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Biomonitoring of Heavy Metals (Cu, Zn, Pb, Cd and Ni) in the West Coast of Peninsular Malaysia Using Giant Mudskipper *Periophthalmodon Schlosseri* (Pallas 1770)

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ABSTRACT

The giant mudskipper *Periophthalmodon schlosseri* (Pallas 1770) is the largest notable species of Malaysian mudskippers and invariable the best known example of resident intertidal fish. This paper present an assessment of heavy metals concentration; Cu, Zn, Pb, Cd and Ni in scales, muscle, bone, gills, operculum, intestine, liver and cartilage of *Periophthalmodon schlosseri* (*P. schlosseri*) with aim of proposing it as a biomonitoring agent in the coastal mudflats. The giant mudskippers; *P. schlosseri* were collected from six sampling sites in the west coast of Peninsular Malaysia and analyzed for heavy metals concentrations; Cu, Zn, Pb, Cd and Ni in scales, muscle, bone, gills, operculum, intestine, liver and cartilage by using an air-acetylene flame atomic absorption spectrophotometer, Perkin Elmer Analyst 800. The levels of heavy metals concentrations in the tissues of *P. schlosseri* varied significantly ($P < 0.05$) with exception of Cu in gills ($P > 0.05$). The studied metals showed different accumulation target in the fish; Cu was observed highest in liver while Zn, Pb, Cd and Ni exhibited their highest concentrations in cartilage. The pattern of metal accumulation was in the order: Zn > Pb > Ni > Cu > Cd, with a similar pattern observed for metal selectivity index (MSI %) value. There was no unique ranking with regards to tissue selectivity index (TSI %) among the tissues, except liver which showed high copper selectivity at all the six sampling sites. However, the result of biota-sediment accumulation factor (BSAF) for Cu and Zn were highest in liver and intestine respectively while the BSAF for Pb, Cd and Ni were highest in cartilage. Results of the present study indicate bioaccumulation of the studied metals in the tissues of giant mudskipper *P. schlosseri* which might suggest it use as a potential bio monitoring agent for heavy metals in the west coast of Peninsular Malaysia.

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Keywords

Biomonitoring, Heavy metals, *Periophthalmodon schlosseri*, West coast Peninsular Malaysia.

ABBREVIATIONS

AAS: atomic absorption spectrophotometer; **ANOVA:** one way analysis of variance; **BSAF:** biota sediment accumulation factor; **°C:** degree Celsius; **CB:** Chemical concentration in the biota; **cm:** centimeter; **CRM:** certified reference material; **CS:** chemical concentration in the sediment; **EFLE:** easily freely leachable and exchangeable; **DDT:** dichlorodiphenyltrichloroethane; **DDW:** double distilled water; **d/w:** dry weight; **g:** gram; **GPS:** global positioning system; **HNO3:** Nitric acid; **IAEA:** international atomic energy agency; **kg:** kilo gram; **mm:** millimeter; **ml:** milliliter; **mg/L:** milligram per liter; **MSI:** metal selectivity index; **MT:** metallothionein; **NRCC:** national research council Canada; **ND:** not detectable; **nm:** nanometer; **%:** percentage; ***P. schlosseri*:** *Periophthalmodon schlosseri*; **QA:** quality assurance; **QC:** quality control; **SET:** sequential extraction technique; **TSI:** tissue selectivity index; **µg/g:** microgram per gram; **µm:** micrometer

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INTRODUCTION

Fishes are one of the most important and largest groups of vertebrates in the aquatic system, that are globally accepted as excellent organisms for the study of heavy metals pollution in aquatic environment. They have been used as one of the most indicative organisms for assessing metal pollution in freshwater and marine systems [1]. Fish are relatively situated at the top of the aquatic food chain; therefore, they can normally accumulate heavy metals from food, water and sediments [2, 3]. Bioaccumulation of metals in fish can be considered as an index of metal pollution in the aquatic bodies [4, 5] and a useful tool to study the biological role of metals present at higher concentrations in fish [6, 7]. The use of fish as aquatic bio monitor/bio indicator has increase significantly over years because they are sensitive indicators of aquatic changes and possess high tissues metals accumulation capacity. After incorporation into the fish body, heavy metals were distributed among different tissues via a process that depends on biological needs [8] and subsequently transferred to higher trophic levels via food chain. However, metals amassing in tissues reveal the past exposure of fish via water or food [9-11] or sediment which can serve as pollution indicator.

Mudskippers are peculiar-looking amphibious fish that are characteristic for mangrove forests and mudflats [12]; they are members of the subfamily Oxudercinae, tribe *Periophthalmini*, family Gobiidae (Gobies) [13]. They are the key species of the soft bottom intertidal areas and mangrove swamps of the Indo-west Pacific region and tropical African coast [13].

The unique behavior and ecology of mudskippers make them potential bio monitoring agents. Mudskippers inhabiting the coastal ecosystems faces a direct exposure to most of the heavy metals and metalloids that are discharged from power, thermal, desalination and water treatment plants, and leakage from oil wells in dissolved as well as dietary phases [14], they are requisites of an ideal bio monitor as they are able to accumulate metals that are relatively easy to measure, easy to identify, abundant in the study area, size large enough to provide sufficient tissues for analyses on individuals and possess a stationary behavior and do not migrate [15, 16]. In particular, mudskippers are known to accumulate higher concentrations of some toxic compounds (e.g. DDT and some heavy metals) in their tissues, relative to other aquatic and benthic species [17]. The mudskippers showed a very high potential for bioaccumulation of different types of pollutants from coastal waters and are very important for bio monitoring the coastal ecosystems [14].

The giant mudskipper, *P. schlosseri* is one of the notable species of mudskipper that spends much of its time out of water [13] and the largest Malaysian mudskipper. The unique characteristics features of this fish being a dominant force on the mudflat, unintimidated by snakes and birds that harassed its smaller relatives [18], top predator in the food chain, big size, time dwell in water and intertidal coastal mudflats make them a good and interesting research organisms for the study of heavy metals accumulation and environmental monitoring assessment. In fact, their robustness to environmental stressors and tolerance to many contaminants give them the capacity to be chronically exposed to toxicants without significant acute effects, while their relatively low trophic status makes them less prone to biomagnifying toxicants [17]. Therefore, they were chosen in this study because they could enable continuous surveillance of pollutant presence in water and/ or sediment where they live.

Although, several studies demonstrated that fishes are important biological indicator for investigation of heavy metal contamination and health risk to both animals directly and human upon consumptions of aquatic foods in different parts of the world [19-23] but only few counted number of researches are available on heavy metals concentrations in *P. schlosseri*. Therefore, the objective of this study is: to assess the concentrations of heavy metals (Cu, Zn Cd, Pb and Ni) in scale, muscle, bone, gills, operculum, intestine, liver and cartilage of giant mudskipper P. schlosseri and determine its potential as a biomonitoring organism for heavy metals pollution in the coastal mudflat environment of west coast of Peninsular Malaysia.

MATERIAL AND METHODS

Ethical approval

The authors declare that this research followed the ethical guidance for animal research. The study protocol and ethics were approved by Postgraduate studies committee/ supervisory committee, Department of Biological Science, Universiti Putra Malaysia.

Sample collection and preparation

Surface sediments and giant mudskipper *Periophthalmodon schlosseri* were collected from six sampling sites in the west coast of Peninsular Malaysia (Figure 1). The samplings were conducted in August 2008 at Segantang Garam and Minyak Beku, in September 2008 at Sungai Tiga and Sungai Puluh and in March and June 2010 at Bagan Lalang and Kuala Juru, respectively. The coordinates of the sampling sites were recorded with Global Positioning System (GPS); (Garmin OREGON 45OT 850 MB waterproof GPS). The coordinates and description of each sampling site were given in Table 1.

Top 3 - 5 cm surface sediments [24, 25] were collected in triplicates from three different points within a certain area (approximately 1 meter radius) from each sampling site using plastic scoop and placed in separate labeled polyethylene plastic bags. Each sediment sample was instantly placed in ice and transported to the laboratory until further analysis. Fish were collected using trap net at almost the same locations were the sediments were collected and placed in a plastic aquarium containing some sediment and water. Fish samples were transported to Ecotoxicology laboratory, Department of Biological Sciences, Universiti Putra Malaysia, were stomach and intestines were emptied and dissected immediately or put in labeled plastic bags and kept in deep freeze at –20˚C until further analysis.

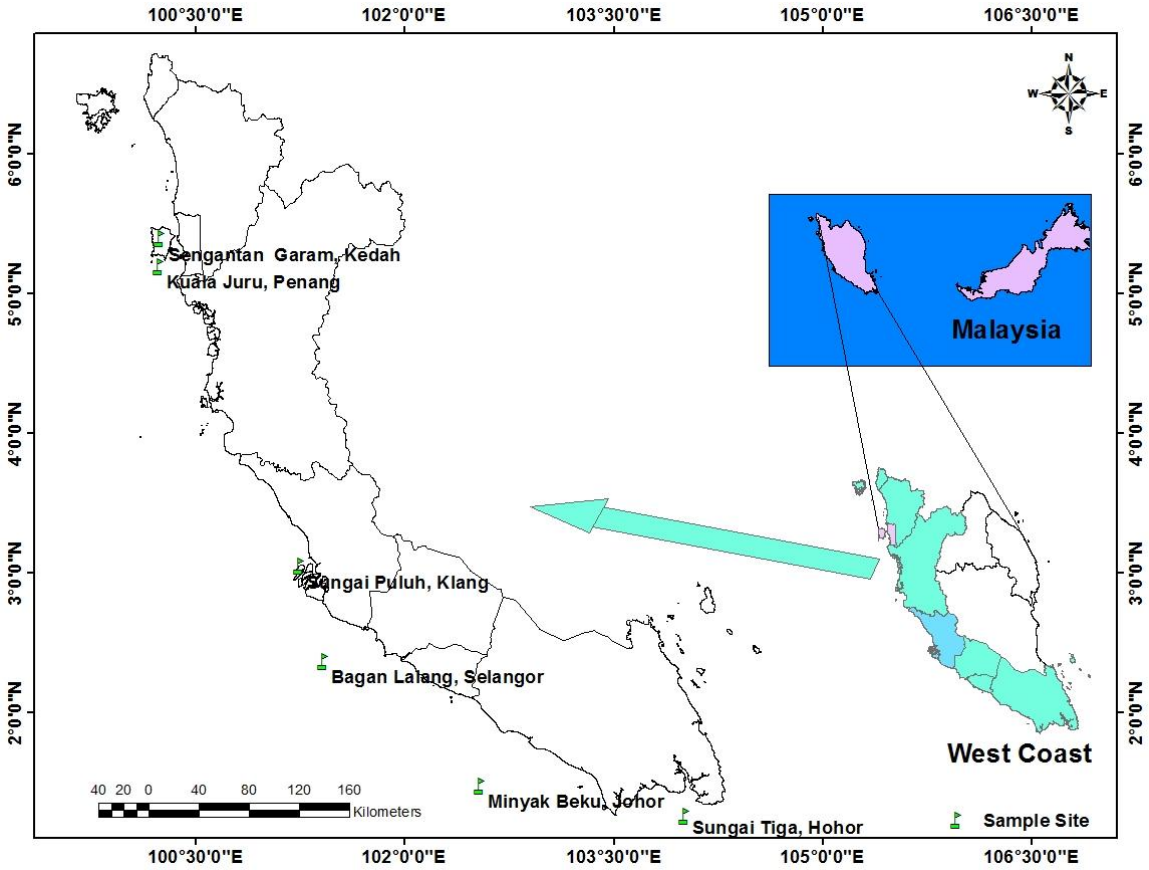

Figure 1. Map of west coast of Peninsular Malaysia showing the six sampling sites.

