



# INTERNATIONAL JOURNAL OF RESEARCH IN PHARMACEUTICAL SCIENCES

Published by JK Welfare &amp; Pharmascope Foundation

Journal Home Page: <https://ijrps.com>

## Bioaccumulation of heavy metals in orange mud crab (*Scylla olivacea*) from Sungai Merbok, Kedah

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### Article History:

Received on: 04.09.2018  
Revised on: 22.12.2018  
Accepted on: 26.12.2018

### Keywords:

Orange Mud Crab  
(*Scylla olivacea*),  
Heavy metal,  
Bioaccumulation

### ABSTRACT

*Scylla olivacea* is the most prevalent edible mud crab species in Peninsular Malaysia. However, mud crab can accumulate high contaminations from the surrounding environment resulting in health risk to consumers. Thus, this study was conducted to determine the selected heavy metal accumulative concentrations in *Scylla olivacea* from Sungai Merbok, Kedah. Adult crab samples were collected every three months for one year (August 2017, November 2017, February 2018 and May 2018) from selected areas in Sungai Merbok, Kedah. The heavy metal accumulation levels in the crab tissues were determined using Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The concentration of heavy metals ranged from 0 to 4.81 ppm for Chromium (Cr), 0 to 0.49 ppm for Manganese (Mn), 0 to 8.3 ppm for Iron (Fe), 0 to 0.33 ppm for Nickel (Ni), 3.2 to 66.07 ppm for Zinc (Zn), 0.33 to 2.55 ppm for Arsenic (As), 0.11 to 0.77 ppm for Lead (Pb), and below the detectable limit for cadmium (Cd). The mean concentration of Cr, Fe and As were  $1.52 \pm 2.26$  ppm,  $2.09 \pm 4.14$  ppm and  $1.68 \pm 0.95$  ppm respectively, slightly higher than the maximum permissible limits set by Joint Food and Agriculture Organization/World Health Organization Expert Committee on Food Additives (JECFA) and Malaysian Food Regulation. This study indicated that the accumulation of Cr, Fe and As was high in the edible tissues of *Scylla olivacea* from Sungai Merbok, Kedah, which can trigger adverse effects on consumer's health. Hence, a detailed investigation needs to be addressed to determine the pollution status in crabs inhabiting along the Sungai Merbok waters.



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ISSN: 0975-7538

DOI: <https://doi.org/10.26452/ijrps.v10i1.1897>

Production and Hosted by

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### INTRODUCTION

Many species of *Scylla* spp exist in river mouths estuarine habitats, and mangrove forests, the *Scylla olivacea* or the orange mud crab is a large swimming crab (Jirapunpipat *et al.*, 2009). *Scylla olivacea* is the significant crab for commercial activities

with a high price in both the export and domestic markets (Keenan, 2004). Water is an essential component of all organisms worldwide. There are huge amounts of multi-pollutants that entering the water bodies and affecting the ecosystem livings, than including the nature and anthropogenic activities, such as urban runoff, wastewater, industries and agriculture (Chen *et al.*, 2003; Bai *et al.*, 2009; Mohamed., 2012 and Sharif *et al.*, 2015). Annually, high concentrations of pollutants adversely affect the life of the fish and the aquatic environment within short and long-term (Massoud *et al.*, 2006). Heavy metals are one of those pollutants that can suspend in the water or accumulate in the different levels, including sediment or even the organisms' bodies. However, the bioaccumulation of heavy metals in the shellfish bodies including crab, which transition by food-chain and affecting the human beings' health (Cheung and Wang, 2008). Globally,

the concern about the accumulation of heavy metals in the aquatic environments has been rising due to the high consumption of fish and other seafood. Therefore, many reports on heavy metal bioaccumulation in the tissue of these organisms have been conducted (Verbeke *et al.*, 2005).

