Journal of the Persian Gulf
(Marine Science)/Vol. 4/No. 14/December 2013/7/39-45

Investigation of Body Size Effect on Bioaccumulation Pattern of Cd, Pb and Ni in the Soft Tissue of Rock Oyster *Saccostrea cucullata* from Laft Port

Alavian Petroody, Somayye sadat[1]; Hamidian, Amir hossein[1*]; Ashrafi, Sohrab[1]; Eagderi, Soheil[2]; Khazaee, Manoochehr[1]

1- Dept. of Environment, Faculty of Natural Resources, University of Tehran, IR Iran
2- Dept. of Fisheries, Faculty of Natural Resources, University of Tehran, IR IRan

Received: July 2013 *Accepted: November 2013*

Abstract

Nowadays, contamination of heavy metals in aquatic ecosystems is one of the major world problems in developing and developed countries. Among various aquatics, shellfish species, especially bivalves can be used as bioindicator, because of purification system, potential bioavailability of impurities in environment as well as accumulation of some heavy metals in their body. Among bivalves, oysters are the most considered in research due to their nutrition importance. In this research, the concentrations of cadmium, nickel and lead were measured in rock oyster *Saccostrea cucullata* soft tissue (sizes from 2 to 7 cm) located in Laft Port using an ICP-OES instrument. Metal concentrations (µg/g DW for length class) of Cd, Pb and Ni were studied in *S. cucullata* to find the relationship with body size. The results of study revealed that smaller mussels in comparison to larger mussels showed higher concentrations of Ni and Pb. However, there was no significant difference between large and small oysters for Cd.According to EPA, FDA standards, the mean concentrations of Cd and Pb in the three categories of length were higher than the permissible limit and as such, its consumption could pose risk to humans and other organisms.

Keywords: *Body size, Metal concentrations, Rock oyster, Laft Port.*

1. Introduction

Aquatic environments receive a large volume of heavy metal pollutants, hydrocarbons, pesticides and organic materials derived from domestic sewage, industrial, mining and agricultural activities in every day period (Canali et al., 1998). Among aquatic ecosystems, coastal zones, estuaries, coral reefs and mangroves, are the most important and productive marine ecosystems. Although, most of aquatic and terrestrial food chains are associated directly or indirectly with these ecosysted, in recent years, these vital ecosystems have been exposed to oil pollution (Butet et al., 2004).

Heavy metals are ecologically important to be investigated, due to aggregation behavior and their toxicity, which results in lower biodiversity in marine ecosystems. Moreover, consuming of contaminated seafood could endanger human health. Heavy metals are persistent pollutants and

* *E-mail: a.hamidian@ut.ac.ir*

noteasily decomposed biologically (Demora et al., 2004). Due to metal toxicity and their ability to accumulate in living organisms, pollution of heavy metals are serious problems (Usero et al., 2005). In food chains, heavy metals can be transferred to higher food levels. Due to accumulation and bio-magnification, the concentrations of pollutants in aquatic bodies are much higher than the environment. Because many marine species are eaten by humans, knowing the normal concentration of metals or their minimum steady concentration in a marine environment, is essential (Ruelas-Inzunza & Paez-Osuna., 2000).

Heavy metals in marine ecosystems are absorbed by bivalves directly in gills through breathing or indirectly as a result of digested food particles (Clark, 2001). Metal absorption ability of bivalves through gills are higher than gastrointestinal tract. During the respiration process, a large volume of water passes through the gills and contaminants are easily absorbed due to high absorption potential of gills (Tinsley, 1979). Since, marine bivalves can filter large amount of water through their gills (Naimo, 1995), which are valnerable to be exposed to these compounds.