Table 1. Names, coordinates and description of sampling sites in the west coast of Peninsular Malaysia

Sampling site	Coordinates	Site description
Segantang Garam, Kedah (S.Garam)	N 05° 39.552′ E 100° 23.983′	Jetty, aqua cultural area and paddy field
Kuala Juru, Penang (K. Juru)	N 05° 19.683′ E 100° 22.949′	Industrial area, urbanization and aquaculture
Sungai Puluh, Klang (Sg. Puluh)	N 03° 04.786′ E 101° 23.903′	Jetty receiving domestic wastes and industrial area
Bagan Lalang, Selangor (Bg. Lalang)	N 02° 36.669′ E101° 41.100′	Recreational and agricultural areas
Minyak Beku, Johor (M.Beku)	N 01° 47.746′ E102° 53.395′	Jetty receiving domestic wastes and shipping activities
Sungai Tiga, Johor (Sg.Tiga)	N 01° 25.841′ E 104° 00.281′	Jetty, agricultural and oil plantation

Metal analysis

Sediment samples were dried in the laboratory using an air-circulating oven to a constant dry weight (d/w) at 80˚C. The dried sediment samples were crushed to powder by using a porcelain mortar and pestle then sieved vigorously to produce homogeneity [26], through a 63 µm stainless steel aperture sieve.

Geochemical fractions of Cu, Zn, Pb, Cd and Ni in the sediments were determined by modified sequential extraction technique (SET) as describe by Badri and Aston [27] and Tessier and Campbell [28]. The four-step extraction procedure of sediment handling and analysis employed using SET was described as:

1) Easily, freely or leachable and exchangeable (EFLE) (Fraction 1); 2) acid reducible (Fraction 2); 3) oxidisable organic (Fraction 3) and; 4) resistant fractions (Fraction 4). The mathematical summation of the first three types of fractions constitutes the "non-resistant phase which is closely related to anthropogenic inputs" [27].

Fish samples were removed from the refrigerator and plastic bags, rinsed with double distilled water (DDW) then thawed at room temperature. About 6-10 *P. schlosseri* from each sampling sites with the recorded body length (mm) and weight (g) were dissected on clean plastic material using stainless steel kits and glass equipment. The dissected parts were pooled into eight different parts namely; scales, muscle, bone, gills, operculum, intestine, liver and cartilage. These tissues were chosen because they play a role in metals uptake, bioaccumulation, formation of metal-complexes, storage and detoxification processes. All the eight parts were dried at 80˚C according to Mucha et al. [29] until a constant dry weight. Sample of each dried part was weighed separately (0.5 - 1.0 g) in triplicate and placed in digestion tubes. To each digestion tube 10 ml concentrated nitric acid (AnalaR grade, BDH 69%) was added and placed in a hot block digester unit at 40˚C for 1 hour (h). The temperature was then increased to 140˚C for at least three hours [26]. The digested samples were diluted to 40ml with DDW. The samples were then filtered through filter papers into pill box and the filtrate was stored until metal determination. The filtrates obtained from sediments and biological samples were determined for Cu, Zn, Pb, Cd and Ni by using an air-acetylene flame atomic absorption spectrophotometer (AAS) Perkin Elmer Analyst 800. Standard solutions were prepared from 1000 mg/L stock solution of each metal (BDH-Spectrosol). Standard solutions were prepared from 1000 mg/L stock solution of each metal (BDH-Spectrosol). The wavelengths for each metal were 324.8, 213.9, 283.3, 228.8 and 232.0 nm for Cu, Zn, Pb, Cd and Ni respectively.

The data were presented in µg/g dry weight. Multiple-level calibration standards were analyzed to generate calibration curves against which sample concentrations were calculated. During the period of AAS metal analysis, a quality control sample was routinely included for every 5 - 10 samples. Procedural blanks and quality control samples made from standard solutions for Cu, Zn, Pb, Cd and Ni were analyzed after every 5 - 10 samples to ensure the sensitivity and recovery of the instrument used. The procedures of quality assurance (QA) and quality control (QC) were employed to ensure the validity of the analytical data [30]. All plastics and glassware used were washed with detergent, Deacon 90, rinsed with double-distilled water and soaked in 10% HNO3 for at least 24 h, then rinsed with double distilled water and allowed to dry at room temperature. The QA and QC were controlled by procedural blanks, sample replicates and certified reference material (CRM). The quality of the method was checked with a CRM for soil from International Atomic Energy Agency (IAEA), Soil-5, Vienna; Austria and Dogfish liver DOLT-3 from National Research Council Canada (NRCC) were analyzed. These were checked to accuracy of the digestion method with the certified values supplied by the IAEA and NRCC. To ensure the sensitivity of the Atomic Absorption Spectrophotometer (AAS) and generate calibration curves against which sample concentrations were calculated. The results of similar digested samples analyzed for Cu, Zn, Pb, Cd, and Ni by the flame AAS Perkin Elmer. An analyst 800 showed acceptable recoveries of the metals; about 94% - 107% for soil and 87% - 110% for dogfish liver.

Heavy Metal Assessment Index in *Periophthalmodon schlosseri*

Metal selectivity index (MSI) and Tissue selectivity index (TSI)

MSI is defined as the relative metal-accumulating capacity of a tissue for a particular metal while TSI is defined as the relative tissue-occupying capacity of a metal in a particular tissue. MSI and TSI were calculated according to Nair et al. [31] as follows:

MSI = (absolute concentration of a metal in a tissue ×100)/Total concentration of all metals in that tissue

TSI = (absolute concentration of a metal in a tissue ×100)/Total concentration of that metal in all tissues

Biota sediment accumulation factor (BSAF)

Biota sediment accumulation factor (BSAF) is a parameter defining bioaccumulation of sediment-associated organic compounds or metals in tissues of the organism [32]. In the present study, the metal bioaccumulation in the tissues of giant mudskipper was evaluated by calculating the BSAF, which is defined as the ratio between the metal concentration in tissues and that in the sediment. Heavy metals accumulation in tissues of *P. schlosseri* from the sediments is expressed by the BSAF.

The BSAF is calculated using the following equation:

BSAF = CB/CS

Where; CB is the chemical concentration in the biota [mass of chemical per kg of biota/dry weight (d/w)], while CS is the concentration in the related sediment (mass of chemical per kg of sediment/d/w).

In the present study, the summation of non-resistant geochemical fractions was used as the total metal concentration (µg/g d/w) in sediment due to its bioavailable characteristics to the living organisms. BSAF is used to calculate the ratio of metal found in the organism to that in the sediments based on the amount of metal it accumulated. BSAF was used to classify the tissues of *P. schlosseri* as a macroconcentrator (BSAF > 2), microconcentrator (1 < BSAF < 2) or deconcentrator (BSAF < 1).

Statistical analysis

All statistical analyses of data were carried out using SPSS statistical package programs version 17 and graphs were plotted with Microsoft EXCEL 2007. Data were tested for the basic assumptions of normality and homogeneity of variance in exploratory data analysis in SPSS 17. One way analysis of variance (ANOVA) was calculated, post host comparison was made using Duncan's multiple range test at 0.05 confidence level.

RESULTS

Heavy metals concentrations in surface sediments

The Biota sediment accumulation factor (BSAF) of heavy metals in the tissues of *P. schlosseri,* was calculated by using the non-resistant geochemical fraction of heavy metals in the surface sediments from the six sampling sites in the west coast of Peninsular Malaysia and was presented in Table 2.

Heavy metals in *P. Schlosseri*

The measured metal concentrations in the tissues of *P. schlosseri* were presented in Table 3, as mean ± SE. The highest Cu concentration was observed in liver as 18.61 µg/g dry weights at M.Beku while cartilage recorded the highest concentrations of Zn, Pb, Cd and Ni as 218.28, 37.71, 4.42 and 121.84 µg/g dry weights at Bg. Lalang, Sg. Tiga, K. Juru, and K. Juru respectively. The lowest concentrations of Cu, Zn, Pb and Cd were found in muscle at Sg. Tiga, K. Juru, M. Beku and Sg. Tiga respectively, while liver recorded the lowest concentration of Ni at Bg. Lalang. Zinc concentrations were the highest in all tissues of *P. schlosseri* from the six sampling sites, except Ni concentration in cartilage at K.Juru.

Metal Selectivity Index (MSI) and Tissue Selectivity Index (TSI)

The MSI (%) and TSI (%) values were calculated for the five metals and presented in Tables 4 and 5 respectively. Based on the MSI (%) values, the affinity for metals in the tissues of *P. schlosseri* can generally be ranked in the order Zn > Pb > Ni > Cu > Cd, irrespective of the type of tissue for most of the sampling sites. Zn stands as the metal being accumulated to the greatest extent in all the tissues, except Ni in muscle and cartilage at K.Juru. The highest TSI (%) values for Cu and Pb were recorded in liver and gills respectively while highest TSI values for Zn, Cd and Ni were recorded in cartilage.

Biota sediment accumulation factor (BSAF)

The Biota sediment accumulation factor (BSAF) for Zn, Cu, Pb, Cd, and Ni in the tissues of *P. schlosseri* based on non – resistant sediments concentration are tabulated in Table 6. The BSAFs indicated bioaccumulation of heavy metals in intestine, liver, operculum, cartilage, gills and bone. Liver and intestine recorded the highest BSAF for Cu and Zn respectively, while the highest BSAF for Pb, Cd and Ni were recorded in cartilage.

[a] deconcentrators; [b] microconcentrators; [c] macroconcentrators

Table 2. Mean concentrations (µg/g d/w, n=3) of non-resistant geochemical fraction of Cu, Zn Pb, Cd and Ni in sediments of west coast of peninsular Malaysia.

Sampling site	Non-resistant Geochemical Fraction of Metals				
	Cu	Zn	Pb	Cd	Ni
S. Garam	4.89	25.51	9.04	0.39	6.10
K. Juru	9.57	296.32	0.00	0.00	0.65
Sg. Puluh	8.28	178.27	20.76	0.31	23.42
Bg. lalang	2.13	36.80	6.85	0.40	4.25
M. Beku	6.01	114.28	19.85	0.34	10.72
Sg. Tiga	3.60	60.25	14.08	0.36	6.42

Table 3. Mean metal concentration (µg/g d/w; ±SE) in the tissues of *P. schlosseri* from west coast of Peninsular Malaysia. Statistical significant ($P < 0.05$) between the sampling sites for a given examined metal was indicated by different superscript alphabets.

