J. Appl. Environ. Biol. Sci., 2(1)59-66, 2012

© 2012, TextRoad Publication

ISSN: 2090-4215
Journal of Applied Environmental and Biological Sciences
www.textroad.com

The Oyster *Saccostrea cucullata* as Biomonitor Agent for Some Metals (Cu, Pb and Ni) from Iranian Coasts of the Oman Sea

Einollahi Peer F[1]*, Safahieh A[2], Savari A[2], Dadollahi Sohrab A[3]

[1]*Institute Research of Hamun International Wetland Zabol university, Zabol, Iran
[2]Department of Biology, Faculty of Oceanography, Khoramshar University of Marin Sciences and Technology, Khoramshahr, Iran
[3]Department of Environment, Faculty of Nutral resources, Khoramshar University of Marin Sciences and Technology, Khoramshahr, Iran

ABSTRACT

This study was carried out with the purpose of using *Saccostrea cucullata* for biological monitoring three metals (Cu, Pb and Ni) on the Iranian coasts along the Oman Sea. Samples of oysters and sediments were collected from the intertidal zone at five different stations during low tide in August and February 2007. Sediment and tissue samples were oven dried and acid digested. The metal contents of samples were analyzed using an atomic absorption spectrophotometer (Unicam 919). The results showed that Cu, Pb and Ni concentrations in the sediment samples are as fallows: 27.4 ±1.1, 32.0 ± 2.2 and 20.8 ± 2.2 $\mu g.g^{-1}$dw respectively. The metals concentration in the soft tissues of oyster was determined as 185.2 ±21.0, 5.1 ± 1.1 and 6.0 ±1.0 $\mu g.g^{-1}$dw for Cu, Pb and Ni respectively. Similarly, Cu, Pb and Ni concentrations in the shells were found to be at 27.1 ± 1.6, 27.3 ± 2.6 and 14.2 ± 2.0 $\mu g.g^{-1}$dw respectively. A Significant correlation was found between Pb concentration in soft tissue and sediment samples. Similarly correlations between metals contents of sediments and hard tissues were found to be significant, suggesting that the soft tissue of *S. cucullata* should be a useful tool for Pb monitoring in the study area. While, the shell is an appropriate biomonitor agent for the all three metals. According to available standards, metals concentration in the soft tissue of oyster was higher than daily human' s consumption.

Key words: Heavy metals, *Saccostrea cucullata*, Biomonitoring, Intertidal zone

INTRODUCTION

Marine pollution is a global environmental problem. Human activities in the coastal area and marine water contribute to the discharge of various kinds of pollutants such as heavy metals into the marine ecosystems (Censi *et al.,* 2006; Pote *et al.,* 2008). The main reason for the metal contamination is considered as persistent and due to their toxic properties, could create several problems for different kinds of marine ecosystems and could be accumulating in marine organisms (Wen *et al.,* 2007; Wcislo *et al.,* 2008). Moreover, their accumulation in marine organisms and biomagnification throughout the food chain may be harmful for human health (Kowalski 1994; Valls and Lorenzo, 2002; Gochfeld 2003; Yi *et al.,* 2008).
Monitoring of heavy metals in marine environment as especially coastal zone is very important to assess the metal contamination in the marine environment (Fowler *et al.,* 2007; Yin *et al.,* 2008). Traditional monitoring of heavy metals in the aquatic environment involves determining and comparing the metal in water, sediment and biota. But, each method present it's own problems and limitations (Agrawal, 2005).

The low concentration of metals in the ambient water makes analysis difficult as contamination problems become significant and pre-concentration is required. The typical large temporal variations in metals concentration in water often warrant frequent sampling and analysis. Another major criticism is that information on bioavailability of metals is not provided (Mashinchian Moradi, 2001; Boening, 1999). The metals concentration in sediment provides a time-integrated estimate of metal levels. However, are significantly affected by particle size, organic content and redox conditions, which cannot be standardized (Tam and Wong, 2000; Santos *et al.,* 2005; Mil-Homens *et al.,* 2007; El Nemr *et al.,* 2007; Karsten *et al.,* 2008).