By measuring the concentration of heavy metals in soft tissues of bivalves, they could be used as bioindicator of heavy metals and coastal environment pollution (Yap et al., 2006). However, the accumulation of heavy metals in tissues of bivalves is also affected by number of intrinsic and extrinsic factors. The extrinsic factors include, spawning season (Lobel et al., 1991) and bivalve size (Lobel et al., 1991; Riget et al., 1996). Researchers have shown that body size might change the absorption of heavy metals (Lobel et al., 1991; Riget et al., 1996). It is clear that body size can affect the rates of metal adsorption and desorption (Phillips & Rainbow., 1993). For instance, effects of body size on physiological changes, such as pumping rate, filtration and respiration in the bivalve *Mytilus edulis* have been reported (Jones et al., 1992). The relationship between heavy metal concentrations and body size of temperate climate mollusks have been well documented (Lobel & Wright., 1982), but such information for tropical and sub-tropical species is rather limited. Therefore, the purpose of this study is to provide relative information to understand physiological strategies of Ni, Cd and Pb accumulation in relation to body size of the rock oyster *S. cucullata* in coast of Laft port, located in the Qeshm Island. The present study was conducted to investigate the effects of length on the accumulation of Ni, Cd and Pb in the rock oyster *S. cucullata*.

2. Materials and Methods

The Laft port is located in Qeshm Island (26˚56’ N, 55˚43’E) (Fig. 1). In the late summer of 2011, 200 oysters (in different sizes) were collected using a chisel and a hammer. Collected specimens were washed with seawater to remove their mud. Samples were placed in a thermal isolation polystyrene containing ice bags and transported into the Environmental Pollution Laboratory at University of Tehran (Fig. 2). In the laboratory, the soft tissues of oysters were dissected by an acid washed plastic knife and located into conical flasks. All glassware used in this study were washed three times with concentrated nitric acid and rinsed three times with distilled water (Einollahi Peer et al., 2010). Soft tissues were weighed to the accuracy of 0.001 g. The samples were placed in an oven for 48 h in a temperature of 110˚C and dry weights of the samples recorded. The dried samples were ashed using a furnace at 450˚C for 72 h (The temperature of the furnace was started at 50˚C and increased to 450˚C in 50˚C intervals in hour). The ashed samples were digested by 10 mL concentrated nitric acid (Merck, Germany) on a hot plate and after acid evaporation the remains were diluted into 25 mL of 1% nitric acid. The concentrations of Cd, Ni and Pb in the solutions were measured by an ICP- OES (GBC Integra XL, Australia) instrument.

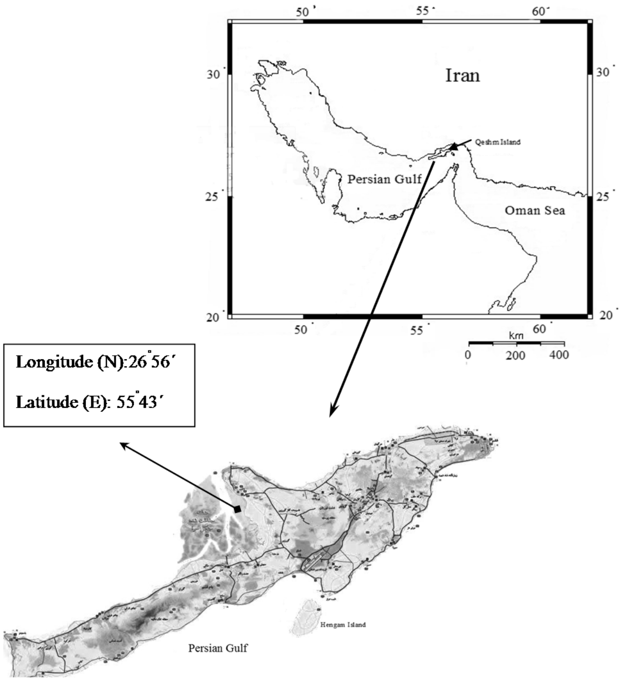

Fig. 1: Location of study area

Fig. 2: *Saccostrea cucullata*

For biometry purposes, some pictures were taken from the shells and transferred to a computer. The maximum lengths of the shells were measured with Image-j software. The lengths were classified in three categories; small (lengths of 2 - 3 cm), medium (lengths of 4 - 5 cm) and large (lengths of 6 - 7 cm), and for each length category, 20 samples were selected from 200 digested samples randomly.

Data normality and homogeneity were examined by Kolmogorov-Smirnov Test. The statistical methods One-way-ANOVA and Duncan's test were used for general and multiple comparisons (at a 99% of confidence), respectively. All statistical analyses were performed by SPSS16 software.