Sampling site	Tissue	Concentration (µg/g dry weight; ± SE)				
		Cu	Zn	Pb	Cd	Ni
S.Garam (n=10)	Scale	7.30^{d}±0.29	102.58^{c}±0.98	22.86^{c}±1.00	1.55^{b}±0.23	8.20^{ab}±0.13
	Muscle	5.23^{c}±0.37	45.24^{bc}±4.74	2.29^{a}±0.61	0.19^{a}±0.15	1.09^{a}±0.49
	Bone	7.16^{c}±0.06	75.76^{b}±0.79	19.01^{c}±2.00	1.58^{b}±0.24	5.06^{a}±0.15
	Gills	8.24^{a} ±0.29	100.40^{bc}±1.95	14.67^{a}±1.52	0.84^{bc}±0.23	3.98^{a}±0.55
	Operculum	7.49^{c}±0.32	109.54^{cd}±2.05	19.98^{c}±0.34	1.09^{bc}±0.19	6.15^{a}±0.89
	Intestine	16.98^{d}±0.32	152.63^{e}±1.69	11.98^{c}±0.16	0.61^{c}±0.18	5.82^{a}±1.16
	Liver	13.47^{b}±0.26	60.27^{c}±2.74	10.11^{d}±0.43	0.49^{ab}±0.22	2.90^{b}±0.41
	Cartilage	9.96^{c}±0.19	72.60^{c}±1.67	34.51^{d}±0.85	0.08^{a}±0.01	17.11^{b}±1.45
K.Juru (n=6)	Scale	3.50^{c}±0.23	68.30^{a}±5.75	1.57^{a}±0.07	1.17^{b}±0.07	14.90^{c}±4.33
	Muscle	1.72^{b}±0.16	12.79^{a}±11.23	0.43^{a}±0.04	0.11^{a}±0.03	21.00^{c}±0.92
	Bone	3.61^{b}±0.08	63.48^{a}±5.68	3.90^{a}±0.08	1.45^{b}±0.23	29.64^{c}±2.67
	Gills	4.52^{a}±0.06	93.37^{ab}±1.65	7.87^{a}±0.30	0.68^{b}±0.23	9.89^{b}±0.29
	Operculum	3.77^{a}±0.08	96.60^{ab}±4.39	3.13^{a}±0.05	1.29^{cd}±0.09	19.91^{b}±3.15
	Intestine	8.96^{b}±0.41	105.26^{c}±3.39	5.83^{b}±0.08	0.40^{bc}±0.10	89.98^{b}±13.59
	Liver	9.30^{a}±0.92	45.36^{b}±1.19	5.42^{bc}±0.96	1.33^{c}±0.12	5.04^{c}±0.70
	Cartilage	8.50^{c}±0.38	54.10^{a}±1.88	9.32^{a}±0.84	4.42^{b}±0.82	121.84^{c}±5.77
Sg.Puluh (n=10)	Scale	7.09^{d}±0.03	96.65^{c}±1.75	19.14^{bc}±0.38	0.49^{a}±0.20	12.35^{bc}±2.12
	Muscle	5.45^{d}±0.28	44.08^{c}±4.93	2.09^{bc}±1.17	0.20^{a}±0.11	8.74^{b}±0.72
	Bone	6.98^{c}±0.06	73.09^{ab}±2.19	15.21^{b}±1.64	0.55^{a}±0.26	12.70^{b}±0.23
	Gills	7.83^{a}±0.37	95.03^{ab}±3.50	12.09^{a}±0.64	0.28^{ab}±0.18	10.87^{b}±0.84
	Operculum	7.81^{c}±0.21	101.36^{bc}±2.45	15.38^{b}±2.93	0.91^{b}±0.12	15.64^{b}±1.04
	Intestine	15.56^{c}±0.79	80.75^{b}±0.72	10.49^{c}±2.30	0.08^{ab}±0.06	12.13^{a}±1.31
	Liver	14.67^{b}±1.18	34.31^{a}±2.11	7.90^{cd}±0.90	0.40^{ab}±0.20	12.51^{e}±0.85
	Cartilage	8.26^{c}±1.10	58.49^{b}±0.90	23.71^{c}±0.49	0.31^{a}±0.15	17.52^{a}±1.44
Bg.Lalang (n=9)	Scale	2.84^{b}±0.17	101.22^{c}±6.44	3.86^{a}±0.32	1.53^{b}±0.15	4.26^{a}±0.76
	Muscle	1.28^{b}±0.11	62.79^{c}±5.71	1.02^{a}±0.07	0.07^{a}±0.02	0.04^{a}±0.02
	Bone	3.32^{b}±0.22	82.97^{b}±2.12	3.21^{a}±0.02	1.76^{b}±0.18	7.71^{a}±0.22
	Gills	15.77^{a}±11.68	117.90^{d}±1.28	12.73^{a}±9.40	1.40^{c}±0.22	3.75^{a}±0.05
	Operculum	3.84^{b}±0.14	133.76^{e}±3.47	3.10^{a}±0.08	1.42^{cd}±0.08	5.03^{a}±0.44
	Intestine	7.10^{a}±0.58	123.53^{d}±1.10	2.36^{a}±0.48	0.59^{c}±0.15	0.04^{a}±0.01
	Liver	17.83^{c}±0.88	89.39^{d}±0.77	2.43^{ab}±0.26	0.60^{b}±0.06	0.04^{a}±0.01
	Cartilage	8.54^{c}±1.50	218.28^{e}±1.32	8.54^{a}±0.85	3.48^{b}±0.58	10.09^{ab}±1.01
M.Beku (n=10)	Scale	3.78^{c}±0.07	84.08^{b}±1.69	18.32^{b}±0.96	1.16^{b}±0.22	6.23^{ab}±0.14
	Muscle	1.21^{b}±0.18	43.40^{bc}±1.86	0.04^{a}±0.00	1.21^{b}±0.04	0.60^{a}±0.29
	Bone	3.38^{b}±0.07	81.50^{b}±4.05	19.53^{c}±1.19	1.70^{b}±0.32	7.89^{a}±0.88
	Gills	4.25^{a}±0.09	106.01^{c}±2.78	11.55^{a}±0.57	1.40^{c}±0.06	3.70^{a}±0.21
	Operculum	4.17^{b}±0.04	115.21^{d}±1.75	17.31^{bc}±0.90	1.49^{d}±0.07	5.40^{a}±0.13
	Intestine	9.75^{b}±0.00	120.46^{d}±1.10	0.20^{a}±0.01	0.04^{a}±0.01	0.16^{a}±0.06
	Liver	18.61^{c}±1.32	84.87^{d}±2.30	0.04^{a}±0.01	0.23^{ab}±0.20	0.67^{a}±0.06
	Cartilage	5.09^{b}±0.58	85.11^{d}±1.10	20.54^{b}±0.91	1.03^{a}±0.01	6.92^{a}±0.58
Sg.Tiga (n=10)	Scale	1.66^{a}±0.11	68.90^{a}±2.86	35.38^{d}±3.08	0.04^{a}±0.01	6.03^{a}±0.25
	Muscle	0.04^{a}±0.00	36.88^{b}±0.44	20.65^{b}±1.75	0.05^{a}±0.02	0.28^{a}±0.06
	Bone	0.83^{a}±0.75	63.40^{a}±4.97	31.71^{d}±0.44	0.13^{a}±0.09	5.82^{a}±0.99
	Gills	0.85^{a}±0.77	89.41^{a}±5.56	28.06^{b}±2.08	0.04^{a}±0.01	3.47^{a}±0.32
	Operculum	0.17^{a}±0.11	90.18^{a}±4.89	33.52^{d}±1.00	0.04^{a}±0.00	6.22^{a}±0.49
	Intestine	9.39^{b}±0.00	66.28^{a}±1.16	28.45^{a}±0.93	0.05^{a}±0.01	4.46^{a}±0.38
	Liver	10.38^{a}±0.88	61.86^{c}±1.75	29.02^{e}±2.18	0.04^{a}±0.01	6.64^{d}±0.41
	Cartilage	0.48^{a}±0.11	76.02^{c}±1.36	37.71^{e}±0.47	0.04^{a}±0.01	12.42^{b}±0.90

Table 4. Metal selectivity index (MSI) in the tissues of *P. schlosseri*.

Sampling site	Tissue	MSI (%)				
		Cu	Zn	Pb	Cd	Ni
S.Garam	Scale	5.12	71.99	16.04	1.09	5.75
	Muscle	9.68	83.72	4.24	0.35	2.02
	Bone	6.59	69.78	17.51	1.46	4.66
	Gills	6.43	78.36	11.45	0.66	3.11
	Operculum	5.19	75.94	13.85	0.76	4.26
	Intestine	9.03	81.18	6.37	0.32	3.10
	Liver	15.44	69.09	11.59	0.56	3.32
	Cartilage	7.42	54.07	25.70	0.06	12.74
K.Juru	Scale	3.91	76.36	1.76	1.31	16.66
	Muscle	4.77	35.48	1.19	0.31	58.25
	Bone	3.54	62.19	3.82	1.42	29.04
	Gills	3.89	80.26	6.77	0.58	8.50
	Operculum	3.02	77.47	2.51	1.03	15.97
	Intestine	4.26	50.02	2.77	0.19	42.76
	Liver	14.00	68.26	8.16	2.00	7.58
	Cartilage	4.29	27.30	4.70	2.23	61.48
Sg.Puluh	Scale	5.22	71.21	14.10	0.36	9.10
	Muscle	9.00	72.79	3.45	0.33	14.43
	Bone	6.43	67.35	14.01	0.51	11.70
	Gills	6.21	75.36	9.59	0.22	8.62
	Operculum	5.54	71.84	10.90	0.64	11.08
	Intestine	13.07	67.85	8.81	0.07	10.19
	Liver	21.02	49.16	11.32	0.57	17.93
	Cartilage	7.63	54.01	21.89	0.29	16.18
Bg.Lalang	Scale	2.50	89.02	3.39	1.35	3.75
	Muscle	1.96	96.30	1.56	0.11	0.06
	Bone	3.35	83.83	3.24	1.78	7.79
	Gills	10.41	77.80	8.40	0.92	2.47
	Operculum	2.61	90.90	2.11	0.97	3.42
	Intestine	5.31	92.45	1.77	0.44	0.03
	Liver	16.17	81.05	2.20	0.54	0.04
	Cartilage	3.43	87.69	3.43	1.40	4.05
M.Beku	Scale	3.33	74.03	16.13	1.02	5.49
	Muscle	2.60	93.41	0.09	2.60	1.29
	Bone	2.96	71.49	17.13	1.49	6.92
	Gills	3.35	83.53	9.10	1.10	2.92
	Operculum	2.90	80.24	12.06	1.04	3.76
	Intestine	7.46	92.23	0.15	0.03	0.12
	Liver	17.82	81.28	0.04	0.22	0.64
	Cartilage	4.29	71.71	17.31	0.87	5.83
Sg.Tiga	Scale	1.48	61.51	31.59	0.04	5.38
	Muscle	0.07	63.70	35.66	0.09	0.48
	Bone	0.81	62.22	31.12	0.13	5.71
	Gills	0.70	73.39	23.03	0.03	2.85
	Operculum	0.13	69.30	25.76	0.03	4.78
	Intestine	8.64	61.01	26.19	0.05	4.11
	Liver	9.62	57.31	26.89	0.04	6.15
	Cartilage	0.38	60.01	29.77	0.03	9.81

