrations of heavy metals in sediment and water allow some aquatic organisms to absorb metals biologically and transfer them through the food chain. *P. schlosseri* can accumulate heavy metals in their tissues because they are at the top of the food chain, they dwell in the benthic mud of the coastal intertidal zones and mangrove forest floors and they are directly exposed to pollutants in estuaries. Estuaries according to Ferreira et al (2019) are a type of habitat that is continuously exposed to heavy metal pollutants.

Table 4

Heavy metals concentrations in *Periophthalmodon schlosseri* (mg kg^{-1} dry weight)

Heavy metals	Station 1			Station 2			Grand average (SD)		
	Liver	*Kidney*	*Skin mucus*	*Liver*	*Kidney*	*Skin mucus*	*Liver*	*Kidney*	*Skin mucus*
Fe	4.123	7.998	3.833	5.234	7.900	3.803			
	8.999	11.119	4.999	14.500	18.010	4.879			
	13.111	15.119	6.999	16.345	17.090	6.500			
Average (SD)	8.744 (4.499)	11.412 (3.570)	5.277 (1.601)	12.026 (5.954)	14.333 (5.590)	5.061 (1.358)	10.385 (5.051)	12.873 (4.490)	5.169 (1.333)
Zn	2.750	1.750	11.498	2.898	2.789	11.780			
	4.495	4.373	10.997	7.878	4.400	10.450			
	5.498	3.499	4.248	9.100	3.500	4.300			
Average (SD)	4.248 (1.391)	3.207 (1.336)	8.914 (4.049)	6.625 (3.285)	3.563 (0.807)	8.843 (3.990)	5.437 (2.605)	3.385 (1.006)	8.879 (3.596)
Cu	0.750	4.427	1.767	0.806	4.410	1.705			
	0.500	1.749	0.375	0.989	1.900	0.350			
	0.999	2.499	0.750	0.980	0.980	0.780			
Average (SD)	0.750 (0.250)	2.892 (1.382)	0.964 (0.720)	0.925 (0.103)	2.430 (1.775)	0.945 (0.692)	0.837 (0.196)	2.661 (1.445)	0.955 (0.632)
Hg	0.411	0.301	0.851	0.399	0.299	0.900			
	0.251	0.317	0.257	0.300	0.400	0.190			
	0.432	0.340	0.457	0.460	0.350	0.399			
Average (SD)	0.365 (0.099)	0.319 (0.020)	0.522 (0.302)	0.386 (0.081)	0.350 (0.051)	0.496 (0.365)	0.376 (0,082)	0.335 (0.038)	0.509 (0.300)
Cd	0.357	0.111	0.167	0.356	0.100	0.160			
	0.250	0.250	0.125	0.400	0.280	0.150			
	0.499	0.125	0.250	0.367	0.200	0.230			
Average (SD)	0.369 (0.125)	0.162 (0.077)	0.181 (0.064)	0.374 (0.023)	0.193 (0.090)	0.180 (0.044)	0.372 (0.080)	0.178 (0.077)	0.180 (0.049)
Pb	0.875	0.311	1.000	0.799	0.301	1.011			
	0.375	0.375	0.500	0.378	0.389	0.490			
	0.624	0.625	0.375	0.100	0.710	0.380			
Average (SD)	0.625 (0.250)	0.437 (0.166)	0.625 (0.331)	0.426 (0.352)	0.467 (0.215)	0.627 (0.337)	0.525 (0.294)	0.452 (0.173)	0.626 (0.299)

Bioaccumulation efficiency. The bio-water accumulation factor (BWAF) is a metric that describes the accumulation of certain heavy metals from water bodies, into ecological receptor networks. The bio-sediment accumulation factor (BSAF) is a metric that describes the accumulation of certain heavy metals from sediments into ecological receptor networks (Jayaprakash et al 2015). Table 5 displays the calculated BWAF and BSAF. BWAF values in the liver, kidney, and skin mucus were higher than BSAF values for all metals examined, except for Hg. The results of this study are consistent with the conclusions of Gawad (2018), which state that the presence of heavy metals in water bodies and sediments causes an accumulation process in fish body tissues. Accumulation can occur as a result of the direct absorption of heavy metals in water and in the food chain.

