Bioavailability and Variability of Cd, Pb, Zn, and Cu Pollution in Soft Tissues and Shell of *Saccostrea cucullata* Collected from the Coast of Qeshm Island, Persian Gulf, Iran

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Received: 19.01.2013 *Accepted: 09.03.2013*

ABSTRACT

Background: Marine pollution is a global environmental problem that its monitoring by ideal biomonitors is of great importance. Marine organisms, especially mussels, have the ability to accumulate metals from the environment; they can be considered as a biomonitoring agent.

Methods: In this study, concentrations of heavy metals were measured in Saccostrea cucullata collected from seven sites on Qeshm Island's Coast. To achieve a digesting sample, each soft tissue was obtained and each of the shell homogeneous powders, 0.8 g and 1 g, respectively, were mixed with 10 mL HNO3 (69%) and poured into a PTFE digestion vessel. The prepared samples were evaluated for Cd, Cu, and Zn by using a flame AAS Model 67OG and for Pb by using a graphite furnace AAS.

Results: The distributions of metals between soft tissues and shells were compared in each sampling site. For seven sites, Cd, Zn, and Cu levels in soft tissues were higher than in the shells, but Pb level was higher in the shells than in the soft tissues. In addition, the results indicated the coefficient of variation (CV) in the soft tissues was lower than the shells for Cd, and in the shells lower than the soft tissues for Pb, whereas the CV values were different in both the soft tissues and shells for Zn and Cu.

Conclusion: The results of this study support using these materials in S. cucullata for biomonitoring. Shells are appropriate for monitoring Pb contamination, and the soft tissues are more apt for monitoring Cd, Zn, and Cu contamination.

Keywords: Biomonitoring, Heavy Metals, Persian Gulf, Qeshm Island, Saccostrea cucullata.

IJT 2013; 836-841

INTRODUCTION

Marine pollution is a global environmental problem that its monitoring by ideal biomonitors is of importance [1, 2]. A biomonitor is described as an organism which can accumulate heavy metals in its tissues [3]. Marine organisms, especially mussels, have the ability to accumulate metals from the environment in their tissues wherever they live [4]. Using mussels as a monitor for marine pollution has been recommended by several researchers [5-7]. Mussels including *S. cucullata* have: 1) the ability to accumulate most of the heavy metals at much higher levels than those found in the water column, because they are filter feeders and filter large volumes of seawater and concentrated metals and other pollutants, so they are representative of the pollution of an area, 2) widespread distribution, 3) sessile behavior or sedentary, and thus easy to sample, 4) the ability to sequester some metals and their lipophilic properties, and 5) the capacity to provide sufficient tissues for contaminant analysis [8,9]. Therefore, they can be designated as an ideal biomonitor. Several studies [10-13] have reported the comparisons of metal levels in the mussels' tissue in

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determining the potential use of shell or soft tissue as a biomonitoring material for heavy metals. The aim of this study was to see if the shell or the soft tissue of *S. cucullata* is a better biomonitoring material for Cd, Pb, Cu, and Zn.

MATERIALS AND METHODS

S. cucullata samples were collected in May 2010 from seven sites on the northern and the southern intertidal coast of Qeshm Island on the Persian Gulf, Iran (Table 1 and Figure 1). At each sampling site, 15 to 23 *S. cucullata* samples of similar sizes were collected by hand from the intertidal coast region, stored in plastic zip bags, and transported to the laboratory under ice. The samples were, then, stored in the laboratory at -20 °C until further analysis. Frozen rocky oysters (S. *cucullata*) were defrosted at room temperature before opening. In the laboratory, the soft tissues of rocky oysters were carefully separated from the shells. Then the shell length of each sample was measured by caliper as displayed in Table 1. After cleaning shells with a jet of tap water, they were washed with deionized distilled water (DDW) and 0.5% of concentrated HNO_3; then tissue rocky oysters samples were dried in an oven at 105 ºC for 72 h in order to achieve constant dry weights (dw). After washing and drying, the soft tissues were individually pulverized in an agate mortar and the shells were powdered by a mixer mill. Eventually, 0.8 g of each soft tissue and 1 g of each of the shell homogeneous fine powders were stored in polythene bags for subsequent analysis [14].