The use of organisms for biomonitoring of heavy metals in marine environment cannot only concentrate metals from water, but also provides a time-integrated estimate on the bioavailable fraction of heavy metals in marine ecosystems (Nicholson & Lam, 2005; Morelli *et al.,* 2009). As a result, biomonitoring process has been widely used to monitoring metals in the last two decades (Zelika *et al.,* 2003; Nicholson & Lam, 2005; Stanly *et al.,* 2008). Different types of organisms may be used for biomonitoring, such as marine algae (Topcuo *et al.,* 2003; Besada *et al.,* 2009) and filter-feeding mollusks (Mashinchian Moradi, 2001; Zorita *et al.,* 2006; Hamed and Emara, 2006). Many studied showed, bivalves do not regulate

***Correspondence Author:** Einollahi Peer F, Institute Research of Hamun International Wetland Zabol university, Zabol, Iran. Fateme.eynollahi@yahoo.com Fax: 05422251521, Mobile 0989155441732

the level of some metals within their body (Stanly *et al.,* 2008) and they can deflect the metals contamination from surrounding area. Thus, bivalves to be considered as good biomonitor agents for heavy metal monitoring in aquatic ecosystems (Elfwing & Tedengreen, 2002; Yap *et al.,* 2003; Zelika *et al.,* 2003; Nicholson *et al.,* 2005; Zorita *et al.,* 2006; Vlahogianni *et al.,* 2007; Maanan, 2008).

Chabahar is a developing area. Water discharge by boats and ships, marine transportation and ballast water discharges are main sources of pollutants in this area. While, food factory, wastewater, industrial and agricultural discharges and dredging are another sources of pollutant in Chabahar coasts. These activities along the Chabahar coasts have caused this area to be exposed to different kinds of pollutants specially heavy metals (Amini Ranjbar, 2006). Although bivalves specially *S. cucullata* are widely distributed in this area, but there is a lack of information related to heavy metal content of them. This study was carried out with purpose using of S. *cucullata* indices biological monitoring of heavy metals in the studied area.

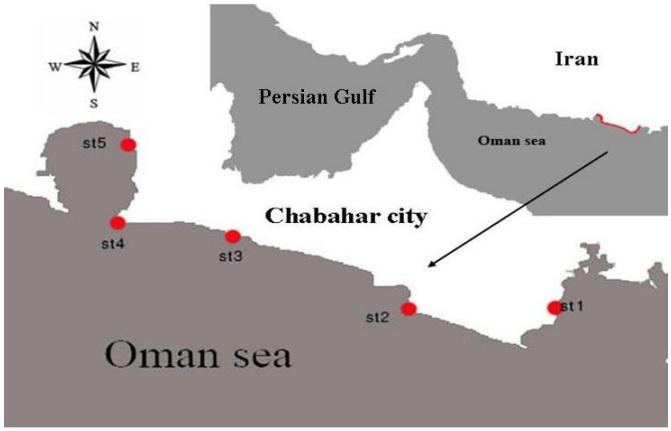

Fig. 1 Map showing study area

MATERIALS AND METHODS

Study area

Five different stations were studied along Chabahar coasts from Guatr bay to Tiss port (Fig.1). The position and the name of each station are shown in table 1.

Sampling and storage

Samples of sediment and oyster were taken during low tide in August and February 2007. The top 3-5 cm of sediments were collected near the oyster habitats. Three replicates of sediments were sampled from each site. Each sediment sample was placed in an acid-washed polyethylene bag and deep-frozen prior to analysis. Thirty oysters of the same size (7.65 to 9.1 cm) were collected from each station. Stainless steel hammer and Rod were used to separate oysters from their surrounding cliffs. The collected oysters were placed in polyethylene containers and all samples transferred to laboratory by using Icebox. All of debris's were removed from each sample in laboratory. After washing shells with double distilled water, oysters were freezed in -20^{o}c freezer until the next step (Orescanin *et al.,* 2006).