The bioaccumulation of heavy metals in fish tissue is has a negative impact on the ecosystem, being one of the pollution entry points. Bioaccumulation in fish tissues is generally influenced by heavy metal concentrations in water and sediment, diet, fish species, excretion and metabolism. According to Mustafa (2020), the elevated BWAF concentration of all metals in the studied organs indicated that these metals underwent bioaccumulation and biomagnification. These results are in accordance with the findings of Jayaprakash et al (2015) which stated that the heavy metals Ni, Pb, Mn, Co, Cd, Fe and Cu had the highest BWAF concentrations in liver tissue.

The value of the bioaccumulation factor indicates the relationship between the heavy metal concentrations in the water and the heavy metal concentrations in the fish organs. The value of BAF from the environment into fish tissue varies based on parameters such as heavy metal type, heavy metal bioavailability, environmental pollution level, age, species and tissue metabolite status. Heavy metal concentrations in water can also fluctuate after prolonged rain or drought (Mustafa 2020). Zhou et al (2008) claimed that fish can be used as an excellent biomonitoring instrument for estimating heavy metal concentrations in water. The high BSAF value for Hg in fish tissue might be attributed to pollution of Hg bound to sediments, due to mining and tailings left over from mining operations and discharged into the waterways (Lin et al 2010). Mercury accumulates in sediments originating from various physical, chemical, biological, geological and anthropogenic processes that occur in the environment. Direct Hg pollution (point sources) is often caused by Hg mining, gold mining operations, ore refining and mercury recycling goods or processes as well as the chlor-alkali industry (Randall & Chattopadhyay 2013). The values of BWAF and BSAF are considered as potential bio-indicators for monitoring the pollution state of the aquatic environment. The BWAF and BSAF tend to vary based on the structure of the food web, trophic level and life history of an organism (Jayaprakash et al 2015).

Table 5

Bio-Water Accumulation Factor (BWAF), Bio-Sediment Accumulation Factor (BSAF), Coefficient of Variation (CV%) of heavy metal concentration values in liver, kidney and skin mucus of *Periophthalmodon schlosseri*

Heavy metals	*BWAF*			*BSAF*			*CV (%)*		
	Liver	*Kidney*	*Skin mucus*	*Liver*	*Kidney*	*Skin mucus*	*Liver*	*Kidney*	*Skin mucus*
Fe	1.310	1.624	0.652	0.001	0.001	0.000	48.634	34.878	25.789
Zn	58.773	36.596	95.987	0.066	0.041	0.108	47.919	29.720	40.497
Cu	28.067	89.190	31.994	0.022	0.071	0.025	23.396	54.308	66.209
Hg	5.007	4.460	6.787	10.288	9.164	13.945	21.748	11.383	58.933
Cd	11.145	5.330	5.410	0.325	0.156	0.158	21.636	43.199	27.045
Pb	2.116	1.821	2.522	0.022	0.019	0.026	55.978	38.215	47.709

The coefficient of variation (CV) was used in this study to evaluate the degree of variability of metal concentrations in liver, kidney, and skin mucus tissues of giant mudskipper fish. The CV (%) value is used to determine the viability of fish body tissues use as biomonitoring instruments for measuring heavy metal pollution (Yap et al 2003). Table 5 shows the a CV sensitivity to the heavy metal accumulation in fish tissues, indicating that the three fish body tissues have the potential to be used as an instrument to evaluate heavy metal contamination of Fe, Zn, Cu, Hg, Pb and Cd. Based on the CV, the liver is recommended as a potential bioindicator to evaluate water pollution by three heavy metals, namely Pb, Fe and Zn. The kidney is recommended as a potential bioindicator instrument to evaluate water pollution by Cu, Cd and Pb. The skin mucus is recommended as a potential bioindicator instrument to evaluate water pollution by Cu, Hg and Pb. The findings of this study suggest that the simultaneous use of liver, kidney and skin mucus tissue as a potential biomonitoring instrument for long-term contamination of Fe, Zn, Cu, Hg, Pb and Cd in the estuary and coastal waters. However, for conservation and animal welfare, it is preferable to use skin mucus as the best bioindicator instrument, based on non-invasive ways of biomonitoring heavy metal pollution in waters (Bulloch et al 2020).