Because of the rapid developing in industrial activities, the levels of those metals in coastal environments have been reaching alarming rates (Ali and Fishar 2005; David 2003). The continued influence of fish and seafood with high concentrations of these contaminants has made many species a source of dangerous metals which transmission to the human beings, such as arsenic (As), lead (Pb), mercury (Hg), and cadmium (Cd) (Castro-Gonzalez and Mendez-Armenta 2008; Khansari *et al.*, 2005). The study of bioaccumulation levels of the harmful heavy metals in mud crabs is still slightly known. The threat of the accumulation of heavy metals in the mud crab lies in transition to the food chain, which contributes to the formation of direct risk to humans and other living organisms.

## MATERIALS AND METHODS

### Study location

The study was conducted in Sungai Petani, a town located in Kuala Muda District, Kedah, Malaysia (Figure 2.1). This location was chosen because of the abundance of many kinds of crabs' species and is easily accessible. This town is usually abbreviated as Sg. Petani or sometimes termed "SP" and it's said to be the largest town in the state of Kedah followed by the state capital Alor Setar which is 55km away from Sg Petani and 33km northeast of George Town, the capital state city of the neighbouring state of Penang. It's located about 35km from Penang in the northern part of Peninsular Malaysia. The city is also the most significant industrial area near Penang which is the third largest city in Malaysia after Johor Bahru. The population of this city is estimated to be 443, 458 dated back to 2010 census data report. A river popularly known as Merbok River is where the samples of this study collected. Its main sources are at Mount Jerai and Sungkap Forest. Its name came to existence after the confluence of Bangkok and Lalang River. It's around 2.5km wide and is known as the historical river of Lembah Bujang.

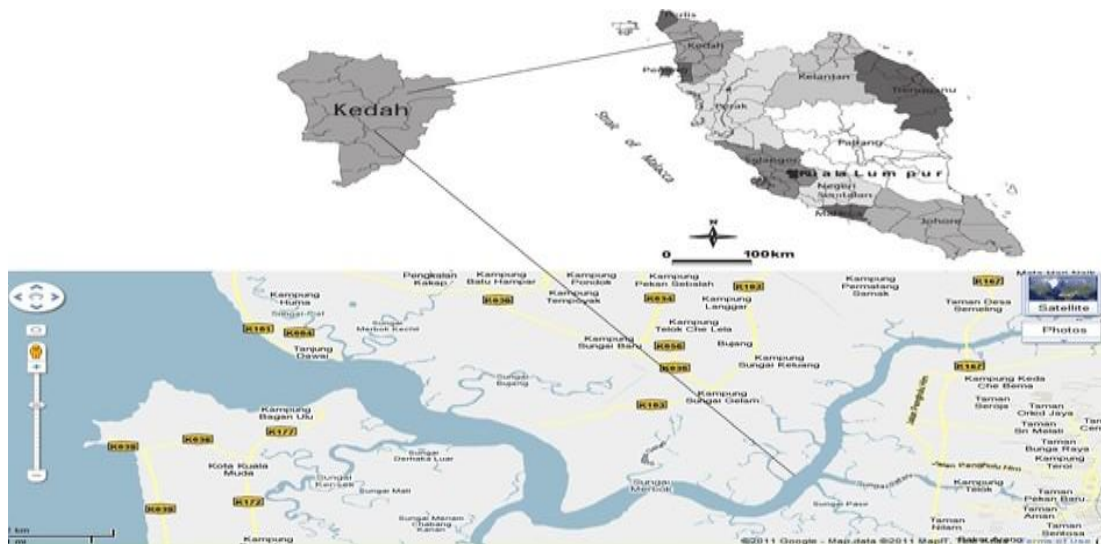
All crabs used in the study from Sungai Petani, a town in Kuala Muda District, Kedah, Malaysia. Samples were collected in batches from first to fourth sampling collection. The crab of *Scylla olivacea* samples was transferred back to the research laboratory of the Faculty of Science and Mathematics. The meats of *S. olivacea* collected from each sampling sites was extracted and sent to a central laboratory to estimate the accumulation of heavy

metals in mud crab *Scylla olivacea* in University Malaysia Pahang. Heavy metal was determined as described by Kamsia *et al.*, (2013) and Ashraf *et al.*, (2012). A concentration of Cr, Mn, Fe, Ni, Zn, As, Cd and Pb was detected by Coupled Plasma- Mass Spectrometry (ICP-MS) with the direct aspiration of the sample in a direct air-acetylene flame.

