Bioaccumulation of Selected Metals in the Blood Cockle 
(*Anadara granosa*) from Langkawi Island, Malaysia

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ABSTRACT

Concentration of metals (Zn, Cu and Pb) in the edible portion of commercially important blood 
cockle (*Anadara granosa*) collected from Langkawi Island, Malaysia was analyzed. Samples were 
collected from 3 different stations namely Kuah (KU), Kuala Triang (KT) and Pantai Kok (PK) during 
2009. Heavy metal concentrations were detected using Inductively Coupled Plasma Mass Spectrometry 
(ICP-MS) with respect to the total body burden (i.e., the observed concentration multiplied with total 
flesh weight). The observed mean concentration of Zn, Cu and Pb at Kuah station was 110.75 µg/g, 
12.35 µg/g and 1.73 µg/g respectively followed by Kuala Triang station with mean Zn, Cu and Pb 
concentration of 62.16 µg/g, 6.72 µg/g and 0.06 µg/g respectively. The lowest concentration of selected 
metals was observed in the samples collected from Pantai Kok station with mean Zn, Cu and Pb 
concentrations of 77.50 µg/g, 8.56 µg/g and 0.04 µg/g respectively. The calculated Bio-concentration 
Factor Value (BCF\text{sample-sediment}) showed the resisting the entry of metals or limited ability of *A.granosa* in 
accumulating selected metals from their environment as indicated by the BCF\text{sa-se} value (< 1) except 
the Zn accumulation from Kuah station (BCF\text{sa-se} value = 2.21). However, the observed metal 
concentration in the sample tissue was lower than the national and International permissible standard 
limit for human consumption.

Key words: Heavy metal, *Anadara granosa*, Bioaccumulation, BCF, Toxicity.

INTRODUCTION

Metal accumulations in aquatic organisms especially filter feeders are of serious concern due 
to their higher bioaccumulative capacity from their external environment which could ultimately 
transferred to higher trophic level via biomagnification process. Due to this ability, bivalves 
are widely used as biological indicators for the assessment of contamination especially in coastal
environment and providing a time integrated indication of environmental contamination (Otchere, 2003). It was also well documented that water bodies contaminated by heavy metals may lead to bioaccumulation in the food chain of an estuarine environment. Normally, such contaminants are transported from its sources via river system and deposited downstream (Hong et al., 2003). Bivalves accumulate higher amount of heavy metals from sediments compared to the surrounding water body due to higher concentration of metal bioavailability in sediments, the direct exposure of bivalve to the sediment bottom and filter feeding nature of bivalves. Metal accumulation in the bivalve tissue is also being affected by the seasonal variation in metals, their spawning period, weight of the organism, growth rate, uptake and excretion rates of metals, besides the physicochemical properties of surrounding water body and sediment bed (Ibrahim, 1995; Otchere et al., 2000). He has also observed the magnitude of metal bioaccumulation in the bivalve tissue varies between species to species and their ability/capacity to regulate or accumulate metals (Otchere et al., 2003). Recent studies have clearly shown that of effluents and associated toxic compounds into aquatic systems represents an ongoing environmental problem due to their possible impact on communities in the receiving aquatic water and a potential effect on human health (Canivet and Gibert, 2002). Toxic metals released into the marine environments tend to accumulate in sediments and subsequently are taken up by filter-feeding organisms. Hence the knowledge on the metal accumulation in filter feeding aquatic organisms is of prime importance for their sustainable management and utilization for human consumption.

Anadara granosa commonly known as blood cockle is an important resource, especially in coastal and island populations, where they provide a main table food in south East Asian countries (Bardach et al., 1972) especially in Malaysia. However, the studies on the bioaccumulation of heavy metals in cockles are still scanty (Ibrahim, 1995; Abdullah et al., 2007). Thus we have analyzed the heavy metal pollution level in the edible portion of Anadara granosa collected from Langkawi Island, one of the busiest tourist spot in North West coast of Peninsular Malaysia.

MATERIAL AND METHODS

Sampling sites
Langkawi is one of the major tourist islands in Malaysia and located in the west coast of Peninsular Malaysia. It is part of Kedah state and governed by the Langkawi Development Authority (LADA). The shores around the main island which consist of soft, silty, and fine sand are protected by several smaller islands surround it. The mean daily temperature in this study area varied from 26.6°C to 29.3°C. Meteorological and bathymetric data showed that the numbers of rain days in a month ranged from none (0) to 27 days and the tides along the Langkawi Island are semi-diurnal type (twice daily) and wind speed was mostly prevailed at 0.3 m/s to 3.3 m/s. Langkawi island is influenced by a wind system (South West Monsoon which blows from May to Sept) that blown from the Indian Ocean which bring monsoon season to Langkawi Island and its adjacent areas.