The relationship between metal contents in water, sediment and in Periophthalmodon schlosseri tissues. Relationships between heavy metal concentrations in water, sediment, tissue, and skin mucus of *P. schlosseri* were calculated and recorded in Table 6. Table 6 demonstrates that there is a significant and positive correlation of 0.98 between the Fe concentrations in the liver and the kidney, indicating that the presence of Fe concentration in the liver tends to increase the Fe concentration in the kidney. In addition, there is also a significant and positive correlation of 0.81, between Fe concentration in the liver and the skin mucus, and 0.73 between the kidney and the skin mucus. According to this study, the accumulation of Fe in the water and sediment increases the Fe concentrations in fish skin tissue and mucus. This is consistent with the high concentration of Fe found in water and sediment.

Table 6

Correlations between the metal concentrations in water and sediment, and those in the liver, kidney and skin mucus of *Periophthalmodon schlosseri*

	Water	*Sediment*	*Liver*	*Kidney*	*Skin mucus*
			Fe		
Water	1				
Sediment	-0.55	1			
Liver	0.24	0.34	1		
Kidney	0.22	0.41	0.98	1	
Skin mucus	-0.12	0.36	0.81	0.73	1
			Zn		
Water	1				
Sediment	0.39	1			
Liver	-0.23	-0.91	1		
Kidney	-0.62	-0.66	0.60	1	
Skin mucus	0.24	0.78	-0.63	-0.19	1
			Cu		
Water	1				
Sediment	0.66	1			
Liver	0.44	0.19	1		
Kidney	-0.22	-0.77	-0.20	1	
Skin mucus	-0.23	-0.88	-0.08	0.89	1
			Hg		
Water	1				
Sediment	N/A	1			
Liver	N/A	0.26	1		
Kidney	N/A	-0.55	-0.20	1	
Skin mucus	N/A	0.89	0.53	-0.77	1
			Cd		
Water	1				
Sediment	-0.87	1			
Liver	0.29	-0.13	1		
Kidney	0.35	-0.68	-0,34	1	
Skin mucus	0.50	-0.27	0.77	-0.38	1
			Pb		
Water	1				
Sediment	0.55	1			
Liver	0.07	-0.24	1		
Kidney	0.44	0.10	-0.64	1	
Skin mucus	-0.45	-0.63	0.81	-0.77	1

There is a significant and negative correlation -0.91 between the concentration of Zn in the sediment and the concentration of Zn in the liver, implying that the increase of the Zn concentration in the sediment tends to decrease the concentration of Zn in the liver. Zn is hypothesized to be utilized as a cofactor by cellular antioxidant enzymes such as superoxide dismutase (SOD), as well as in the activities of other enzymes such as carbonic anhydrase enzymes involved in respiration. As a result, the amount of Zn in the liver is reduced. Furthermore, there is also a significant and positive correlation of 0.78 between the concentration of Zn in the sediment and the concentration of Zn in skin mucus, indicating that the concentration of Zn in the sediment tends to increase the concentration of Zn in the skin mucus. This finding is consistent with the character of *P. schlosseri*, which is more active in mud-shaped sediments, where pollutants in the sediments directly come into contact with skin mucus. These findings are also in accordance with the findings of Buhari & Ismail (2016) which showed a significant and positive correlation between the concentrations of Cu, Zn, Pb, Cd and Ni in several *P. schlosseri* tissues with the metal concentrations in sediment from the West Coast of Peninsular Malaysia, thus indicating the ability of the fish to accumulate heavy metals in their tissues. *P. schlosseri* can accumulate heavy metals in their tissues because they are at the top of the food chain, dwell in the benthic mud of the coastal intertidal zones and mangrove forest floors, and are directly exposed to pollutants in estuaries.