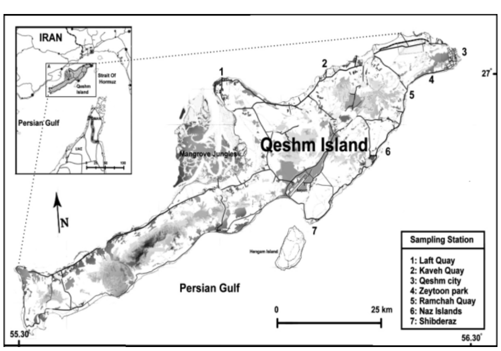

Figure 1. Location of all sampling sites for *S. cucullata* in the northern and the southern intertidal Coast of Qeshm Island on the Persian Gulf, Iran.

Table 1. Location of sampling sites for *S. cucullata*, number of samples analysed (N), and shell length (mm).

Site description	Shell length (min–max)	N	Location	Sampling Stations
a busy jetty	42.61 – 54.53	15	26°56'37''N, 55°45'35''E	**1**
a jetty	36.75 – 51.39	20	26°55'19''N, 55°57'17''E	**2**
urban area	38.09 – 49.73	18	26°56'25''N, 56°16'31''E	**3**
urban area	38.66 – 51.51	16	26°55'28''N, 56°16'04''E	**4**
recreational area	37.93 – 50.19	19	26°53'27''N, 56°09'37''E	**5**
a busy jetty	36.26 – 49.23	23	26°48'49''N, 56°06'56''E	**6**
Pristine area	37.65 – 48.44	20	26°41'16''N, 55°55'45''E	**7**

To achieve a digesting sample, each soft tissue was extracted and each of the shell homogeneous powders, 0.8 g and 1 g, respectively, were mixed with 10 mL HNO3 (69%) and poured into a PTFE digestion vessel. For 1 hour, the samples were heated in a hot-block digester at a temperature of 40 ºC. Eventually, those fully digested were heated for 3 hours in a hot-block digester at a temperature of 140 ºC. The digested samples were, then, diluted to 25 mL volume with DDW. After filtration, the prepared samples were evaluated for Cd, Cu, and Zn by using a flame atomic absorption spectrophotometer (Model 67OG) and concentration of Pb was measured by using a graphite furnace atomic absorption spectrophotometer (Model 67OG). Analysis of the samples was performed in the Laboratory of Tarbiat Modares University, Iran [15].

All statistical analyses were performed using SPSS software 17.0. To determine the coefficient of variation (CV), MINITAB statistical package 14.0 was employed with the untransformed data. Nonparametric comparisons (Wilcoxon Signed Rank Test) were applied to test the differences among metal concentrations in different tissues of *S. cucullata*. Level of significance for all tests was set at 0.05.

RESULTS

The results obtained for concentrations of four metals (Pb, Cd, Cu, and Zn) measured in the shells and soft tissues, and the standard length of mussels sampled from seven sites of Qeshm Island's coast are presented in Table 2. Heavy metal concentrations in the soft tissue of *S. cucullata* were compared to the mussel shell, and the ratios of shell/soft tissue were calculated and results are presented in Table 2. The metal levels in the soft tissue of *S. cucullata* from the seven sites ranged from 0.661 to 2.403 µg g^{-1} for Cd, 0.153 to 0.601 µg g^{-1} for Pb, 448.681 to 2445.761 µg g^{-1} for Zn, and 32.852 to 282.201 µg g^{-1} for Cu, while those in the shells ranged from 0.005 to 0.017 µg g^{-1} for Cd, 0.172 to 2.201 µg g^{-1} for Pb, 14.113 to 190.701 µg g^{-1} for Zn, and 1.132 to 9.754 µg g^{-1} for Cu. Accordingly, metal concentrations in the soft tissues in comparison with the mussel shells from a total of seven sites were shell < soft tissue for Cd, shell > soft tissue for Pb, shell < soft tissue for Cu, and shell < soft tissue for Zn. These results indicated that the concentrations of Cd, Pb, Cu, and Zn in the soft tissues and the shells of *S. cucullata* were different. Also, according to Table 2, the comparison between metals concentrations in shells and tissues were as follows: Cd concentrations in the shells were 0.003 to 0.025 times lower than in the soft tissues, Pb concentrations in the shells were 1.155 to 3.728 times higher than in the soft tissues, Zn concentrations in the shells were 0.009 to 0.275 times lower than in the soft tissues, and Cu concentrations in the shells were 0.007 to 0.296 times lower than in the shells of *S. cucullata*.