Table 5. Tissue selectivity index (TSI) in the tissues of *P. schlosseri.*

Sampling site	Tissues	TSI (%)				
		Cu	Zn	Pb	Cd	Ni
S.Garam	Scale	9.63	14.27	16.88	24.11	16.30
	Muscle	6.90	6.29	1.69	2.95	2.17
	Bone	9.44	10.54	14.04	24.57	10.06
	Gills	10.87	13.96	10.83	13.06	7.91
	Operculum	9.88	15.23	14.76	16.95	12.22
	Intestine	22.39	21.23	8.85	9.49	11.57
	Liver	17.76	8.38	7.47	7.62	5.76
	Cartilage	13.13	10.10	25.49	1.24	34.01
K.Juru	Scale	7.98	12.67	4.19	10.78	4.77
	Muscle	3.92	2.37	1.15	1.01	6.73
	Bone	8.23	11.77	10.41	13.36	9.49
	Gills	10.30	17.31	21.00	6.27	3.17
	Operculum	8.59	17.91	8.35	11.89	6.38
	Intestine	20.42	19.52	15.56	3.69	28.82
	Liver	21.19	8.41	14.46	12.26	1.61
	Cartilage	19.37	10.03	24.87	40.74	39.03
Sg.Puluh	Scale	9.63	16.56	18.05	15.22	12.05
	Muscle	7.40	7.55	1.97	6.21	8.53
	Bone	9.48	12.52	14.35	17.08	12.40
	Gills	10.63	16.28	11.40	8.70	10.61
	Operculum	10.60	17.36	14.51	28.26	15.26
	Intestine	21.13	13.83	9.90	2.48	11.84
	Liver	19.92	5.88	7.45	12.42	12.21
	Cartilage	11.22	10.02	22.37	9.63	17.10
Bg.Lalang	Scale	4.69	10.89	10.36	14.10	13.76
	Muscle	2.12	6.75	2.74	0.65	0.13
	Bone	5.49	8.92	8.62	16.22	24.90
	Gills	26.06	12.68	34.17	12.90	12.11
	Operculum	6.35	14.39	8.32	13.09	16.25
	Intestine	11.73	13.29	6.34	5.44	0.13
	Liver	29.46	9.61	6.52	5.53	0.13
	Cartilage	14.11	23.48	22.93	32.07	32.59
M.Beku	Scale	7.52	11.67	20.93	14.04	19.73
	Muscle	2.41	6.02	0.05	14.65	1.90
	Bone	6.73	11.31	22.31	20.58	24.99
	Gills	8.46	14.71	13.20	16.95	11.72
	Operculum	8.30	15.99	19.78	18.04	17.10
	Intestine	19.41	16.72	0.23	0.48	0.51
	Liver	37.04	11.78	0.05	2.78	2.12
	Cartilage	10.13	11.81	23.47	12.47	21.92
Sg.Tiga	Scale	6.97	12.46	14.47	9.30	13.30
	Muscle	0.17	6.67	8.45	11.63	0.62
	Bone	3.49	11.47	12.97	30.23	12.84
	Gills	3.57	16.17	11.48	9.30	7.65
	Operculum	0.71	16.31	13.71	9.30	13.72
	Intestine	39.45	11.99	11.64	11.63	9.84
	Liver	43.61	11.19	11.87	9.30	14.64
	Cartilage	2.02	13.75	15.42	9.30	27.39

Table 6. Metals Biota sediment accumulation factor (BSAF) in the tissues of *P. schlosseri* from west coast of Peninsular Malaysia.

Sampling site	Tissues	Cu	Zn	Pb	Cd	Ni
S. Garam	**Scale**	1.49^{b}	4.02^{c}	2.53^{c}	3.97^{c}	1.35^{b}
K. Juru		0.37^{a}	0.23^{a}	0.15^{a}	2.93^{c}	22.89^{c}
Sg. Puluh		0.86^{a}	0.54^{a}	0.92^{a}	1.49^{b}	0.53^{a}
Bg. lalang		1.33^{b}	2.75^{c}	0.56^{a}	4.94^{c}	1.00^{b}
M. Beku		0.63^{a}	0.74^{a}	0.92^{a}	3.44^{c}	0.58^{a}
Sg. Tiga		0.46^{a}	1.14^{b}	2.51^{c}	0.11^{a}	0.94^{a}
S. Garam	**Muscle**	1.07^{b}	1.77^{b}	0.25^{a}	0.48^{a}	0.18^{a}
K. Juru		0.18^{a}	0.04^{a}	0.04^{a}	0.27^{a}	32.27^{c}
Sg. Puluh		0.66^{a}	0.25^{a}	0.10^{a}	0.60^{a}	0.37^{a}
Bg. lalang		0.60^{a}	1.71^{b}	0.15^{a}	0.21^{a}	0.01^{a}
M. Beku		0.20^{a}	0.38^{a}	0.00^{a}	3.59^{c}	0.06^{a}
Sg. Tiga		0.01^{a}	0.61^{a}	1.47^{b}	0.15^{a}	0.04^{a}
S.Garam	**Bone**	1.46^{b}	2.97^{c}	2.10^{c}	4.05^{c}	0.83^{a}
K. Juru		0.38^{a}	0.21^{a}	0.37^{a}	3.62^{c}	45.54^{c}
Sg. Puluh		0.84^{a}	0.41^{a}	0.73^{a}	1.65^{b}	0.54^{a}
Bg. lalang		1.56^{b}	2.25^{c}	0.47^{a}	5.71^{c}	1.82^{b}
M. Beku		0.56^{a}	0.71^{a}	0.98^{a}	5.05^{c}	0.74^{a}
Sg. Tiga		0.23^{a}	1.05^{b}	2.25^{c}	0.37^{a}	0.91^{a}
S.Garam	**Gills**	1.68^{b}	3.94^{c}	1.62^{b}	2.14^{c}	0.65^{a}
K. Juru		0.47^{a}	0.32^{a}	0.75^{a}	1.70^{b}	15.19^{c}
Sg. Puluh		0.95^{a}	0.53^{a}	0.58^{a}	0.85^{a}	0.46^{a}
Bg. lalang		7.39^{c}	3.20^{c}	1.86^{b}	4.52^{c}	0.88^{a}
M. Beku		0.71^{a}	0.93^{a}	0.58^{a}	4.15^{c}	0.35^{a}
Sg.Tiga		0.24^{a}	1.48^{b}	1.99^{b}	0.11^{a}	0.54^{a}
S. Garam	**Operculum**	1.53^{b}	4.29^{c}	2.21^{c}	2.79^{c}	1.01^{b}
K. Juru		0.39^{a}	0.33^{a}	0.30^{a}	3.23^{c}	30.59^{c}
Sg. Puluh		0.94^{a}	0.57^{a}	0.74^{a}	2.74^{c}	0.67^{a}
Bg. lalang		1.80^{b}	3.64^{c}	0.45^{a}	4.59^{c}	1.18^{b}
M. Beku		0.69^{a}	1.01^{b}	0.87^{a}	4.42^{c}	0.50^{a}
Sg. Tiga		0.05^{a}	1.50^{b}	2.38^{c}	0.11^{a}	0.97a
S. Garam	**Intestine**	3.47^{c}	5.98^{c}	1.33^{b}	1.56^{b}	0.95^{a}
K. Juru		0.94^{a}	0.36^{a}	0.55^{a}	1.00^{b}	138.23^{c}
Sg. Puluh		1.88^{b}	0.45^{a}	0.51^{a}	0.24^{a}	0.52^{a}
Bg. lalang		3.33^{c}	3.36^{c}	0.34^{a}	1.91^{b}	0.01^{a}
M. Beku		1.62^{b}	1.05^{b}	0.01^{a}	0.11^{a}	0.01^{a}
Sg. Tiga		2.61^{c}	1.10^{b}	2.02^{c}	0.14^{a}	0.69^{a}
S.Garam	**Liver**	2.75^{c}	2.36^{c}	1.12^{b}	1.26^{b}	0.48^{a}
K. Juru		0.97^{a}	0.15^{a}	0.52^{a}	3.32^{c}	7.74^{c}
Sg. Puluh		1.77^{b}	0.19^{a}	0.38^{a}	1.21^{b}	0.53^{a}
Bg. lalang		8.36^{c}	2.43^{c}	0.35^{a}	1.94^{b}	0.01^{a}
M. Beku		3.10^{a}	0.74^{a}	0.00^{a}	0.67^{a}	0.06^{a}
Sg. Tiga		2.89^{c}	1.03^{b}	2.06^{c}	0.11^{a}	1.03^{b}
S. Garam	**Cartilage**	2.03^{c}	2.85	3.82^{c}	0.20^{a}	2.81^{c}
K. Juru		0.89^{a}	0.18^{a}	0.89^{a}	11.04^{c}	187.17^{c}
Sg. Puluh		1.00^{b}	0.33^{a}	1.14^{b}	0.95^{a}	0.75^{a}
Bg. lalang		4.00^{c}	5.93^{c}	1.25^{b}	11.25^{c}	2.37^{c}
M. Beku		0.85^{a}	0.74^{a}	1.03^{b}	3.07^{c}	0.65^{a}
Sg. Tiga		0.13^{a}	1.26^{b}	2.68^{c}	0.11^{a}	1.93^{b}

Table 7. Heavy metals concentrations (µg/g) in fish species from Malaysia and other regions of the world.

Location	Species	Cu	Zn	Pb	Cd	Ni	Reference
South-western Mediterranean (coast of Sfax)	*Diplodus annularis* *Solea vulgaris* *Liza aurata*	ND-40.81	106-284.90	0.17-5.06	0.10-6.47	0.08-6.42	[76]
Iranian coastline of the Caspian Sea	*Acipenser persicus* *Acipenser stellatus* *Huso huso*	-	11.17-182.26	0.55-18.52	0.001-4.06	-	[81]
Red Sea, Egypt	*Epinephelus sp.,* *Caranx sp.,* *Scarus gibbus,* *Nemipterus japonicus,* *Sardinella sp.,* *Synodus sp.,* *Carangoides bajad,* *Lutjanus bohar,* *Thunnus albacares,* *Gerres oyena,* *Sargocentron spiniferum,* *Siganus rivulatus,* *Lethrinus sp.* *Trachurus mediterraneus*	0.17-18.62	1.17-64.61	0.14-6.93	0.03-8.37	-	[41]
Coastal Waters of Kapar and Mersing, Malaysia	*Arius thalassinus* *Johnius belangeri*	0.66 -55.87	13.12 -739.6	-	0.027-13.35	-	[114]
Kapar Coastal Waters, Malaysia	*Pennahia anea* *Arius maculatus*	0.83 -55.01	17.7-555.8	0.07-1.96	0.02-1.08	-	[80]
Pulicat Lake, North of Chennai, Southeast Coast of India	*Labeo rohita* *Cirrhina mrigala*	BDL-0.19	0.08-0.55	1.06-5.20	0.04-0.15	-	[73]
Coastal waters of Mersing, Malaysia	*Megalaspis cordyla* *Arius thalassinus* *Johnius belangeri*	1.51-26.0	17.54-365.1	0.12-2.03	0.02-6.14	-	[77]
Masan Bay, Korea	*Mugil cephalus , Enedrias nebulosus, Pleuronichthys cornutus,* *Conger myriaster,* *Acanthogobius flavimanus,* *Hexagrammos otakii,* *Sebastiscus marmoratus*	0.18-0.25	6.33-12.90	0.04-0.15	0.01	0.02	[75]
South West Malaysian Coast	*Nemipterus japonicas,* *Chirocentrus dorab,* *Lutjanus sebae,* *Otolithes ruber* *Pampus argenteus*	-	2.33-10.52	0.04-0.26	-	-	[87]