Sample preparation

In order to dry samples, Sediments were oven dried at 105 ^{o}C for at least 16 h until a constant weight was obtained (Delman *et al.,* 2006). Afterwards, they were sieved through a 0.6 mm stainless steel sieve and shaken vigorously to produce homogeneity. Then they were powdered using glass mortar and stored in polyethylene pillboxes until digestion. Oysters were taken from freezer and were placed in the laboratory to melt their ice. Soft tissues were separated from the shell by using stainless steel knife and both soft and hard tissues were oven dried at 80^{o}C until constant weight was obtained (Yap *et al.,* 2002; Orescanin *et al.,* 2006). The dried samples from each station were then pooled together in order to obtain sufficient amount of tissues for metal analysis. They were powdered using glass mortar and were stored in polyethylene pillboxes until digestion. They oyster shells were also washed using percentage0.5 nitric acid and oven dried. Then they were powdered and stored as the same procedure as for soft tissues (Yap *et al.,* 2003).

Heavy metals analysis

for the analysis of total Cu, Pb and Ni concentrations in sediment, 1g of each dried sample was digested in a combination of concentrated nitric acid (65%, Merck, Darmstadt, Germany) and perchloric acid (60% Merck) in the ratio 4:1, first at low temperature (40^0C) for 1 h and then the temperature was increased to 140^0C for 3 h (Orescanin *et al.,* 3006). The metal analysis for both tissues was performed with the same method. 1 g of each dry sample from soft tissue and shell of oyster were digested in pure nitric acid (65% Merck). The samples were predigested first for 1 hour in 40^0C and then digestion was continued for 3 hours in 140^0C. After digestion, samples were cooled in laboratory temperature, diluted to certain volume using double distilled water and filtered by filter paper (Whatman 42µ) (Yap *et al.,* 2002). Heavy metals analysis was performed by using Unicom 919 an air-acetylene atomic absorption spectrophotometer.

Data analysis

All data were analyzed statistically by using SPSS version 13. The data were tested for normal distribution first. After ensuring normal distribution of data, the One-way analysis of Variance (ANOVA) was used to find any significant difference between metals concentration in samples. If significant difference was observed, Tukey post hoc test was used to determine different kinds. The Pearson's correlation coefficient was applied to determine the relationship and the significant levels between any two variables.

Table 1. Position of stations along Chabahar coasts

station		latitudes	
St1	Guatr	*E 61 ° 30 ' 9/3 "*	*N 25 ° 9 ' 53/1 "*
St2	Beris	*E 61 ° 10.3 ' 6/5 "*	*N 25 ° 8 ' 55/7 "*
St3	Ramin	*E 60 ° 44 ' 50 "*	*N 25 ° 16 ' 9/3 "*
St4	Shahid Beheshti	*E 60 ° 37 ' 12/7 "*	*N 25 ° 21 ' 92/9 "*
St5	Tiss	*E 60 ° 37 ' 21/5 "*	*N 25 ° 17 ' 39/2 "*

Table 2. The mean of Cu concentration (µg.g^{-1} dry weight) in sediment and different tissues of *S. cucullata* in summer and winter

	sediment		Soft tissue		shell	
station	summer	winter	summer	winter	summer	winter
Guatr	38.47±10	16.22±1.88	201.31±27.83	289.93±93	39.88±0.89	14.92±1.90
Beriss	38.69±0.62	16.96±2.26	183+54±35.9	102.46±9.14	39.96±0.35	13.83±1.12
Ramin	37.88±0.17	16.90±2.20	195.96±6.40	371.16±44.05	40.04±0.88	16.67±1.27
Beheshti	38.07±0.25	15.87±1.25	89.70±7.67	84.96±17.23	37.15±5.46	14.16±1.57
Tiss	38.29±0.36	16.25±1.12	137.52±10.73	195.30±19.65	40.57±0.71	15.88±1.55