2. Results

The results of the morphometric measurements indicated that shell lengths ranged from 2 to 7 cm and tissue weights from 0.107 to 6.352 g. Results (Table. 1) showed significant relationships between lengths and the concentrations of Pb and Ni ($P < 0.01$), but no significance correlation between length and Cd concentrations in the tissues were observed. The results of Duncan Test (Table. 2) revealed that shellfish species with small shells had the highest concentrations of Pb and Ni. Due to shell size length, two different classes of shells based on Pb and Ni concentrations were recognized.

Table 1. Results of ANOVA: length effects on metal concentrations (Ni, Cd and Pb)

Factor	Metal	df	F	P
Length	Ni	59	9.391	0.001
Length	Cd	59	0.480	0.623
Length	Pb	59	7.450	0.002

Table 2: The average concentration of metals in the soft tissue of oyster (µg/g dry weight)

Length category \ Metal	Ni	Cd	Pb
1	74.68 ^{a}	5.65 ^{a}	18.65 ^{a}
2	35.61 ^{b}	5.36 ^{a}	9.92 ^{b}
3	30.67 ^{b}	4.76 ^{a}	9.19 ^{b}

Letters in the columns are in relation to the result of Duncan Test. lack of similar letters in each column indicates significant difference at 1% level (a > b).

The comparison of Pb, Ni and Cd concentration in the muscle of edible oyster and levels of regulation and safety tips are presented in Table 3.

3. Discussion

In this research, the effects of shell size on Ni and Pb concentrations in soft tissues of oyster *S. cucullata* were investigated. The results indicated that Ni and Pb concentrations in small oysters (class one) were significantly higher than those of large oysters (classes 2 and 3). Previous studies reported negative relationships between body size and accumulation of Pb and Ni and stated that concentrations of metals decreased in oysters with increasing body size (Williamson, 1980; Amiard et al., 1986; Martincic et al., 1992) . Williamson (1980) reported higher concentrations of Cd, Pb and Zn in smaller snails and concluded that increase in body size reduced concentration of heavy metals. Savari (1991) showed that the increase of length of the shells reduced concentration of Ni in soft tissues of *Cerastoderma edule*.

Table 3: The comparison of results with the levels of regulation and safety tips

standard	Ni	Cd	Pb	Referenc
(EPA) ppm dry weight	80	4	1.7	EPA Guidana Document
FDA	80	3	1	Mortazavy, 2002
Length category 1	74.68	5.65*	18.65*	Present study
Length category 2	35.61	5.36*	9.92*	"
Length category 3	30.67	4.76*	9.19*	"

* The symbol indicated that data are higher than standard level.

In this study, shells with 2 and 3 cm in length were counted as one year old and immature oysters (Ashjaardalan, 2000). Therefore, class 1 is regarded as immature oysters, while classes 2 and 3 as mature oysters. Because immature or small bivalves had faster growth rate and higher metabolic rate than larger ones, they needed more food and filter feeding than large clams (Savari, 1991). This is not only caused pumping higher volumes of water per unit of their body mass, compared to adults, but also affected the rates of absorption and elimination of metals (Yap et al., 2009). Researchers have indicated that smaller bivalves had higher concentrations of Ni and Pb than bigger bivalves, due to the lack of factors like spawn that leads to sudden loss of heavy metal and body weight. According to the metabolic requirements of body and surface to volume ratio, bivalves with different sizes accumulated different amounts of metals (Hedouin et al., 2006). Reducing of metal concentrations with increasing body size suggested that a significant part of the metal content was surface adsorbed. Smaller bivalves had larger surface to volume ratio, therefore, metal concentrations in smaller bivalves were higher than in adults (Jones et al., 1992).

The results of study revealed that the size of oyster had no correlation with Cd bioaccumulation and Cd concentrations in oysters with different lengths which were almost constant. This is similar to what was reported by Saavedra (2003), that expressed Cd accumulation in bivalve *Mytilus galloprovincialis* which was independent of size. This might be because Cd in oyster tissues (unlike other metals) did not decrease in spawn period (Zarogian, 1980). During spawn period, mature oysters (classes 2 and 3) did not excrete Cd from their body. Because Cd was similar to essential metals, such as Zn and Ca, oysters could have mistaken them with other metals and did not excrete them (Fan, 2002).