There is a significant and positive correlation of 0.89 between the concentration of Cu in the kidney and the skin mucus, which means that the concentration of Cu in the kidney tends to increase the concentration of Cu in the skin mucus. In addition, there is also a significant and negative correlation between Cu concentrations in sediment and liver (-0.77) and sediment and skin mucus (-0.88). This indicates that the increase in Cu concentration in the sediment tends to decrease the Cu concentration in the liver and skin mucus. In this study, an increase in the Cu concentration in the sediment was linked to an increase in the Cu concentration in the kidney, which was followed by an increase in the concentration of Cu in the skin mucus.

Hg concentration in the sediment and skin mucus has a significant and positive correlation of 0.89, indicating that the concentration of Hg in the sediment tends to increase the concentration of Hg in the skin mucus. Additionally, there is also a significant and negative correlation of -0.77 between Hg concentration in the kidney and skin mucus. The concentration of Cd in the liver and skin mucus has a significant and positive connection of 0.77, indicating that Cd concentration in the liver tends to increase the Cd concentration in skin mucus. In addition, there is a -0.87 correlation between the concentration of Cd in the water and the concentration of Cd in the sediment, indicating that the concentration of Cd in the water tends to decrease the concentration of Cd in the sediment. An increase of the Cd concentration in water produces an increase in the concentration of Cd in the liver, which is followed by an increase in the concentration of Cd in the skin mucus. An increase of the Cd concentration in water bodies isn't always accompanied by increases of the Cd concentration in sediment.

The concentration of Pb in the liver and skin mucus has a significant and positive association of 0.81, indicating that the concentration of Pb in the liver tends to increase the concentration of Pb in the skin mucus. There is also a significant and negative correlation of -0.77 between the concentration of Pb in the kidney and the concentration of Pb in the skin mucus, which implies that as the concentration of Pb in the kidney increases, the concentration of Pb in the skin mucus tends to decrease. An increase of the Pb concentration in the liver was accompanied by a rise of the Pb concentration in the skin mucus, but an increase of the Pb concentration in the kidney did not always lead to an increase in Pb concentration in the skin mucus.

The findings of this study indicate that there is a correlation between the concentration of heavy metals in water, sediment, fish tissue and skin mucus of giant mudskipper fish. Heavy metal concentrations in mud sediment, which serves as a habitat for fish, is positively and significantly correlated with heavy metal concentrations in skin mucus. Following an increase in heavy metal concentrations in the liver and kidney, heavy metal concentrations in skin mucus also increased. This result supports the findings Pandiyan et al (2021) claiming that sediment is a potential source of heavy metals for aquatic biota. Heavy metals from various sources tend to settle in sediments or are released through food chains and food webs, resulting in bioaccumulation and biomagnification in the aquatic ecosystem. The results of this study provide evidence that the giant mudskipper fish has the potential as a bioindicator of heavy metal pollution in estuaries, which is consistent with the findings of Waykar & Petare (2016) stating that the indicator species or sentinel organisms accumulate pollutants in their body tissues at a higher rate than their surroundings.

Heavy metals bioaccumulation in indicator species can reflect metal concentrations in the surrounding water and sediment, making it a useful indicator of environmental quality. The presence of heavy metal concentrations in skin mucus suggests that *P. schlosseri* skin mucus could be used for non-invasive heavy metal pollution biomonitoring in estuaries, in compliance with the conservation plans requiring the preservation of fish species, particularly of endemic, unique and uncommon species (Dzul-Caamal et al 2016). Following this, subsequent research suggests developing innovative non-invasive approaches using fish skin mucus matrix to evaluate fish health and the effects of heavy metal pollution (Santoso et al 2020a; Bulloch et al 2020).