Table 3. The mean of Pb concentration (µg.g^{-1} dry weight) in sediment and different tissues of *S. cucullata* in summer and winter

	sediment		Soft tissue		shell	
station	summer	winter	summer	winter	summer	winter
Guatr	50.25±1.53	13.33±1.29	5.36±1.37	3.83±1.71	43.71±2.71	11.30±1.35
Beriss	44.93±2.65	15.79±1.52	5.37±0.77	4.26±1.69	38.75±3.04	14.37±1.76
Ramin	56.45±4.06	15.61±1.90	5.91±0.56	5.26±1.13	46.11±4.27	14.92±1.36
Beheshti	51.78±3.27	12.09±0.35	5.15±0.60	4.70±1.00	44.68±3.27	12.79±2.21
Tiss	48.48±4.60	10.98±1.14	5.97±0.54	4.71±0.96	41.76±4.01	12.23±0.85

Table 4. The mean of Ni concentration (µg.g^{-1} dry weight) in sediment and different tissues of *S. cucullata* in summer and winter

	sediment		Soft tissue		shell	
station	**summer**	**winter**	**summer**	**winter**	**summer**	**winter**
Guatr	25.69±2.22	12.38±1.86	9.39±1.79	6.40±0.62	20.91±2.98	**5.47±1.28**
Beriss	30.28±2.40	17.10±1.78	5.31±1.05	7.19±0.76	20.24±2.66	**7.79±0.48**
Ramin	28.42±2.54	14.46±2.33	1.89±1.09	6.94±0.71	24.41±3.53	**7.79±0.71**
Beheshti	26.41±5.42	13.55±0.01	6.36±0.91	5.67±0.69	21.55±3.71	**6.74±1.24**
Tiss	**23.74±1.06**	**15.91±2.71**	**4.36±1.86**	**6.05±0.56**	**22.36±3.00**	**6.09±0.85**

Table 5. The Correlation between heavy metal concentrations in sediment and different tissues of *S. cucullata* in Iranian coast of the Oman sea

		Cu	Pb	Ni
Soft tissue	**sediment**	R^2=0.08 A=44.15 B=-152.9	R^2=0.61* A=0.02 B=4.18	**R^2=0.04 A=0.05 B=4.74**
shell	**sediment**	**R^2=0.99* A=1.11 B=-3.31**	**R^2=0.99* A=0.80 B=2.27**	**R^2=0.89* A=1.13 B=-9.35**

*Levels of significance are indicated as P < 0.05.

Table 6- A comparison of mean heavy metals concentration (µg/g wet weight) in the soft tissue of oyster from intertidal zone of the Chabahar coasts

Standard	Cu	Pb	Ni	reference
WHO	10	---	0.2	Shulkin et al. 2003
FAO	30	0.5	0.5	Shulkin et al. 2003
USFDA	11.5	1.70	0.8	Liu and Kueh, 2005
ICES	---	1.2	---	Chang et al. 2007
ISIRI	20	---	1	Darvish 2007
Soft tissue	27.4	0.73	0.87	Present study

RESULTS

The concentration of Cu, Pb and Ni in sediment and different tissues of oyster *S. cucullata* in different season are given in tables 2-4 respectively. Cu, Pb and Ni concentrations in sediment from five different stations were measured 15.87 to 38.69 µg.g^{-1}, 10.98 to 56.45 µg.g^{-1} and 12.38 to 30.28 µg.g^{-1} respectively. This indicated, the order of metal concentrations in the sediment of abundance: Pb>Cu>Ni. Similarly, the metal concentrations ranged in the shell were obtained 13.83 to 46.04 µg.g^{-1}, 11.30 to 40.11 µg.g^{-1} and 5.47 to 24.41 µg.g^{-1} for Cu, Pb and Ni respectively. However, this order of metal accumulation was different from that found in the soft tissue. Which had the following order of abundance: Cu>Pb>Ni. 84.70 to 289.93 µg.g^{-1}, 3.83 to 5.97 µg.g^{-1} and 1.89 to 9.39 µg.g^{-1} were measured for Cu, Pb and Ni in soft tissue of oyster in different stations during summer and winter. Pb concentration in shell was found to be higher than soft tissue. Unlike Pb, Cu concentration was found to be higher in the soft tissues.