The results presented in Table 3 showed that the coefficient of variation (CV) (%) in the soft tissues were lower than the shells of *S. cucullata* in each sampling site for Cd. The results also showed lower degrees of variability in the shells than the soft tissues of *S. cucullata* in each sampling site for Pb. Furthermore, CV values (%) were different in the soft tissues and the shells of *S. cucullata* in each sampling site for Zn and Cu. The lower CV indicates that there is more precision in the determination of the biomonitoring material for heavy metals in the shells or soft tissues.

Table 2. Mean concentrations (µg g^{-1}dry weight) of Cd, Pb, and Zn in total soft tissue and total shell of *S. cucullata* collected from the northern and the southern intertidal Coast of Qeshm Island on the Persian Gulf, Iran.

Cu_{Sh}/Cu_{ST}	Cu_{ST}	Cu_{Sh}	Zn_{Sh}/Zn_{ST}	Zn_{ST}	Zn_{Sh}	Pb_{Sh}/Pb_{ST}	Pb_{ST}	Pb_{Sh}	Cd_{Sh}/Cd_{ST}	Cd_{ST}	Cd_{Sh}	Stations
0.014	282.201	4.091	0.009	2445.761	21.951	3.728	0.601	2.201	0.009	1.301	0.012	**1**
0.007	163.712	1.132	0.015	1136.292	17.432	2.509	0.512	1.282	0.025	0.661	0.017	**2**
0.019	127.831	2.431	0.031	448.681	14.113	3.627	0.515	1.851	0.016	1.012	0.016	**3**
0.296	32.852	9.754	0.095	1301.702	123.821	1.476	0.213	0.314	0.010	1.306	0.014	**4**
0.071	50.284	3.565	0.275	693.611	190.701	1.185	0.271	0.322	0.003	1.910	0.005	**5**
0.086	33.451	2.911	0.076	807.333	62.012	1.155	0.454	0.521	0.004	2.403	0.009	**6**
0.219	40.523	8.902	0.103	1255.551	129.631	1.133	0.153	0.172	0.008	1.240	0.010	**7**

Sh: Shell, ST: Soft tissue

Table 3. Comparison between coefficients of variation (CV %) of heavy metal concentrations in the soft tissues and the shells of *S. cucullata* collected from the Coast of Qeshm Island on the Persian Gulf, Iran.

Zn(Sh)	Zn(ST)	Cu (Sh)	Cu(ST)	Pb(Sh)	Pb (ST)	Cd (Sh)	Cd(ST)	Stations
27.2	19.8	75.6	16.5	29.9	34.7	35.9	12.3	**1**
32.2	38.1	76.4	22.6	37.7	40.5	39.4	32.7	**2**
25.7	18.3	122.5	18.3	24.3	28.4	44.6	40.1	**3**
41.2	62.9	77.4	26.6	29.1	108.4	54.1	17.2	**4**
17.1	74.2	26.6	32.2	77.1	114.8	100.7	27.6	**5**
22.9	50.8	55.7	34.5	61.7	62.2	66.4	44.5	**6**
38.9	42.7	50.6	37.5	40.4	74.2	40.7	16.1	**7**