Gulf of Aqaba, Jordan	*Abudefduf saxatili, Chaetodon austricus, Chaetodon fasciatus, Epinephelus fasciatus, Fistularia petimba, Kyphosus sp., Mugil sp, Mulloidichthys auriflama, Parupenus cyclostomus, Polysteganus coeruleopunctatus, Thalassorna sp.*	ND-42.6	1.90-271.70	ND-35.0	ND-10.4	ND-23.0	[115]
Poompuhar coast, India	*Mugil cephalus*	20.48	156.78	ND	ND	0.004	[78]
East and west coast of Peninsular Malaysia	*Decapterus kurroides, Decapterus macrosoma, Megalaspis cordyla, Parastromateus Niger, Scomberoides lysan, Selar crumenophthalmus, Paraplagusia Bilineata, Pomadasys Kaakan, Nemipterus Furcosus, Euthynnus affinis, Rastrelliger kanagurta,* Acanthopagrus latus,	0.86-29.0	17.1-953	0.01-0.88	0.001-33.8	NA	[82]
Victoria Harbour, Hong Kong	*Siganus oramin*	5.7-26.6	66.6-192.9	19.1-94.6	3.3-18.0	-	[83]
Zhejiang Coastal Area, China	*Periophthalmus sericus*	1.31	-	0.064	0.03	19.9	[88]
Sunderban mangrove, India	*Liza parsia*	5.85-28.65	21.25-88.58	-	-	-	[89]
Gulf and Gulf of Oman	*Epinephelus coioides and Lethrinus nebulosus*	0.235-276.0	1.82-240	< 0.001-0.55	<0.001-195.0	<0.01-0.111	[85]
Coastal waters of Uruguay, Uruguay	*Odontesthes sp., Mugil platanus, Micropogonias furnieri, Urophycis brasiliensis, Cynoscion guatucupa, Menticirrhus americanus, and Mustelus schmitti.*	< 0.63-493.0	12.0-214.0	-	-	-	[86]
Coastal Lagoon, Eastern Gulf of California Mexico	*Cathorops fuerthii, Mugil cephalus, Oistithonema libertate, Seriola lalandi, Carcharhinus leucas, Cynoscion xanthulus,*	ND-535.0	6.0-2341.0	0.50-9.20	0.02-165.0	-	[84]

	Galeichthys peruvianus, Lutjanus Colorado and Sphyrna lewini,						
El-Mex Bay, Egypt	*Siganus rivulatus and Sargus sargus*	1.12-44.23	4.03-283.04	1.03-11.97	0.11-2.56	-	[93]
French coast of the Eastern English Channel and Southern Bight of the North Sea	*Pleuronectes platessa, Limanda limanda, Platichthys flesus and Gadus morua*	0.78-52.20	-	ND-0.38	0.004-1.1	-	[92]
Tagus Estuary, Portugal	*Liza ramada, Solea senegalensis and Pomatochistus minutus*	0.94-4.40	18.5-138.2	1.10-8.90	18.5-138.2	-	[74]
Esmoriz-Paramos coastal Lagoon, Portugal	*Liza saliens*	< 2.6-262.1	25.7-88.6	BDL	-	-	[90]
Camargue, French coast	*Anguilla anguilla*	24.0-43.0	104.0-128.0	0.16-0.2	0.04-0.09	-	[91]
West coast, Peninsular Malaysia	*P. schlosseri*	0.04-18.61	12.79-152.63	0.04-37.71	0.04-4.42	0.04-121.84	Present study

ND: Not detectable, BDL: Below detection limit

Table 8. Comparison of heavy metal concentration (µg/g) in giant mudskipper *P. schlosseri* with other species of mudskippers from other geographical regions.

Sample area/country	Fish species	Cu	Zn	Pb	Cd	Ni	Reference
Mai Po Bay, Hong Kong	*Boleophthalmus pectinirostris*	2.08	84.61	1.56	0.012	-	[116]
Coast of Tanzania	*Periophthalmus argentilineatus*	2.90-5.50	148.0-219.0	1.9-20.9	0.2-1.5	1.3-3.1	[117]
Dumai waters, Indonesia	*Periophthalmus sp*	-	-	1.30-2.88	0.01-0.41	4.06-8.95	[118]
Zhejiang Coastal Area, China	*Periophthalmus sericus*	1.31	-	0.064	0.03	19.9	[88]
Intertidal areas, Peninsular Malaysia	*P.schlosseri*	0.8-6.6	16.0-57.8	2.8-14.9	0.4-1.8	-	[119]
Northern coast of Hormuz Strait (Persian Gulf)	*Periophthalmus waltoni*	-	61.94-263.88	2.33-2.50	-	0.35-12.5	[94]
West coast, Peninsular Malaysia	*P. schlosseri*	0.04-18.61	12.79-152.63	0.04-37.71	0.04-4.42	0.04-121.84	Present study

DISCUSSION

The mean concentrations of metals in mudskipper *P. schlosseri* and their standard errors were presented in Table 3. Analysis of variance (ANOVA) followed by Duncan's multiple comparisons analysis showed that significant difference ($P < 0.05$) were observed in tissues metals concentrations in *P. schlosseri* except Cu in gills which shows no significant difference ($P > 0.05$) between the sampling sites.

The tissues of *P. schlosseri* showed different pattern of metals accumulation, but high concentration of Zn and low concentration of Cd were generally observed in the studied tissues.

The highest mean concentration of Cu was recorded in liver at M. Beku. The accumulation of essential heavy metal in the liver is associated with its role in metabolism. Cu is an essential element that is carefully regulated by physiological mechanisms in most organisms [1]. High levels of Cu in hepatic tissues are usually related to a natural binding proteins such as metallothioneins (MT) [33, 34]. It is well known that large amount of metallothionein induction occurs in the liver tissue of fishes. The high levels of Cu in the liver can be ascribed to the binding of Cu to metallothioneins (MT), which serves as a detoxification mechanisms [35, 36] and important role it plays in Cu homeostasis [37].

Non - significant difference ($P > 0.05$) was observed in liver Cu concentration between M. Beku and Bg. Lalang. Liver Cu was higher at these two sampling sites compared to others sites that recorded higher bioavailable Cu, which could be related to the fish size. It was observed that small fish shows higher Cu concentration in liver than big fish. And it is well known that metabolic activity of a young marine animal is normally higher than that of an old individual. Thus, metal accumulation was shown to be higher in younger individuals than the older ones [38, 39]. The mechanisms of neutralization harmful compounds are not developed sufficiently in young organisms. Therefore, larger amounts of toxins can accumulate in their bodies [40]. This result is in agreement with several studies conducted on heavy metals concentrations in different fish species which showed high Cu concentration in liver tissues [8, 41, 42].

The highest concentrations of Zn, Pb, Cd and Ni were found in cartilage. It has been shown that highly mineralized materials like cartilage accumulate, at a varying intensity, both the essential (including Fe, Zn, Cu, Mn) and non-essential (Pb, Cd, Hg) trace elements [43]. Zinc is one of the most important environmental toxicants [44]; yet also perform essential roles in a wide range of biological processes [45]. Zinc is the second most abundant trace element in the body after iron [46]. It functions as a cofactor where many enzymes depends upon it as well as body cells [47]. However, Zinc plays an important part in metabolic processes and the metal is also involved in ossification and acts on cartilage growth especially in young organisms [48, 49].

Lead (Pb) is a persistent heavy metal which has been characterized as a priority hazardous substance [50]. The highest mean concentration of Pb in the present study was observed in cartilage, calcareous tissue has great affinity for Pb, and once bound, Pb is practically immobile. Osteoblasts and chondocytes seem to be important target cells for the toxic effects of Pb [51].

The high concentration of Pb in cartilage is probably due to it higher affinity to osteocalcin than calcium. Increases amount of mineral bound to osteocalcin [52, 53] has shown to interfere with Ca^{2+} signalling in cells by competing for calcium binding sites [54]. Indeed, it has been shown that Pb^{2+} can be carried into cells via Ca^{2+} channels [55, 56]. While the low concentration in muscle and liver could be attributed to metal binding proteins such as metallothioneins which is low in muscle [57] and do not binds Pb in the liver [58].

Cadmium is a non-essential highly toxic and ecotoxic metal [59] with limited biological function [60] and of considerable environmental and occupational concern. It is a common pollutant in surface water and can cause adverse effects on fish and other organisms inhabiting these bodies of water [61, 62]. Cadmium is well known to disturb Ca homeostasis in both fish and mammals [63, 64]. Direct uptake of Cd by fish from the water is mainly in its free ionic form (Cd^{2+}) [65] and the indirect exposure is possible as dietary means when consumer organisms subsequently ingest metals bio accumulated in organisms at a lower trophic level with the potential for effects or bioaccumulation [66]. In fishes, Cd can cause very adverse effects because its blocks sulfhydryl groups in the enzymes and competes for binding sites that are essential for normal enzymes functioning [67]. It competes for binding site with calcium because of their resemblance. Cadmium is believed to share a common transport pathway with Ca^{2+} [64, 68] it seems possible that Cd^{2+} can mimic Ca^{2+} at and in Ca^{2+} channels in order to gain entry into the target cells. Experimental evidence indicates that Cd^{2+} may interact with membrane transporters involved in the uptake of nutritive metals, such as Ca^{2+}, Fe, and Zn, as a means to gain entry into target cells of organs affected adversely by this metal. This uptake has been proposed recently to occur through a mechanism of ionic mimicry [69],

whereby Cd^{2+} mimics the divalent cationic species one or more of these nutritive metals at the binding site of one or more carrier proteins and/or channels that transport these metals. Recently, some studies have shown that when Cd^{2+} enters into the body it interferes with metabolic pathways of some elements including Zn, Cu, Ca, among others and causes significant damage to their biological activities [70].

Nickel is well known contaminant with low rate of bioaccumulation in fish tissues. Nickel is classified as borderline element between hard and soft acid acceptor in chemical reactions with donor atoms. It occurs principally as Ni^{2+} and relatively non-toxic to fresh and marine water fishes but exposure to low level over extended periods may results in a number of toxicological effects. Cartilage recorded the highest mean concentration of Ni. The highest concentration of nickel in cartilage may be attributed to the competitive nature of Ni^{2+} with Ca^{2+} for the same transport mechanism. The metal accumulation in different fish organs depends on their physiological role, behavior and feeding habits, as well as regulatory ability, as reported by Clearwater et al. [71].

In general the accumulation of these metals by the cartilage was due to their calcium mimicking effects which they displaced it from its binding sites. Concentrations of Cd and Ni were found higher in cartilage than sediment, suggesting a higher rate of accumulation of these metals by cartilage, while the lowest concentrations of metals were found in muscle which could be attributed metal binding proteins such as metallothionein which is low in muscle. It's important to mention that accumulation of trace elements in muscle is relatively lower than other tissues due to the fact that muscle does not come into direct contact with the metals as it is totally covered externally by the skin and also it is not an active site for detoxification and therefore transport of trace metals from other tissues to muscle [72]. Metal accumulation in fish are mainly focused on metabolically active tissues and muscle tissues while the metal accumulation in cartilage and its potential use in monitoring programs have been neglected and does not received proper attention. Therefore, at the time of this study no available data on heavy metals concentration in fish cartilage to compare with the present findings.