Sample collection and preparation
A total of 45 similar size samples were handpicked during low tide from each station during 2009 (Figure 1) together with the sediment samples in triplicates. Samples were packed in clean plastic bags in iced condition and immediately transferred into laboratory for further analysis. Prior to the analysis bivalve samples were cleaned with running tap water to remove the sand particles and weighed. A 2×2cm size soft tissue was excised from each sample and cleaned with running tap water followed by distilled water to remove the debris and sand particle. All the bivalve samples were made into triplicate and kept in oven at 70°C for 72 hours. Sediment samples were air dried for 7 days and transferred to the oven for complete drying process.

Analytical Method
Dried bivalve samples were ground to a fine powder using sterile mortar and pestle and a constant weight of 0.5g dried tissue were weighed and transferred to the Teflon beakers for acid digestion process. Samples for heavy metal analysis (including blanks) were digested in Teflon beakers containing 10 mL of concentrated HNO₃, 2 mL of concentrated H₂SO₄, 3 mL of H₂O₂ and 2 mL of HCl on the hot plate at 60°C for about two hours to ensure complete digestion of all organic matter.
(Kamaruzzaman et al., 2007). After a complete digestion, the solution was cooled at room temperature and the samples were filtered and transferred to falcon tubes and double washed with ultrapure water to ensure the complete transfer of digested sample. The digested sample was made up to 50 mL using 5% HNO₃ as the diluent. The standard reference material (SRM 1566b, Oyster Tissue [for bivalve sample] and estuarine sediment, SRM 1646a [for sediment sample]) were also subjected to the same procedure. Closed digestion method was adopted to detect the heavy metal level in sediment samples as explained by Kamaruzzaman et al., (2008). The concentrations of heavy metals were detected by using Inductively Coupled Plasma-Mass Spectrometry (ICP-MS). The results of the standard reference material were in good agreement with the certified values (Table 1).

**Data analysis**

Bio-concentration factor (BCF<sub>sample/sediment</sub>) value was calculated by finding the ratio of metal concentration in the mollusk to that in sediment. BCF value > 1 is indicating that the mollusk contained more metals than the sediment. Whereas BCF value < 1 is a sign of limited ability of selected samples in accumulating the heavy metals from their surrounding environment. Analysis of Variance test (ANOVA) test was performed to determine the significant variation in metal accumulation by bivalve samples collected from different stations. The detected concentration of metals in the sample tissue was compared with previous studies, national and international standard for human consumption. (BCF). The detected concentration of selected metals in the sample was compared with national and international permissible limits for human consumption besides comparing with other studies.

**RESULTS AND DISCUSSION**

The detected concentration of Zn, Cu and Pb in the soft tissue of *Anadara granosa* and sediment samples collected from different sampling sites is given in Table 2. The mean concentration of Zn in *A.granosa* tissue was higher in Kuah station samples (110.75 ± 11.00 µg/g) followed by Pantai Kok samples (77.50 ± 5.62 µg/g) and Kuala Triang samples (62.16 ± 15.35 µg/g). The mean concentration of Zn in sediment samples was higher in Pantai Kok station (102.6 µg/g) followed by Kuala Triang (84.26 µg/g) and Kuah (50.12 µg/g).

<table>
<thead>
<tr>
<th>Stations</th>
<th><em>Anadara granosa</em> (µg/g dry weight)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Zn</td>
</tr>
<tr>
<td>Kuah</td>
<td>110.75 ± 11.00</td>
</tr>
<tr>
<td>Kuala Triang</td>
<td>62.16 ± 15.35</td>
</tr>
<tr>
<td>Pantai Kok</td>
<td>77.50 ± 5.62</td>
</tr>
<tr>
<td>Sediment (µg/g)</td>
<td></td>
</tr>
<tr>
<td>Kuah</td>
<td>50.12</td>
</tr>
<tr>
<td>Kuala Triang</td>
<td>84.26</td>
</tr>
<tr>
<td>Pantai Kok</td>
<td>102.6</td>
</tr>
</tbody>
</table>

Table 1: Recovery percentage of selected metals observed in ICPMS reading using Oyster Tissue, 1566b as a reference standard material

<table>
<thead>
<tr>
<th>Elements</th>
<th>Certified value</th>
<th>Analysis value</th>
<th>Recovery (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cu</td>
<td>71.6 ± 1.6</td>
<td>70.6 ± 0.19</td>
<td>98.60</td>
</tr>
<tr>
<td>Zn</td>
<td>1424 ± 46</td>
<td>1485 ± 12</td>
<td>104.28</td>
</tr>
<tr>
<td>Pb</td>
<td>0.308 ± 0.009</td>
<td>0.298 ± 0.210</td>
<td>96.75</td>
</tr>
</tbody>
</table>
Calculated $BCF_{\text{sample-sediment}}$ value showed that *A.granosa* samples from Kuah station tend to accumulate higher amount of Zn from sediment environment ($BCF_{\text{sa-se}} = 2.21$) where as *A.granosa* from other sampling stations have limited ability to accumulate Zn from sediment environment ($BCF_{\text{sa-se}} = < \sim 0.74$). Higher concentrations of Zn in sampling stations are identified mostly caused by its application as an anticorrosive agent in boat paint (Greenwood and Earnshaw, 1997), and used as antifouling paint (Konstantinou and Albanis, 2004). There are many tourist boats as well as fishing boats in these areas that might introduce Zn into the environment. Kamaruzzaman et al., (2006) stated that painting activities of fishing boats and the used of antirust paint in fishing and shipping industries may affect the levels of Zn in the sediments.