Javanshir et al. (2009) expressed that absorption of heavy metals (Cd in this work) might change when concentration of dissolved calcium changes. Cd and Ca have very similar atomic radii (Yap et al., 2004), thus Cd might be absorbed through Ca channels (Banaoui et al., 2004). Oysters have calcareous shells and they require Ca for shell growing. Due to high concentration of Cd or low Ca concentration in the surrounding environment, oysters might use Cd instead of Ca. This might be the reason that why no significant differences found for Cd concentrations in mature and immature oysters.

Different results in the study of relationship between body size and accumulation of Cd in oysters have been reported by some researchers. For example, Pashaei Rad (2010) expressed that there is significant positive correlation between the concentration of Cd and the length of *Amiantis umbonella* oyster. They stated that bigger clams were older than smaller ones; therefore, they were exposed to heavy metals for longer periods and accumulated them in their body. Khristoforova (1990) demonstrated that due to a reduction in the ability of older clams in excreting Cd, cadmium concentration increased with age.

Metal concentrations can increase, decrease, or remain constant with body size (Boyden, 1974). For example, a wide range of results have been reported for Cd in mussels in different studies, and positive (Harris et al., 1979; Lobe1 & Wright., 1982), negative (Phillips, 1976; Cossa et al., 1980; Borchardt et al., 1988), or no (Phillips, 1976; Fischer, 1983; Popham & d'Auria., 1983) correlations were found between Cd concentration in mussels and body size.

A number of growth-related changes in physiology, gonad development, food preferences, and metallothionein lysosome detoxification systems cause variations in Cd accumulation trends. Variations in certain intrinsic and extrinsic properties, including species, ploidy, habitat, age and weight, season, source of Cd exposure, and metal detoxification systems could affect the results of studies (Rasmussen, 2007).

The comparison of obtained heavy metal concentrations are three categories of length in edible oyster with EPA and FDA standards, (Table. 3) indicated that concentration of Cd and Pb were higher than permissible level. Also the results showed that the concentration of these metals were high in small oysters (2-3 cm than oysters with a length greater than 3 cm). These oysters are amongst important sources of nutrition of native people, therefore high consumption of Loyesters, especially those with a length less than 4 cm, could introduce high concentration of Cd and Pb (per gram of consumption) to the body of consumer than those with a greater length.

Pb is absorbed by human body, via inhalation but, Cd enters the body mainly via foods such as liver, mushroom, river oyster and accumulates in kidney finally. High levels of Cd in body can cause diarrhea, abdominal pain and vomiting, broken bones, infertility and sterility, damage the central nervous system and immune system and cause mental disorders, DNA destruction, peptic ulcer and cancer.

In aquatic ecosystems, Cd accumulates more in river oyster, shrimps, crabs and fish. Different concentrations of Cd in human body, accumulate more in liver and kidney and cause high blood pressure, liver disease and brain and spinal disorders. Calcium in the bones could be replaced by cadmium, so it could cause softening of bones or brittle bones in humans (Abbe et al., 2000). Therefore, necessitating constant environmental monitoring in studied area.

References

Abbe, G. R., Riedel, G. F. and Sanders, J. G., 2000. Factor that influence the accumulation of copper and cadmium by transplanted eastern oyster (Crassostrea virginica) in the patuxent River, Mary land. Marine Environmental Research. 49(4): 377-396.

Amiard, J. C., Amiard- Triquet, C., Berthet, B. and Metayer, C., 1986. Contribution to the ecotoxicological study of cadmium, lead, copper and zinc in the mussel *Mytilus edulis* I: Field study. Marine Biology. 90(3): 425-431.

Ashjaardalan, A., 2000. Distribution and biology of growth and reproduction in oyster *Saccostrea cucullata* off the coast of Oman. Dissertation, Islamic Azad Aniversity, Science & Research Branch of Tehran, Iran, Pp. 220. (In Persian)

Banaoui, A., Chiffoleau, J. F., Moukrim, A., Burgeot, T., Kaaya, A., Auger, D. and Rozuel, R., 2004. Trace metal distribution in the mussel *Perna perna* along the Moroccan coast. Marine Pollution Bulletin. 48(3-4): 385-390.