The metals investigated were wide ranged, but the study focused on eight metals: Chromium (Cr), Manganese (Mn), Iron (Fe), Nickel (Ni), Zinc (Zn), Arsenic (As), Cadmium (Cd) and Lead (Pb). The heavy metal accumulation levels in the crab tissues were then determined using Inductively Coupled Plasma Mass Spectrometry (ICP-MS). The concentration of heavy metals ranged from 0 to 4.81 ppm for Chromium (Cr), 0 to 0.49 ppm for Manganese (Mn), 0 to 8.3 ppm for Iron (Fe), 0 to 0.33 ppm for Nickel (Ni), 3.2 to 66.07 ppm for Zinc (Zn), 0.33 to 2.55 ppm for Arsenic (As), 0.11 to 0.77 ppm for Lead (Pb), and below the detectable limit for cadmium (Cd). The mean concentration of Cr, Fe and as were  $1.52 \pm 2.26$  ppm,  $2.09 \pm 4.14$  ppm and  $1.68 \pm 0.95$  ppm, respectively shown in table 3.1, slightly higher than the maximum permissible limits set by Joint Food and Agriculture Organization/World Health Organization Expert Committee on Food Additives (JECFA) and Malaysian Food Regulation (1996).

Analysing each metal (Table 1), we discovered that Cr concentration varied from 1.19 ppm in August, 4.81 ppm in November, 0.01 ppm in February and 0.06 ppm in May with an average value of 1.52 ppm. The highest value of Cr was 4.81 ppm in November, whereas the lowest value was at 0.01 ppm during February. While the concentration of Mn varied from 0.00ppm in August, 0.61 ppm in November, 0.04 ppm in February and 0.49 ppm in May with an average value of 0.28 ppm. The highest value of Mn was 0.61 ppm in November, whereas the lowest value was at 0.00 ppm during August. Iron (Fe) varied from 0.05 ppm in August, 0.02 ppm in November, 8.30 ppm in February and 0.00 ppm in May with an average value of 2.09 ppm. The highest value of Fe was 8.30 ppm in February, whereas the lowest value was at 0.00 ppm during May. Nickel (Ni) concentration of Ni varied from 0.33ppm in August, 0.00 ppm in November, 0.00 ppm in February and 0.00 ppm in May with an average value of 0.08 ppm. The highest value of Ni was 0.33 ppm in August r, whereas the lowest value was at 0.00 ppm during November, February and May.

Zinc (Zn) varied from 47.80 ppm in August, 37.22 ppm in November, 3.20 ppm in February and 66.07 ppm in May with an average value of 38.57 ppm. The highest value of Fe was 66.07 ppm in February,



**Figure 1: A state map of Kedah, Malaysia showing Sungai Petani where the river Merbok located and was used for sample collections**

**Table 1: Mean concentration values for each metal**

Metals	Aug 2017	Nov 2017	Feb 2018	May 2018	Min	SD	Permissible limit
Chromium (Cr)	1.19	4.81	0.01	0.06	1.52	2.26	1.2*
Manganese (Mn)	0.00	0.61	0.04	0.49	0.28	0.31	10*
Iron (Fe)	0.05	0.02	8.30	0.00	2.09	4.14	0.8*
Nickel (Ni)	0.33	0.00	0.00	0.00	0.08	0.16	0.3*
Zinc (Zn)	47.80	37.22	3.20	66.07	38.57	26.42	100***
Arsenic (As)	1.80	2.55	0.33	2.05	1.68	0.95	1***
Cadmium (Cd)	0.00	0.00	0.00	0.00	0.00	0.00	1***
Lead (Pb)	0.43	0.77	0.11	0.52	0.46	0.27	2***