Results indicated the pattern of metals accumulation in the sediment and shell of oyster were more and less similar (Pb>Cu>Ni). While, the pattern of metals accumulation in soft tissue was Cu>Pb≥Ni. Cu concentration was found to be higher in the soft tissues. Unlike Cu, the concentration of Pb in the shell was about 3 times higher than the soft tissues.

The assessment for the potential use of the total soft tissue and shell of *S. cucullata* as a biomonitoring tool for Cu, Pb and Ni was based on the following. The relationship between metals concentration in the different tissues of oyster and those in the sediment are presented in table 5. No significant correlation was found for Cu and Ni between the sediment and the total soft tissue of oyster (P>0.05). While, the correlation between Pb concentration in the soft tissue of *S. cucullata* and sediment was to be significant (R^2 = 0.61*, P<0.05). A significant correlation was found for Cu, Pb and Ni concentrations between the shell of oyster and the sediment (P<0.05).

DISCUSSION

They heavy metals concentration in sediment and different tissues of oyster were measured in intertidal zone of Chabahar coasts along the north coasts of the Oman sea. Results showed the order of metals accumulation in the sediment and shell of oyster are similar to Pb>Cu>Ni. While, the order of metal accumulation in the soft tissue of oyster is like to Cu>Pb≥Ni. These patterns indicated, the heavy metals concentration in the soft tissue of oyster may be regulate by physiological process and Excess concentration of heavy metals were accumulate in the shell, to have less effect on the oyster (Ballan-Dufrancias *et al.,* 2001). Thus, the metals accumulation in the sediment and shell are similar. While, this pattern is not like to order metals accumulation in the soft tissue of oyster. Cravo *et al.,* (2004) suggested that mollusk species might use their shell for sequestration of a part of up-taken heavy metals. This could be a part for detoxification of non-essential and excess essential metals taken in mollusks. On the other hand, the levels of Pb in the shell of Rock oyster from Chabahar coasts was found 3-4 times higher than those measured in the soft tissue of oyster and The soft tissue of oyster has high concentration of the Cu. Cu is an essential metal for oysters. They are use the Cu to make haemocyanin for respiratory pigments (Launestein *et al.,* 2002; Caussy *et al.,* 2003؛ Conner & Launestein, 2005). While, Pb is not essential element for oysters (Boening, 1999). Meanwhile, The shell matrix has a higher capacity for incorporation of these metals than the soft tissue (Ballan-Dufrancias *et al.,* 2001; Lauenstein *et al.,* 2002). Gillikin *et al.,* (2005) were compared the concentration of heavy metals between shell and soft tissue of *Mercenaria mercenaria* in Carolina. They were found, heavy metals concentration in the shell has not changed during 1949 to 2002. They were suggested a little change in metals concentration in the shell may be lack of effect of physiological processes.

In addition, the increase of Pb in the shell of Oyster may be due to crystalline structure of the shell. The Pb ion is then to several times tendency to bind with carbonate ions in the calcareous structure (Babukutty & Chako, 1995). Thus, the high concentration of Pb in the shell of oyster may be due to replace of Pb ions in the calcareous structure of the shell.