Conclusions. This paper focused on six heavy metals (Fe, Zn, Cu, Hg, Cd and Pb) found in *P. schlosseri*, as well as in the water and sediment from the coastal wetlands of Kuala Lupak Estuary of the Barito River, Indonesia. The giant mudskipper fish lives in estuaries' water and muddy sediments. This species is abundant and easy to collect, which makes it a good candidate for biomonitoring in estuaries and coastal wetlands. The average heavy metal concentrations in water at all research locations decrease as follows: Fe>Pb>Zn>Hg>Cd>Cu. The average heavy metal concentrations in sediments decrease as follows: Fe>Zn>Cu>Pb>Cd>Hg. Heavy metal concentrations in all fish tissues of *P. schlosseri* were higher than the heavy metal concentrations in water bodies. The highest concentration of Fe was found in the kidney; the highest concentration of Zn was found in skin mucus; the highest concentration of Cu was found in the kidney; the highest concentration of Hg was found in skin mucus; the highest concentration of Cd was found in the liver and the highest concentration of Pb was found in skin mucus. In all fish tissues, Fe concentration was the highest. The bio-water accumulation factors of giant mudskippers tissue and skin mucus were significantly higher than the bio-sediment accumulation factors, implying that this species can be used as a bioindicator for certain heavy metals in water. Because the coefficients of variation (CV) of the heavy metal accumulation in fish tissues differ, these three fish body tissues can be used simultaneously to evaluate heavy metal contamination of Fe, Zn, Cu, Hg, Pb and Cd. Based on the CV value, the liver has the potential as a bioindicator of Pb, Fe and Zn pollution in the water. Meanwhile, the kidney has the potential as a bioindicator of Cu, Cd, and Pb pollution in the water. Furthermore, skin mucus has the potential as a bioindicator of Cu, Hg and Pb pollution in the water. Heavy metal concentrations in water, sediment, liver, kidney and skin mucus all had a correlation. Heavy metal concentrations in mud sediment, which serves as a habitat for fish, are positively correlated with heavy metal concentrations in skin mucus. The ability of *P. schlosseri* to accumulate heavy metals in their tissues is demonstrated in this study. This study indicates that giant mudskipper fish can bioaccumulate various heavy metals to varying degrees in their tissues, allowing them to be utilized as biomonitoring agents to evaluate the heavy metal pollution in coastal wetlands. According to the findings of this study, authorities should focus their efforts on protecting coastal wetland ecosystems or estuaries against heavy metal pollution.

Acknowledgements. The authors thankfully acknowledge the Rector of Lambung Mangkurat University for providing research funding, through the PDWM program, in 2021.

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Received: 02 August 2021. Accepted: 08 October 2021. Published online: 20 October 2021.
Authors:
Heri Budi Santoso, Lambung Mangkurat University, Faculty of Mathematics and Natural Sciences, Department of Biology, Banjarbaru, South Kalimantan, Indonesia; Doctoral Program of Agricultural Science, Lambung Mangkurat University, Banjarbaru, South Kalimantan, Indonesia, e-mail: heribudisantoso@ulm.ac.id
Hidatarurrahmah, Lambung Mangkurat University, Faculty of Mathematics and Natural Sciences, Department of Biology, Banjarbaru, South Kalimantan, Indonesia, e-mail: hidayaturrahmah@ulm.ac.id
Eko Suhartono, Lambung Mangkurat University, Faculty of Medicine, Department of Medical Chemistry/Biochemistry, Banjarbaru, South Kalimantan, Indonesia, e-mail: ekoantioxidant@gmail.com
Rizmi Yunita, Lambung Mangkurat University, Faculty of Fisheries and Marine Sciences, Department of Aquatic Resources Management, Banjarbaru, South Kalimantan, Indonesia, e-mail: rizmiyunita@ulm.ac.id
Danang Biyatmoko, Lambung Mangkurat University, Faculty of Agriculture, Department of Animal Science,Banjarbaru, South Kalimantan, Indonesia, e-mail: danangbiyatmoko@ulm.ac.id
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