Borchardt, T., Burchert, S., Hablizel, H., Karbe, L. and Zeitner, R., 1988. Trace metal concentrations in mussels: comparison between estuarine, coastal and offshore regions in southeastern North Sea from 1983 to 1986. Marine Ecology-Progress

Series. 42(1): 17-31.

Boyden, C. R., 1974. Trace element content and body size in molluscs. Nature. 251(5473): 311-314.

Butet, I., Tanguy, A. and Moraga, D., 2004. Response of the Pacific oyster *Crassostrea gigas* to hydrocarbon contamination under experimental conditions. Gene. 329: 147-154.

Canali, M., Ay, O. and Kalay, M., 1998. Levels of heavy matal (Cd, Pb, Cu, Cr and Ni) in tissue of *Cyprinus carpio*, *Barbus capito* and *Chondrostoma regium* from the Seyhan river. Turkish Journal of Zoology. 22: 149-157.

Clark, R., 2001. Marine Pollution (5th Ed.). Oxford University Press, USA, Pp.248.

Cossa, D., Bourget, E., Pouliot, D., Piuze, J. and Chanut, J. P., 1980. Geographical and seasonal variations in the relation between trace metals content and body weight in *Mytilus edulis*. Marine Biology. 58(1): 7-14.

Demora, S., Fowler, S. W., Wyse, E. and Azemard, S., 2004. Distribution of heavy metals in marine bivalves, fish and coastal sediments in the Persian Gulf and Gulf of Oman. Marine Pollution Bulletin.49(5-6): 410-424.

Einollahi Peer, F., Safahieh, A., Dadollahi Sohrab, A. and Pakzad Tochaii, S., 2010. Heavy metal concentrations in rock oyster *Soccostrea cucullata* from Iranian coasts of the Oman sea. Trakia Journal of Sciences. 8: 79-86.

Fan, W. H., Wang, W. X. and Chen, J. S., 2002. Geochemistry of Cd, Cr and Zn in highly contaminated sediments and its influences on assimilation by marine bivalves. Environmental Science and Technology. 36(23): 5164-5171.

Fischer, H., 1983. Shell weight as an independent variable in relation to cadmium content of molluscs. Marine Ecology-Progress Series. 12(1): 59-75.

Harris, J. E., Fabris, G. J., Statham, P. J. and Tawfik, F., 1979. Biogeochemistry of selected heavy metals in Western Port, Victoria, and use of invertebrates as indicators with emphasis on *Mytilus edulis planulatus*. Australian Journal of Marine and Freshwater Research.30(2): 159-178.

Hedouin, L., Metian, M., Teyssie, J. L., Fowler, S. W., Fichez, R. and Warnau, M., 2006. Allometric relationships in the bioconcentration of heavy metals by the edible clam (*Gafrarium tumidum*). Science of the Total Environment. 366(1): 154-163.

Javanshir, A., Shapoori, M., Azarbad, H., M.Vaghefi, A. R. and Danekar, A., 2009. Influence of calcium presence on the absorption of cadmium by the rock oyster *Saccostrea cucullata* from Persian Gulf (Ostreidae; Bivalvia) in laboratory conditions. Journal of Ecology and the Natural Environment. 1(8): 178-183.

Jones, H. D., Richards, O. G. and Southern, T. A., 1992. Gill dimension, water pumping rate and body size in the mussel *Mytilus edulis (L.)*. Journal of Experimental Marine Biology and Ecology. 155(2): 213-237.

Khristoforova, N. K. and Chernova, E. N., 1990. Trace element composition of giant oyster from Posyet Bay Sea of Japan. Soviet Journal of Marine Biology. 15(5): 340-346.

Lobel, P. B. and Wright, D. A., 1982. Relationship between body zinc concentration and allometric growth measurements in the mussel *Mytilus edulis*. Marine Biology. 66(2): 145-150.

Lobel, P. B., Bajdik, C. D., Jackson, S. E. and Longerich, H. P., 1991. Improved protocol for collecting mussel watch specimens taking into account sex, size, condition, shell, shape and chronological age. Archives of Environmental Contamination and Toxicology. 21(3): 409-414.

Martincic, D., Kwokal, Z., Peharec, Z., Margus, D. and Branica, D., 1992. Distribution of Zn, Pb, Cd and Cu between sea water and transplanted mussel (Mytilus edulis). Science of the Total Environment. 119: 211-230.