Sh: Shell, ST: Soft Tissue

DISCUSSION

The results of this study indicated that in each of the seven sites, the concentration of Cd in the soft tissues of *S. cucullata* was significantly higher than that of the shells (Table 2). For Cd, in each of the seven sites, the CV values (%) in the soft tissues were lower than those for the shells (Table 3). Therefore, Cd concentrates in soft tissue more than in shell, and the lower degrees of Cd variability (CV) in the soft tissues showed soft tissue to be generally a more sensitive and accurate biomonitoring material for Cd than the shell of *S. cucullata*. Several studies have shown that in the species examined in the present study (*S. cucullata*) and some other oysters (e.g. *Crassostrea virginica*), the ability exists to concentrate Cd to a great degree in soft tissues [16,17]. Passive mechanism may be the way for the uptake of Cd by the oysters [18]. Furthermore, oysters are capable of accumulating extremely high levels of Cd in soft tissue portions [19].

The results presented in Tables 2 and 3 showed that the shells of *S. cucullata* could be a suitable and worthy biomonitoring material, especially for Pb since the shells would concentrate higher levels of Pb than the soft tissue and the lower degrees of variability (CV) occurred in the shells compared to the soft tissues of *S. cucullata* in each sampling site for Pb. A previous study demonstrated that a higher concentration of Pb was found in the shells than the soft tissues, and there also was found a lower variability for Pb concentration in the shells than in the soft tissues in many species of mussels, including *Saccostrea cucullata*, *Saccostrea camercialis*, *Mytilus edulis*, *Perna viridis*, and *Crassostrea virginica* [20-22]. It is probably for this reason that high concentrations of Pb in the shells due to the crystalline structures of the shell matrix have a higher capacity for absorption of these metals compared to the soft tissues [23].

Mussels can tolerate very high concentrations of Zn and Cu (without apparent detrimental effects); as a result, these metals are essential elements for mussels [24,25]. Cu makes haemocyanin, which serves as a respiratory pigment for mussels, and Zn is required for metabolism in soft tissues [26]. The results of the present study showed very high concentrations of Zn and Cu in the soft tissues in comparison with the shells in each sampling site (Table 2). The results of previous studies agree with the results of this study, namely, high concentrations of Zn and Cu found in the soft tissues than in the shells in *S. cucullata* and various mussels [6,9]. However, CV values (%) were different in the soft tissues and shells of *S. cucullata* for Zn and Cu in each sampling site shown in Table 3. Hence, the question arises if the shell or the soft tissue of *S. cucullata* is a better biomonitoring material for Zn and Cu. Based on the very high concentrations of Zn and Cu in the soft tissues and not in the shells of *S. cucullata* in each sampling site, the soft tissues of *S. cucullata* can be used as a better biomonitoring material for Zn and Cu.

CONCLUSION

In summary, the findings of this study show the presence of Cd, Pb, Cu, and Zn concentrations in the soft tissues and the shells of *S. cucullata* from the northern and the southern intertidal coast of Qeshm Island, Persian Gulf, Iran, and specify which tissues of *S. cucullata* are more useful as a biomonitoring material for these heavy metals (Cd, Pb, Zn, and Cu). Also, the obtained results show that the soft tissue of *S. cucullata* is a more sensitive biomonitoring organ for Cd compared to the shell, whereas for Pb, the shell of *S. cucullata* was a more suitable biomonitoring organ than the soft tissue. Finally, the very high concentrations of Zn and Cu found in the soft tissue rather than the shell of *S. cucullata* show that the soft tissue can be used as the biomonitoring material of choice for Zn and Cu.

ACKNOWLEDGEMENTS

This study was supported by the Ministry of Science and Technology, Iran, which provided partial funding for this project. The authors would like to thank Miss Ansari, Mr Barani, and Mrs. Rahimi from the Faculty of Natural Resources and Marine Sciences, Tarbiat Modares University, Tehran, Iran, for their kind assistance in the collection of oyster samples.

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