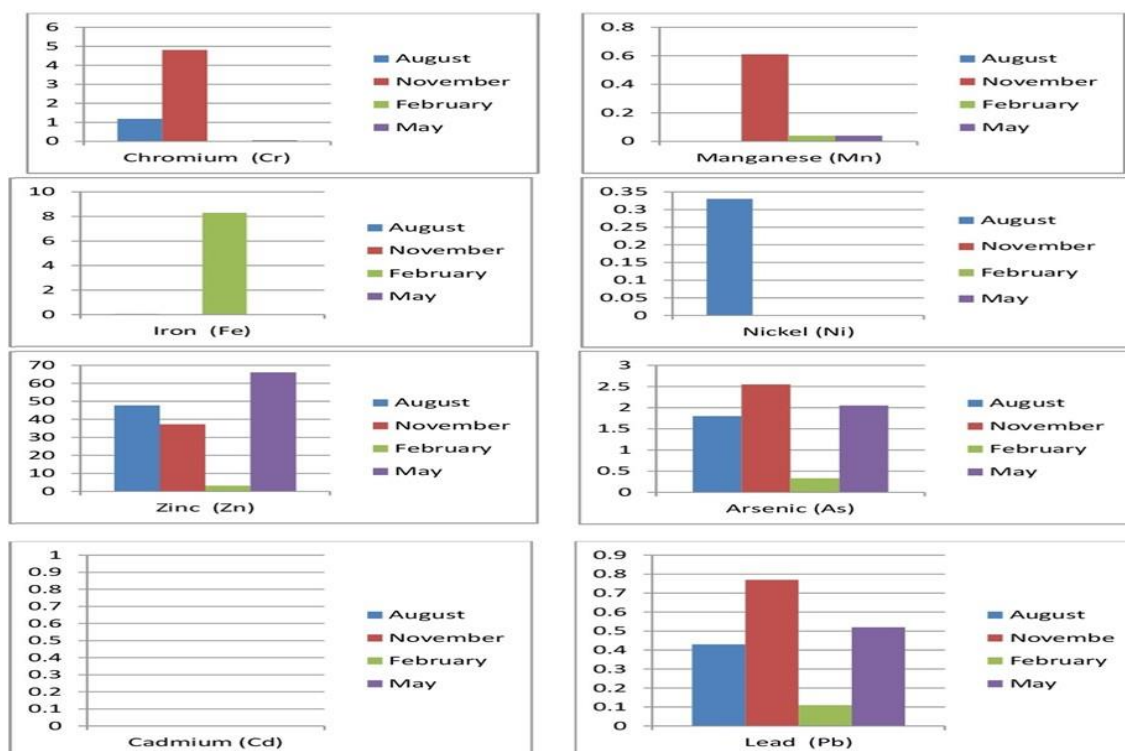
whereas the lowest value was at 3.20 ppm during February. Arsenic (As) varied from 1.80 ppm in August, 2.55 ppm in November, 0.33 ppm in February and 2.05 ppm in May with an average value of 1.68 ppm. The highest value of as was 2.55 ppm in November, whereas the lowest value was at 0.33 ppm during February. Cadmium (Cd) varied from 0.00 ppm in August, 0.00 ppm in November, 0.00 ppm in February and 0.00 ppm in May with an average value of 0.00 ppm. A concentration of Cd in all sampling was 0.00 ppm. Lead (Pb) varied from 0.43 ppm in August, 0.77 ppm in November, 0.11 ppm in February and 0.52 ppm in May with an average value of 0.46 ppm. The highest value of Fe was 0.77 ppm in November, whereas the lowest value was at 0.11ppm during February.

**DISCUSSION**

Of the eight metals studied in the present work, Zn, Ni, Cr and Fe are essential elements while Pb, As, Mn and Cd are non-essential elements for most of the living organisms (Serafim, 2012). Zn is an important element for normal growth, reproduction and longevity of animals, its accumulation in the fish organs was very high when compared with the other four metals. Zn and the other essential metal

was at greater concentration when compared with the other non-essential metals (Pb, As and Cd).