The results of the present study were comparable to reported metals concentration in the tissues of different fish species from other geographical regions of the world as shown in Table 7. The concentration of Cu in the tissues of giant mudskipper *P. schlosseri* was above the reported concentrations from Pulicat Lake, North of Chennai, Southeast Coast of India [73], Tagrus Estuary, Portugal [74] and Masan Bay, Korea [75], but below the concentrations from Coast of Sfax [76] Coastal Waters of Kapar and Mersing, Malaysia [77], Poompuhar coast, India [78]) and Gulf of Aqaba, Jordan [79]. Compared to the concentrations of Zn reported from Kapar coastal waters, Malaysia [80], Coastal waters of Mersing, Malaysia [77], Iranian Coastline of Caspian Sea [81], East and west coast of Peninsular Malaysia [82], Victoria Harbour, Hong Kong [83], Coastal Lagoon, Mexico [84], Gulf of Oman [85] and Coastal waters of Uruguay [86] the concentration of Zn in the tissues of *P. schlosseri* was low but above South west Malaysia coast [87], Red sea , Egypt [41], Zhejiang coastal area, China [88], Sunderban mangrove, India [89]) and Esmoriz-Paramos coastal Lagoon, Portugal [90]. The concentrations of Pb and Cd in the tissues of the present study were higher than Iranian Coastline of Caspian Sea [81], Kapar coastal waters, Malaysia [80], Pulicat Lake, North of Chennai, Southeast Coast of India [73], Camargue coast, France [91], Southern Bight of the North Sea, England [92] and El-Mex Bay, Egypt [93]. The concentrations of Ni in this study were found higher than all the reported Ni concentrations in Table 7.

Comparison between heavy metal concentrations in giant mudskipper *P. schlosseri* with other species of mudskippers from various geographical regions (Table 8.) revealed higher concentrations of Cu, Zn, Pb, Cd and Ni in *P. schlosseri* than in other species of mudskippers with exception of Zn at Northern coast of Hormuz Strait, Persian Gulf [94]. Concentration of Cu, Zn, Pb and Cd were found higher in the present study than in *P. schlosseri* from intertidal areas at Morib and Remis in Peninsular Malaysia.

The calculated MSI (%) values in most of the studied tissues indicated Zn as the metal with highest value. Zinc is an essential micronutrient in all marine organisms and a cofactor in nearly 300 enzymes; it is therefore not surprising the concentration of this metal was found highest in all the examined tissues. Although fish bio accumulate Pb from seawater in proportion to its concentration in solution, Pb is not very bio available or toxic to marine animals [95], the metal (Pb) was ranked second based on MSI (%) in this study. It was observed that, fish have the ability to accumulate heavy metal in their tissues and organs in different amounts. These differences result from different affinity of metals to fish tissues, different uptake, deposition and excretion rates [96]. The differences in concentration of metals in different parts of an organism could be attributed to the tendency of metals to bind to various molecular groups found within the cells of organisms as well as the degree of exposure to metal as influenced by its metabolic characteristics and position in the food chain [97]. Zinc, Pb and Cd indicated high MSI values in muscle, whereas Cu and Ni showed high MSI value in liver and cartilage respectively. High accumulation of heavy metals in muscles may be due to its strong binding with cysteine residues of metallothionein [98] and the feeding habit of the fish. High

concentration of heavy metals in muscles were reported by different authors; Cd [99], Zn and Pb Abdallah [100], many studies have attributed high metal accumulation in muscle to the feeding habit of the fish.

The tissue selectivity index (TSI) has not shown any unique ranking with regards to the studied tissues except for liver which showed high copper selectivity at all the six sampling sites. The specific tissues in which certain metals can be retained depend on the properties of the element, metabolic turnover and the state of the organism [101]. Furthermore, tissue specific accumulation has been proposed as a key biomarker to assess the effect of the chronic exposure of metals in aquatic organisms [102].

Based on the BSAF values, most of the tissues of *P. schlosseri* accumulated higher Cd and Ni and could be classified as macro concentrators of these metals. Liver record the highest BSAF value for Cu while intestine recorded the highest BSAF for Zn. It is generally accepted that heavy metal uptake occurs mainly from water, food and sediment. Digestive tissue is observed as one of the major routes for uptake of metal in diet or sediment by aquatic organism [103]. It has been suggested that diet is a much more significant contributor for nutritionally essential metals such as Cu and Zn [104].

Many researchers have stated that dietary Zn is the fundamental reason for increased Zn in marine fish [105]. Miller et al. [106] has reported that the higher Cu accumulation in liver of rainbow trout increased as the Cu concentration increased in the diet.

Even though there was no clear evidence about Cu and Zn dietary but according to Bordajandi et al. [107], the diet has remarkable role in the bio concentration process for some metals, mainly for Cu and Zn. Liver is an active organ in fish and high amount of Cu accumulation can possibly be attributed to the involvement of liver in detoxification and removal of toxic substances circulating in the blood stream [108]. Metabolic tissues such as liver, intestine and kidneys that are involved in the process of digestion, accumulate trace metals more than any other tissues [109-111]. The highest BSAF for Pb, Cd and Ni were recorded in cartilage. The variations in heavy metals contents in fish tissues were attributed to affinity of tissues to the metals and variation in heavy metals content in the environment. Other factors, such as sex and size may also influence metal bioaccumulation [112, 113]. Nickel has the highest BSAF values and was therefore considered as the most bio accumulated of all the metals studied. While liver, intestine and cartilage showed greater capacity for metal bioaccumulation than other tissues analyzed in this study. It could be suggested based on BSAF values that, liver and intestine were good accumulators of Cu and Zn respectively while cartilage for Pb, Cd and Ni. The non-essential metals especially Cd and Ni were higher than the essential metals i.e. Cu and Zn in the tissues of *P. schlosseri*, because essential metals like Cu and Zn are carefully regulated by physiological mechanisms in most organisms [101]. On the other hand, the non-essential metals compete for calcium binding sites and they are not regulated by the organisms. Based on the BSAF values, most tissues of *P. schlosseri* accumulated higher Cd and Ni and could be classified as macro concentrators of these metals.

CONCLUSION

The present study was carried out to provide information on the use of giant mudskipper *P.schlosseri* as a biomonitoring agent of heavy metals in the coastal environment. Results obtained from the study indicated high metal accumulation in liver, intestine and cartilage of giant mudskipper *P.schlosseri*. It was concluded that *P.schlosseri*, can accumulate metals in higher concentrations than the quantity in its surrounding environment and it might be considered as a bio monitor of heavy metals pollution in marine coastal environment. However, there is need for further research on heavy metals content in giant mudskipper in order to make them excellent organisms in Eco toxicological research.

Authors' Contributions

Prof. Dr. Ahmad Ismail participated in the design of the research, supervision and intellectual corrections of the manuscript. Tijjani Rufa'i Buhari has participated in design and planning of the experiments, data collection, analyses, interpretation and drafting of manuscript. All authors of this research paper have directly participated in the planning, analysis of the study and have approved the final version submitted.

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Competing interests

The authors declare that they have no competing interests.

REFERENCES

1. Erdorul Ö, Ates DA. 2006. Determination of cadmium and copper in fish samples from Sir and Menzelet dam lake Kahramanmaras, Turkey. *Environmental Monitoring and Assessment*, 117(1-3): 281.
2. Yilmaz F, Ozdemir N, Demirak A, Tuna AL. 2007. Heavy metal levels in two fish species *Leuciscus cephalus* and *Lepomis gibbosus*. *Food Chemistry*, 100(2): 830-835.
3. Zhao S, Feng C, Quan W, Chen X, Niu J, Shen Z. 2012. Role of living environments in the accumulation characteristics of heavy metals in fishes and crabs in the Yangtze River Estuary, China. *Marine Pollution Bulletin*, 64(6): 1163-1171.
4. Karadede-Akin H, Ünlü E. 2007. Heavy metal concentrations in water, sediment, fish and some benthic organisms from Tigris River, Turkey. *Environmental Monitoring and Assessment*, 131(1): 323-337.
5. Tawari-Fufeyin P, Ekaye SA. 2007. Fish species diversity as indicator of pollution in Ikpoba River, Benin City, Nigeria. *Reviews in Fish Biology and Fisheries*, 17(1): 21-30.
6. Anim AK, Ahialey EK, Duodu GO, Ackah M, Bentil NO. 2011. Accumulation profile of heavy metals in fish samples from Nsawam, along the Densu River, Ghana. *Research Journal of Environmental and Earth Sciences*, 3(1): 56-60.
7. Dural M, Göksu MZL, Özak AA. 2007. Investigation of heavy metal levels in economically important fish species captured from the Tuzla lagoon. *Food Chemistry*, 102(1): 415-421.
8. Zubcov E, Zubcov N, Ene A, Biletchi L. 2012. Assessment of copper and zinc levels in fish from freshwater ecosystems of Moldova. *Environmental Science and Pollution Research*, 19(6): 2238-2247.
9. Jabeen G, Javed M, Azmat. 2012. Assessment of heavy metals in the fish collected from the river Ravi, Pakistan. *Pakistan Veterinary Journal*, 32(1): 107-111.
10. Lenhardt M, Jarić I, Višnjić-Jeftić Ž, Skorić S, Gačić Z, Pucar M, et al. 2012. Concentrations of 17 elements in muscle, gills, liver and gonads of five economically important fish species from the Danube River. *Knowledge and Management of Aquatic Ecosystems*, (407): 02.
11. Qiu Y-W. 2015. Bioaccumulation of heavy metals both in wild and mariculture food chains in Daya Bay, South China. *Estuarine, Coastal and Shelf Science*, 163: 7-14.
12. Kruitwagen G. 2007. Ecology and toxicology of mangrove fauna in Tanzania: with particular reference to the mudskipper. PhD Thesis; Radbound University of Nijmegen, Nijmegen.
13. Murdy EO. 1989. A taxonomic revision and cladistic analysis of the oxudercine gobies (Gobiidae: Oxudercinae). The Australian Museum. ISBN 0-7305-6374-X.
14. Ansari AA, Trivedi S, Saggu S, Rehman H. 2014. Mudskipper: A biological indicator for environmental monitoring and assessment of coastal waters. *Journal of Entomology and Zoology Studies*, 2(6): 22-33.
15. Rainbow PS. 1996. Biomonitoring of heavy metal availability in the marine environment: Marine Pollution Bulletin, 1995, 31/4-12 (183-192). Summ. in Engl. *Environmental Pollution*, 94(2): 244.
16. Rainbow PS, Phillips DJH. 1993. Cosmopolitan biomonitors of trace metals. *Marine Pollution Bulletin*, 26(11): 593-601.
17. Polgar G, Lim R, 2011. Mudskippers: Human use, ecotoxicology and biomonitoring of mangrove and other soft bottom intertidal ecosystems. In: Mangroves: Ecology, Biology and Taxonomy. Nova Science. ISBN 1617289914, 9781617289910.
18. Murdy EO. 1986. Mudskippers of Malaysia: the lords of the mudflat. Freshwater Marine Aquarium 9:20-23.
19. Authman MMN, Zaki MS, Khallaf EA, Abbas HH. 2015. Use of fish as bio-indicator of the effects of heavy metals pollution. *Journal of Aquaculture Research & Development*, 6(4): 1.
20. Montalvo C, Aguilar CA, Amador LE, Cerón JG, Cerón RM, Anguebes F, et al. 2014. Metal contents in sediments (Cd, Cu, Mg, Fe, Mn) as indicators of pollution of Palizada River, Mexico. *Environment and Pollution*, 3(4): 89.
21. Kuklina I, Kouba A, Kozák P. 2013. Real-time monitoring of water quality using fish and crayfish as bio-indicators: a review. *Environmental Monitoring and Assessment*, 185(6): 5043-5053.
22. Elnabris KJ, Muzyed SK, El-Ashgar NM. 2013. Heavy metal concentrations in some commercially important fishes and their contribution to heavy metals exposure in Palestinian people of Gaza Strip (Palestine). *Journal of the Association of Arab Universities for Basic and Applied Sciences*, 13(1): 44-51.
23. Zhou Q, Zhang J, Fu J, Shi J, Jiang G. 2008. Biomonitoring: an appealing tool for assessment of metal pollution in the aquatic ecosystem. *Analytica chimica acta*, 606(2):135-150.
24. Ismail A. 1993. Heavy metal concentrations in sediments off Bintulu, Malaysia. *Marine Pollution Bulletin*, 26(12): 706-707.
25. Zulkifli SZ, Ismail A, Mohamat-Yusuff F, Arai T, Miyazaki N. 2010. Johor Strait as a hotspot for trace elements contamination in Peninsular Malaysia. *Bulletin of environmental contamination and toxicology*, 84(5): 568-573.
26. Ismail A. 1993. Heavy metal concentrations in sediments off Bintulu, Malaysia. *Marine Pollution Bulletin*, 27(12): 706-707.
27. Badri MA, Aston SR. 1983. Observations on heavy metal geochemical associations in polluted and non-polluted estuarine sediments. *Environmental Pollution Series B, Chemical and Physical*, 6(3): 181-193.
28. Tessier A, Campbell PGC. 1987. Partitioning of trace metals in sediments: relationships with bioavailability. *Hydrobiologia*, 149(1): 43-52.