<table>
<thead>
<tr>
<th>Elements</th>
<th>Present study</th>
<th>Other study (Abdullah et al., 2007)</th>
<th>National and International Standards</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Kuah Kuala Triang Pantai Kok</td>
<td>Likas estuary Kota Belud estuary</td>
<td>FAO/WHO MFR (ppm) (ppm)</td>
</tr>
<tr>
<td>Cu</td>
<td>12.35 6.72 8.56</td>
<td>6.89 ± 1.66 3.51 – 9.80</td>
<td>10 30</td>
</tr>
<tr>
<td>Zn</td>
<td>110.75 62.16 77.50</td>
<td>96.0 ± 17.0 68.8 – 137.7</td>
<td>150 100</td>
</tr>
<tr>
<td>Pb</td>
<td>1.73 0.06 0.04</td>
<td>4.74 ± 2.37 0.51 – 14.46</td>
<td>1.5 2</td>
</tr>
</tbody>
</table>

Highest concentration of Cu in tissue samples were observed in Kuah station (12.35 µg/ g dw), followed by Pantai Kok (8.56 µg/g dw), and Kuala Triang (6.72 µg/g dw), whereas the concentration in sediment was higher at Kuah station (38.24 µg/g) followed by Pantai Kok (26.99 µg/g) and Kuala Triang (18.23 µg/g). Bivalve from harbor areas are typically high in copper (WHO, 1998), reflecting the increased environmental abundance of this element from sources such as algacides and anti-fouling paints. The higher concentrations of Cu in Kuah samples might probably due to existing activities such as loading and off loading of fishes, cleaning and maintenance of boats, antifouling paint applications, and fuelling the crafts. $BCF_{\text{sample-sediment}}$ value (< 0.4) clearly showed the limited intake of Cu by *A.granosa* samples from all the sampling areas.

The highest concentration of Pb was found at Kuah samples (1.73 µg/ g dw) and lowest was 0.04 µg/ g dw at Pantai Kok. Although blood cockle tissues have relatively low Pb contents, their Pb compositions reflect mixing of natural Pb with modern anthropogenic Pb emission sources (high Pb level in sediment). These emission sources include unleaded automotive gasoline, diesel fuel and leaded aviation gasoline (Jalal et al., 2009). However, the calculated BCF value proved the least Pb accumulative nature of *A.granosa* species from the sampled stations. The flow of heavy metal in *A.granosa* tissue and sediment samples were in Zn > Cu > Pb in all the stations which is well corresponded with previous studies (Abdullah et al., 2007). It was also well documented that organisms tend to accumulate light metals such as Zn, Cu and Fe in higher concentration in their tissue than the heavy metals such as Pb, Cd and Hg due to their significant role as a precursors in various enzymatic reactions (White and Rainbow, 1985; Mizrahi and Achituv, 1989; Kuzmina and Ushakova, 2008).

Similar results were reflected in this study where the bioaccumulation of Zn and Cu was multifold higher in the sample tissue than Pb accumulation. There was no significant variation in the metal concentration observed between the bivalve samples collected from Kuala Triang and Pantai Kok, where as it significantly varied with samples from Kuah stations (P < 0.05). The observed highest
concentration of Zn in the *A. granosa* samples from Kuah station (110.75 µg/g dw) has exceeded the national maximum permissible limit for human consumption (MFR, 1985), however it is still accepted for human consumption based on international standard (FAO/WHO, 1984). The detected concentration of Zn in present study was also higher compared to previous studies (Table 3). In contrast to Zn level, Cu and Pb concentration in the sample tissue exceeded international standard limit, while it is still under national permissible standard for human consumption. However, Pb concentration in the samples from present study was comparatively lower than the samples from East Malaysian (*i.e.*, Sabah samples), where as there was no significant variation in Cu concentration between Peninsular and East Malaysian samples.

**CONCLUSION**

The findings of this study showed that the *Anadara granosa* from Langkawi coastal waters tend to accumulate heavy metals lower than the national and international permissible limits and hence safety to be used for human consumption. It also proved the limited ability of *A. granosa* in accumulating various heavy metals from its habitat in different rates. Nevertheless, high concentration of metals in surrounding sediment might partially from anthropogenic activities which would reflect in metal levels in aquatic organisms especially bivalves. Hence, constant monitoring of heavy metal pollution in Langkawi Island would help in upgraded environmental management programs.

**ACKNOWLEDGEMENTS**

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