The amounts of heavy metals allowed in food varied from country to country and based on both WHO recommendations and local requirements. Seafood is a good source of dietary copper, which is an essential element available to humans (WHO, 1996). Cadmium is highly toxic; it is associated with nephrotoxic effects particularly long-term exposure may cause bone damage. The threshold concentration of cadmium in seafood design for human consumption set by the European Commission is 0.1mg/gw.w, the guideline limit set for Cd by FAO, (1983) is 0.05mg/kg for fish. According to Jakimska *et al.*, (2011), it is the diet of an animal that dictates its accumulation of metals in its tissue. The more we consume mud crab that shows the type of results above the more exposed to bio-accumulation and its consequences. Cadmium accumulation was generally very low in all three tissues, while Cu and Pb were accumulated at levels between those of Zn and Cd. The non-essential metals (Cd, Pb) have no metabolic role in crustaceans, and the tissue contents of these metals are not regulated.



**Figure 2: Seasonal variation of (Cr, Mn, Fe, Ni, Zn, As, Cd and Pb) concentrations in Mud crab (*Scylla olivacea*)**

Nonessential metals do not present any function for the crustacean's metabolism and are by consequent not regulated by the organism. The amount of Cd and Pb in crustaceans can, thus, serve as an indication of environmental levels of these metals.

Another marine study reveals that the Pb contents were in the range between 0.33 to 0.93 mg/kg in muscles tissues of fish in Black and Aegean seas (Uluozlu *et al.*, 2007). Concentrations of Lead in mud crab were found to be in the range of 0.43 to 0.46 in the present study. Similarly, Çoğun *et al.*, (2017) have also reported that the Pb concentrations in the hepatopancreas of site 2 crabs were several times higher than at site 1 ( $77.75 \pm 3.30$  µg/g dry weight and  $20.25 \pm 3.22$  µg/g dry weight, respectively). Pb levels in gill were two-fold higher at site 2 than at site 1. Zinc concentrations were the highest among the eight metals in all three tissues of crabs collected, and this is in line with the previous study (Çoğun *et al.*, 2017).

Arsenic is a naturally occurring element that is common in soils, water and living organisms. Acute high-level exposure to arsenic can lead to vomiting, diarrhoea, anaemia, liver damage and death. Long term exposure is thought to be linked to skin disease, hypertension, some forms of diabetes and cancer (Centeno *et al.*, 2005). Most arsenic in our diet is present in organic form (WHO, 2011). The present study of arsenic was  $1.68 \pm 0.95$  which is relatively similar to Johnson *et al.*, 2011. Although total as levels in these species mostly exceeded 1.0

µg/g with the highest value of  $3.1$  µg/g wet wt, they may not constitute a risk for human health since most arsenic in marine organisms is in the non-toxic organic form (70-95%) (Maher, 1983).

In general, sediment-inhabiting crabs accumulate heavy metals to high concentrations. Normally acceptable limits for Cd and Pb in crustacean are 3 and 1.5 respectively (USFDA 2003). We found that measured metal concentrations in orange mud crab (*Scylla olivacea*) C. *sapidus* muscles were lower than the range value reported by USFDA (2003) except Zn. The concentration of various metals in the important tissues of crabs is well known (Chou *et al.*, 2002; Firat *et al.*, 2008).

### CONCLUSION

This research has presented data on the concentrations of heavy metals in orange mud crab (*Scylla olivacea*) from Sungai Merbok, Kedah, Malaysia. The objectives of this study were to define the accumulation of the eight heavy metals discussed in the paper. A concentration of metals in crab tissues revealed that Zn, Cr and As are highest whereas Mn, Cd, Pb and Fe were expressed lowly in their tissue. Among the metals analysed Zn, Cr and As we're always highest while Cd, Mn, and Pb was lowest in the mud crab tissues. There were monthly variation and differences for eight metals in the crab tissues. Metal concentrations of tissues were higher in August, November and February and lowered in May and February. The order of metals

concentration in *Scylla olivacea* tissues was Zn >Fe>Cr>As>Pb>Mn>Cd.

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