29. Mucha AP, Vasconcelos MTSD, Bordalo AA. 2003. Macrobenthic community in the Douro estuary: relations with trace metals and natural sediment characteristics. *Environmental Pollution*, 121(2): 169-180.
30. Ismail A, Yap CK, Fong CF. 2004. Concentrations of Cd, Cu and Zn in sediments collected from urban lakes at Kelana Jaya, Peninsular Malaysia. *Wetlands*, 2(4): 248-258.
31. Nair M, Jayalakshmy KV, Balachandran KK, Joseph T. 2006. Bioaccumulation of toxic metals by fish in a semi-enclosed tropical ecosystem. *Environmental Forensics*, 7(3): 197-206.
32. Kleinov K, Nichols J, Hayton W, Mc Kim J, Barron M. 2008. Toxicokinetics in fish. In: *The toxicology of fishes*. CRC PressTaylor and Francis Group LLC. ISBN-13: 978-0-415-24868-6.
33. Görür FK, Keser R, Akçay N, Dizman S. 2012. Radioactivity and heavy metal concentrations of some commercial fish species consumed in the Black Sea Region of Turkey. *Chemosphere*, 87(4): 356-361.
34. Qadir A, Malik RN. 2011. Heavy metals in eight edible fish species from two polluted tributaries (Aik and Palkhu) of the River Chenab, Pakistan. *Biological trace element research*, 143(3): 1524-1540.
35. Hogstrand C, Haux C. 1991. Binding and detoxification of heavy metals in lower vertebrates with reference to metallothionein. *Comparative Biochemistry and Physiology Part C: Comparative Pharmacology*, 100(1-2): 137-141.
36. Amiard JC, Amiard-Triquet C, Barka S, Pellerin J, Rainbow PS. 2006. Metallothioneins in aquatic invertebrates: Their role in metal detoxification and their use as biomarkers. *Aquatic Toxicology*, 76(2): 160-202.
37. Grosell MH, Hogstrand C, Wood CM. 1997. Cu uptake and turnover in both Cu-acclimated and non-acclimated rainbow trout (Oncorhynchus mykiss). *Aquatic Toxicology*, 38(4): 257-276.
38. Nussey HH. 2000. Bioaccumulation of chromium, manganese, nickel and lead in the tissues of the moggel, Labeo umbratus (Cyprinidae), from Witbank Dam, Mpumalanga. *Water SA-Pretoria*, 26(2): 269-284.
39. Widianarko B, Van Gestel CAM, Verweij RA, Van Straalen NM. 2000. Associations between trace metals in sediment, water, and guppy, Poecilia reticulata (Peters), from urban streams of Semarang, Indonesia. *Ecotoxicology and environmental safety*, 46(1): 101-107.
40. Gašpić ZK, Zvonarić T, Vrgoč N, Odžak N, Barić A. 2002. Cadmium and lead in selected tissues of two commercially important fish species from the Adriatic Sea. *Water research* 2, 36(20): 5023-5028.
41. El-Moselhy KM, Othman AI, El-Azem HA, El-Metwally MEA. 2014. Bioaccumulation of heavy metals in some tissues of fish in the Red Sea, Egypt. *Egyptian Journal of Basic and Applied Sciences*, 1(2): 97-105.
42. Wei Y, Zhang J, Zhang D, Tu T, Luo L. 2014. Metal concentrations in various fish organs of different fish species from Poyang Lake, China. *Ecotoxicology and environmental safety*, 104: 182-188.
43. Kalisińska E, Salicki W, Kavetska KM, Ligocki M. 2007. Trace metal concentrations are higher in cartilage than in bones of scaup and pochard wintering in Poland. *Science of the Total Environment*, 388(1): 90-103.
44. Eisler R. 1993. Zinc Hazards to Fish, Wildlife, and Invertebrates: A Synoptic Review: U.S. Department of the Interior, Fish and Wildlife Service.
45. Watanabe T, Kiron V, Satoh S. 1997. Trace minerals in fish nutrition. *Aquaculture*, 151(1): 185-207.
46. Choi DW, Koh JY. 1998. Zinc and brain injury. *Annual review of neuroscience*, 21(1): 347-375.
47. Al-Ouran N. 2005. Environmental assessment, documentation and spatial modeling of heavy metal pollution along the Jordan Gulf of Aqaba using coral reefs as environmental indicator. PhD Thesis; Department of Hydrogeology and Environment, Julius-Maximilians-University of Würzburg.
48. Lai YL, Yamaguchi M. 2005. Effects of copper on bone component in the femoral tissues of rats: anabolic effect of zinc is weakened by copper. *Biological and Pharmaceutical Bulletin*, 28(12): 2296-2301.
49. Brandão-Neto J, Stefan V, Mendonça BB, Bloise W, Castro AVB. 1995. The essential role of zinc in growth. *Nutrition Research*, 15(3): 335-358.
50. Sfakianakis DG, Renieri E, Kentouri M, Tsatsakis AM. 2015. Effect of heavy metals on fish larvae deformities: a review. *Environmental research*, 137: 246-255.
51. Puzas JE, Sickel MJ, Felter ME. 1992. Osteoblasts and chondrocytes are important target cells for the toxic effects of lead. *Neurotoxicology*, 13(4): 783.
52. Dowd TL, Rosen JF, Gundberg CM, Gupta RK. 1994. The displacement of calcium from osteocalcin at submicromolar concentrations of free lead. *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease*, 1226(2): 131-137.
53. Dowd TL, Rosen JF, Mints L, Gundberg CM. 2001. The effect of Pb 2+ on the structure and hydroxyapatite binding properties of osteocalcin. *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease*, 1535(2): 153-163.
54. Pounds JG, Long GJ, Rosen JF. 1991. Cellular and molecular toxicity of lead in bone. *Environmental health perspectives*, 91: 17.
55. Kerper LE, Hinkle PM. 1997. Cellular uptake of lead is activated by depletion of intracellular calcium stores. *Journal of Biological Chemistry*, 272(13): 8346-8352.
56. Kerper LE, Hinkle PM. 1997. Lead uptake in brain capillary endothelial cells: activation by calcium store depletion. *Toxicology and applied pharmacology*, 146(1): 127-133.
57. Allen-Gil SM, Martynov VG. 1995. Heavy metal burdens in nine species of freshwater and anadromous fish from the Pechora River, northern Russia. *Science of the Total Environment*, 160: 653-659.
58. Ploetz DM, Fitts BE, Rice TM. 2007. Differential accumulation of heavy metals in muscle and liver of a marine fish,(King Mackerel, *Scomberomorus cavalla* Cuvier) from the Northern Gulf of Mexico, USA. *Bulletin of environmental contamination and toxicology*, 78(2): 134-137.

59. Stancheva M, Makedonski L, Petrova E. 2013. Determination of heavy metals (Pb, Cd, As and Hg) in black sea grey mullet (Mugil cephalus). *Bulgarian Journal of Agricultural Science*, 19(1): 30-34.
60. Lane TW, Morel FMM. 2000. A biological function for cadmium in marine diatoms. *Proceedings of the National Academy of Sciences*, 97(9): 4627-4631.
61. Gravel A, Campbell PG, Hontela A. 2005. Disruption of the hypothalamo-pituitary-interrenal axis in 1+ yellow perch (*Perca flavescens*) chronically exposed to metals in the environment. *Canadian Journal of Fisheries and Aquatic Sciences*, 62(5): 982-990.
62. Perceval O, Couillard Y, Pinel-Alloul B, Giguère A, Campbell PGC. 2004. Metal-induced stress in bivalves living along a gradient of Cd contamination: relating sub-cellular metal distribution to population-level responses. *Aquatic Toxicology*, 69(4): 327-345.
63. Giles MA. 1984. Electrolyte and water balance in plasma and urine of rainbow trout (*Salmo gairdneri*) during chronic exposure to cadmium. *Canadian Journal of Fisheries and Aquatic Sciences*, 41(11): 1678-1685.
64. Verbost PM, Van Rooij J, Flik G, Lock RAC, Bonga SEW. 1989. The movement of cadmium through freshwater trout branchial epithelium and its interference with calcium transport. *Journal of experimental biology*, 145(1): 185-197.
65. Okocha RC, Adedeji OB. 2011. Overview of cadmium toxicity in fish. *Journal of Applied Sciences Research*, 7(7): 1195-1207.
66. Drexler J, Fisher N, Henningsen G, Lanno R, McGeer J, Sappington K, et al. 2003. Issue paper on the bioavailability and bioaccumulation of metals. In: *US Environmental Protection Agency, Risk Assessment Forum, Washington, DC, USA*.
67. Allen P. 1994. Mercury accumulation profiles and their modification by interaction with cadmium and lead in the soft tissues of the cichlid *Oreochromis aureus* during chronic exposure. *Bulletin of environmental contamination and toxicology*, 53(5): 684-692.
68. Verbost PM, Flik G, Lock RA, Bonga SEW: Cadmium inhibition of Ca2+ uptake in rainbow trout gills. *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology* 1987, 253(2):R216-R221.
69. Zalups RK, Ahmad S: Molecular handling of cadmium in transporting epithelia. *Toxicology and applied pharmacology* 2003, 186(3):163-188.
70. Malekpouri P, Moshtaghie AA, Kazemian M, Soltani M. 2011. Protective effect of zinc on related parameters to bone metabolism in common carp fish (*Cyprinus carpio* L.) intoxified with cadmium. *Fish Physiology and Biochemistry*, 37(1): 187-196.
71. Clearwater SJ, Farag AM, Meyer JS. 2002. Bioavailability and toxicity of dietborne copper and zinc to fish. *Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology*, 132(3): 269-313.
72. Alhashemi AH, Sekhavatjou MS, Kiabi BH, Karbassi AR. 2012. Bioaccumulation of trace elements in water, sediment, and six fish species from a freshwater wetland, Iran. *Microchemical Journal*, 104: 1-6.
73. Batvari B, Prabhu D, Kamalakannan S, Krishnamurthy RR. 2015. Heavy metals accumulation in two fish species (*Labeo rohita* and *Cirrhina mrigala*) from Pulicat Lake, North of Chennai, Southeast Coast of India. *J Chem Pharm Res*, 7(3): 951-956.
74. França S, Vinagre C, Caçador I, Cabral HN. 2005. Heavy metal concentrations in sediment, benthic invertebrates and fish in three salt marsh areas subjected to different pollution loads in the Tagus Estuary (Portugal). *Marine Pollution Bulletin*, 50(9): 998-1003.
75. Kwon Y-T, Lee C-W. 2001. Ecological risk assessment of sediment in wastewater discharging area by means of metal speciation. *Microchemical Journal*, 70(3): 255-264.
76. Ben SZ, Ayadi H. 2016. Heavy metal accumulation in Diplodus annularis, Liza aurata, and *Solea vulgaris* relevant to their concentration in water and sediment from the southwestern Mediterranean (coast of Sfax). *Environmental science and pollution research international*, 23(14):13895-13906.
77. Fathi HB, Othman MS, Mazlan AG, Arshad A, Amin SM, Simon KD. 2013. Trace metals in muscle, liver and gill tissues of marine fishes from Mersing, Eastern coast of peninsular Malaysia: concentration and assessment of human health risk. *Asian Journal of Animal and Veterinary Advances*, 8(2): 227-236.
78. Prasath P, Khan TH. 2008. Impact of Tsunami on the heavy metal accumulation in water, Sediments and fish at Poompuhar coast, Southeast Coast of India. *Journal of Chemistry*, 5(1): 16-22.
79. Abu Hilal AH, Ismail NS. 2008. Heavy metals in eleven common species of fish from the Gulf of Aqaba, Red Sea. *Jordan Journal of Biological Sciences*, 1: 13 -18.
80. Bashir FA, Alhemmali EM. 2015. Analysis of some heavy metal in marine fish in muscle, liver and gill tissue in two marine fish spices from Kapar coastal waters, Malaysia. The Second Symposium on Theories and Applications of Basic and Biosciences Vol. (2) Issue (1).
81. Mashroofeh A, Bakhtiari AR, Pourkazemi M, Rasouli S. 2013. Bioaccumulation of Cd, Pb and Zn in the edible and inedible tissues of three sturgeon species in the Iranian coastline of the Caspian Sea. *Chemosphere*, 90(2): 573-580.
82. Agusa T, Kunito T, Yasunaga G, Iwata H, Subramanian A, Ismail A, et al. 2005. Concentrations of trace elements in marine fish and its risk assessment in Malaysia. *Marine Pollution Bulletin*, 51(8): 896-911.
83. Chan KM. 1995. Concentrations of copper, zinc, cadmium and lead in rabbitfish (*Siganus oramin*) collected in Victoria Harbour, Hong Kong. *Marine Pollution Bulletin*, 31(4-12): 277-280.
84. Ruelas-Inzunza J, Paez-Osuna F. 2007. Essential and toxic metals in nine fish species for human consumption from two coastal lagoons in the Eastern Gulf of California. *Journal of Environmental Science and Health Part A*, 42(10): 1411-1416.
85. de Mora S, Fowler SW, Wyse E, Azemard S. 2004. Distribution of heavy metals in marine bivalves, fish and coastal sediments in the Gulf and Gulf of Oman. *Marine Pollution Bulletin*, 49(5): 410-424.