Naimo,T. J., 1995. A review of the effects of heavy metals on freshwater mussels. Ecotoxicology.

4(6): 341-362.

Pashaeirad, S., Saeedi, H., Abtahi, B. and Kiabi, B., 2010. Accumulation of some heavy metals in soft tissue and shell of edible bivalve Amiantis umbonella (Lamarck, 1818) in Bandar Abbas coast, the Persian Gulf. Journal of Animal Environment. 2(2): 9-22.

Phillips, D. J. H., 1976. The common mussel Mytilus edulis as an indicator of pollution by zinc, cadmium, lead and copper. l. Effects of environmental variables on uptake of metals. Marine Biology. 38(1): 59-69.

Phillips, D. J. H. and Rainbow, P. S., 1993. Biomonitoring of trace aquatic contaminants. (London: Elsevier sience publishers).

Popham, J. D. and Auria, J. M.D., 1983. Combined effect of body size, season and location on trace element levels in mussels (*Mytilus edulis*). Archives of Environmental Contamination and Toxicology. 12(1): 1-14.

Rasmussen, R. S., Morrissey, M. T. and Cheney, D., 2007. Effect of age and tissue weight on the cadmium concentration in Pacific oysters (*Crassostrea gigas*). Journal of Shellfish Research. 26(1): 173-179.

Rabinson, W. A., Maher, W. A., Krikoea, F., Nell, J. A. and Hand, R., 2005. The use of the oyster Saccostrea glomerata as a biomonitor of trace metal contamination: intra-sample, local scale and temporal variability and its implications for biomonitoring. Journal of Environmental Monitoring. 7(3): 208-223.

Riget, F., Johansen, P. and Asmund, G., 1996. Influence of length on element concentrations in the blue mussels (*Mytilus edulis*). Marine Pollution Bulletin. 32(10): 745-751.

Ruelas-Inzunza, J. R. and Paez-Osuna, F., 2000. Comparative bioavailability of trace metals using three filter-feeder organisms in a subtropical coastal environment (Southeast Gulf of California). Environmental Pollution. 107(3): 437-444.

Savari, A., Lockwood, A. P. M. and Sheader, A., 1991. Effects of season and size (age) on heavy metal concentrations of the common cockle (Cerastoderma edule (L.)) from Southampton Water. Journal of Molluscan Studies. 57(1): 45-57.

Saavedra Y., Gonza´lez A., Ferna´ndez P. and Blanco J: The effect of size on trace metal levels in raft cultivated mussels (*Mytilus galloprovincialis*). The Science of the Total Environment. 2003: 318 (2004): 115–124.

Tinsley, L. J., 1979. Chemical concepts in pollution behavior. New York, USA, Pp 57-59.

Usero, J., Morillo, J. and Gracia, I., 2005. Heavy metal concentrations in molluscs from the Atlantic coast of southern Spain. Chemosphere. 59(8): 1175-1181.

Williamson, P. D., 1980. Variables affecting body burdens of lead, Zinc and cadmium in a road side population of the snail *Cepaea hortensis* Muller. Oecologia (Ber.). 44(2): 213-220.

Yap, C. K., Ismail, A., Omar, H. and Tan, S. G., 2004. Toxicities and tolerances of Cd, Cu, Pb and Zn in a primary consumer (*Perna viridis*). Environment International. 29(8): 1097-1104.

Yap, C. K., Ismail, A., Edward, F. B., Tan, S. G. & Siraj, S. S., 2006. Use of different soft tissues of Perna viridis as biomonitors of bioavailability and contamination by heavy metals (Cd, Cu, Fe, Pb, Ni and Zn) in semi-enclosed intertidal water, the Johore Straits. Toxicological and Environmental Chemistry. 88(4): 683- 695.

Yap, C. K., Ismail, A. & Tan, S. G., 2009. Effect of body size on heavy metal contents and concentrations in green-lipped mussel *Perna viridis* (Linnaeus) from Malaysian Coastal Water. Pertanika Journal of Science & Technology. 17(1): 61-68.

Zarogian, G. E., 1980. *Crassostrea virginica* as indicator of cadmium pollution. Marine Biology. 58(1): 275-284.