86. Viana F, Huertas R, Danulat E. 2005. Heavy metal levels in fish from coastal waters of Uruguay. *Archives of environmental contamination and toxicology*, 48(4): 530-537.

87. Kamaruzzaman BY, Rina Z, John BA, Jalal KCA. 2011. Heavy metal accumulation in commercially important fishes of South West Malaysian coast. *Research Journal of Environmental Sciences*, 5(6): 595.

88. Fang J, Wang KX, Tang JL, Wang M, Ren SJ, Wu HY, et al. 2004. Copper, lead, zinc, cadmium, mercury, and arsenic in marine products of commerce from Zhejiang coastal area, China, May 1998. *Bulletin of environmental contamination and toxicology*, 73(3): 583-590.

89. Saha M, Sarkar SK, Bhattacharya B. 2006. Interspecific variation in heavy metal body concentrations in biota of Sunderban mangrove wetland, northeast India. *Environment International*, 32(2): 203-207.

90. Fernandes C, Fontaínhas-Fernandes A, Cabral D, Salgado MA. 2008. Heavy metals in water, sediment and tissues of *Liza saliens* from Esmoriz–Paramos lagoon, Portugal. *Environmental Monitoring and Assessment*, 136(1): 267-275.

91. Batty J, Pain D, Caurant F. 1996. Metal concentrations in eels *Anguilla anguilla* from the Camargue region of France. *Biological conservation*, 76(1): 17-23.

92. Henry F, Amara R, Courcot L, Lacouture D, Bertho ML. 2004. Heavy metals in four fish species from the French coast of the Eastern English Channel and Southern Bight of the North Sea. *Environment International*, 30(5): 675-683.

93. Masoud MS, El-Samra MI, El-Sadawy MM. 2007. Heavy-metal distribution and risk assessment of sediment and fish from El-Mex Bay, Alexandria, Egypt. *Chemistry and Ecology*, 23(3): 201-216.

94. Sarhadizadeh N, Afkhami M, Ehsanpour M, Bastami KD. 2014. Heavy metal pollution monitoring in the northern coast of Hormuz Strait (Persian Gulf): plasma enzyme variations in *Periophthalmus waltoni*. *Comparative Clinical Pathology*, 23(4): 1063-1067.

95. Dalman Ö, Demirak A, Balcı A. 2006. Determination of heavy metals (Cd, Pb) and trace elements (Cu, Zn) in sediments and fish of the Southeastern Aegean Sea (Turkey) by atomic absorption spectrometry. *Food Chemistry*, 95(1): 157-162.

96. Jezierska B, Witeska Mg. 2014. The metal uptake and accumulation in fish living in polluted waters. In: *Soil and water pollution monitoring, protection and remediation*. Springer. ISBN-101-4020-4727-4 (PB).

97. Shivakumar CK, Thippeswamy B, Tejaswikumar MV, Prashanthakumar SM. 2014. Bioaccumulation of heavy metals and its effect on organs of edible fishes located in Bhandra River, Karnatka. International Journal of Research in Fisheries and Aquaculture, 4(2): 90-98.

98. Tayel S, Yacoub AM, Mahmoud S. 2008. Histopathological and haematological responses to freshwater pollution in the Nile Catfish Clarias gariepinus. *Journal of Egyptian Academic Society for Environmental Development*, 9: 43-60.

99. Al-Busaidi M, Yesudhason P, Al-Mughairi S, Al-Rahbi WAK, Al-Harthy KS, Al-Mazrooei NA, et al. 2011. Toxic metals in commercial marine fish in Oman with reference to national and international standards. *Chemosphere*, 85(1): 67-73.

100. Abdallah MAM. 2008. Trace element levels in some commercially valuable fish species from coastal waters of Mediterranean Sea, Egypt. *Journal of Marine Systems*, 73(1): 114-122.

101. Blagojević J, Jovanović V, Stamenković G, Jojić V, Bugarski-Stanojević V, Adnađević T, et al. 2012. Age differences in bioaccumulation of heavy metals in populations of the black-striped field mouse, Apodemusagrarius (Rodentia, Mammalia). *International Journal of Environmental Research*, 6(4): 1045-1052.

102. Kim S-G, Kang J-C. 2004. Effect of dietary copper exposure on accumulation, growth and hematological parameters of the juvenile rockfish, Sebastes schlegeli. *Marine Environmental Research*, 58(1): 65-82.

103. Lapointe D, Couture P. 2009. Influence of the route of exposure on the accumulation and subcellular distribution of nickel and thallium in juvenile fathead minnows (*Pimephales promelas*). *Archives of environmental contamination and toxicology*, 57(3): 571.

104. Spry DJ, Hodson PV, Wood CM. 1988. Relative contributions of dietary and waterborne zinc in the rainbow trout, *Salmo gairdneri*. *Canadian Journal of Fisheries and Aquatic Sciences*, 45(1): 32-41.

105. Xu Y, Wang W-X. 2002. Exposure and potential food chain transfer factor of Cd, Se and Zn in marine fish Lutjanus argentimaculatus. *Marine Ecology Progress Series*, 238: 173-186.

106. Miller PA, Lanno RP, McMaster ME, Dixon DG. 1993. Relative contributions of dietary and waterborne copper to tissue copper burdens and waterborne-copper tolerance in rainbow trout (*Oncorhynchus mykiss*). *Canadian Journal of Fisheries and Aquatic Sciences*, 50(8): 1683-1689.

107. Bordajandi LR, Gomez G, Fernandez MA, Abad E, Rivera J, Gonzalez MJ. 2003. Study on PCBs, PCDD/Fs, organochlorine pesticides, heavy metals and arsenic content in freshwater fish species from the River Turia (Spain). *Chemosphere*, 53(2): 163-171.

108. Asagba SO, Eriyamremu GE, Igberaese ME. 2008. Bioaccumulation of cadmium and its biochemical effect on selected tissues of the catfish (*Clarias gariepinus*). *Fish Physiology and Biochemistry*, 34(1): 61-69.

109. Grotto D, Batista BL, Carneiro MFH, Barbosa Jr F. 2012. Evaluation by ICP-MS of Essential, Nonessential and Toxic Elements in Brazilian Fish and Seafood Samples. *Food and Nutrition Sciences*, 3(09): 1252.

110. Ibrahim IAJ, Zwein LH, Al-Shwaikh RM. 2013. Bacterial and heavy metals analyses in fish at Shawaka area of Tigris River. *Chem Mater Research*, 3(7): 94-99.

111. Mohammadnabizadeh S, Pourkhabbaz A, Afshari R. 2014. Analysis and determination of trace metals (nickel, cadmium, chromium, and lead) in tissues of *Pampus argenteus* and *Platycephalus indicus* in the Hara Reserve, Iran. *Journal of toxicology*, 1-6.

112. Al-Yousuf MH, El-Shahawi MS, Al-Ghais SM. 2000. Trace metals in liver, skin and muscle of *Lethrinus lentjan* fish species in relation to body length and sex. *Science of the Total Environment*, 256(2): 87-94.
113. Canli M, Atli G. 2003. The relationships between heavy metal (Cd, Cr, Cu, Fe, Pb, Zn) levels and the size of six Mediterranean fish species. *Environmental Pollution*, 121(1): 129-136.
114. Bashir FH, Othman MS, Mazlan AG, Rahim SM, Simon KD. 2013. Heavy metal concentration in fishes from the coastal waters of Kapar and Mersing, Malaysia. *Turkish Journal of Fisheries and Aquatic Sciences*, *13*(2): 375-382.
115. Hilala AHA, Ismailb NS. 2008. Heavy Metals in Eleven Common Species of Fish from the Gulf of Aqaba, Red Sea. *Jordan Journal of Biological Sciences*, 1: 13 - 18.
116. Wong MH. 2004. Wetlands ecosystems in Asia: function and management. Elsevier. ISBN 0-444-51691-3.
117. Kruitwagen G, Pratap HB, Covaci A, Bonga SEW. 2008. Status of pollution in mangrove ecosystems along the coast of Tanzania. *Marine Pollution Bulletin*, 56(5): 1022-1031.
118. Amin B, Nurrachmi I. 1999. Mudskipper as bioindicator of heavy metal pollution in Dumai waters. *Jurnal Natur Indonesia (Indonesia)*, 1: 19-24.
119. Ikram MM, Ismail A, Yap CK, Azwady AAN. 2010. Levels of heavy metals (Zn, Cu, Cd, and Pb) in mudskippers (*Periophthalmodon schlosseri*) and sediments collected from intertidal areas at Morib and Remis, Peninsular Malaysia. *Toxicological and Environ Chemistry*, 92(8): 1471-1486.

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