Biomonitors organisms accumulate the heavy metals from ambient bioavailable sources of the trace metal over a period (Rainbow, 2006). Thus, the accumulated heavy metals in difference tissue of biomonitor organism are a measure of the total integrated bioavailability of those metals to that organism at studied area along the previous period (Saed, 2001). Some mollusks including oysters are used for biomonitoring programs in marine ecosystems. The oyster *S. cucullata* like other mollusks is suspension feeder and can uptake they heavy metals from suspended sediment (Coles *et al.,* 1997; Caihuan & Wang, 2001). This oyster has a wide distribution in the Chabahar coasts and may be a suitable bioindicator organism for heavy metals in this area. The correlation between Cu, Pb and Ni concentration in the shell and total shell of oyster *S. cucullata* and those in ambient sediment was studied. The stronger correlation coefficients were found between heavy metals concentration in the sediment and total shell. No significant relationship was found between metals concentration except Pb in the soft tissue and ambient sediments. The biological control of heavy metals in the soft tissue puts limitation for use of soft tissue of oyster for biomonitoring of heavy metals. Thus, the soft tissue of oyster is more suitable as an indicator of Pb contamination in this area and the shell of *S. cucullata* should be a useful tool for monitoring of all studied metals in the study area.

Shulkin *et al.,* (2003) were found a significant correlation between Cu, Zn, Pb, Cd and Ni in the soft tissues of both mussel (*Crenomytilus grayanus*),oyster (*Crassostrea gigas*) and in their ambient sediment. They were suggested these mollusks can be use mainly for the monitoring of low and moderate contaminated in Japanian coasts.

Yap *et al.,* (2002) were studied the correlation between Cd, Cu, Pb and Zn concentration in the soft tissue of *P. veridis* and surface sediment in Malasia coasts. They were found a significant correlation between Pb and Ni in soft tissue and their ambient sediment. But, no significant correlation was found for Cu and Ni in this area. In according these results they are concluded, the soft tissue of mussel could be a useful tool for Cd and Pb in Malaysia coasts. A significant correlation was found for Cu, Zn, Pb, Cd and Ni in soft tissue of mussel (*Crenomytilus grayanus*), oyster (*Crassostrea gigas*) and ambient sediment in northern coast of the Japan. So the soft tissue of these mollusks was found to be a good tool for these metal biomonitoring in this studied area (Shulkin *et al.,* 2003). Szefer *et al*., (2002) were found a significant relationship between Hg, Cd, Pb, Ag, Cu, Pb, Cr, Co, Mg and Fe in the soft tissue and byssus of *Metilus edulis* and surface sediment in Polish coasts in Baltic Sea. They were suggested, the soft tissue and byssus of *M. edulis* could be suitable for these metals biomonitoring in this area.

Some heavy metals are necessary for human health. Nevertheless, the concentration exceeding levels considered harmful to human consumption. This oyster is edible and human can be eating this oyster. Meanwhile, Heavy metals concentration in the soft tissue of oyster were compared with WHO (World Health Organization), FAO (Food and Agriculture Organization), ICES (International Council for the Exploration of the Sea), USFAD (the Food and Drug Administration) and ISIRI (The Institute of Standards & Industrial Research of Iran) standards for daily consumption (table 6). According to these results, The Cu concentration was higher than WHO, USFDA and IRISI standards. Ni concentration was higher than the FAO, WHO and USFDA. However, Pb was higher only than the FAO standard. Consequently, these metals concentration are higher than daily consumption.

Conclusion

This study was showed the pattern of metals accumulation in the soft and hard tissue of *S. cucullata* is different. The difference pattern of metals accumulation in the soft tissue could be related to the biological role of that metal in the body. The stronger correlation coefficients were obtained for metals concentration between sediment and shell of oyster. The significant correlation and the lower degrees of heavy metals variability in the total shells of *S. cucullata* suggested, it to be generally a more sensitive and precise biomonitoring material for heavy metals than the soft tissues of *S. cucullata*. Soft tissue of *S. cucullata* for Pb and shell of this oyster was useful tool for biomonitoring of total metals studied Iranian coasts of the sea of Oman. According to some available standards, the metals concentration in the soft tissue of oyster was higher than daily consumption for human.

Acknowledgments: They authors are would like to thanks from the Iranian National Institute of Oceanography (Chabahar station, Iran) and the Department of Environment in the Sistan and Baluchestan province for their assistance.

REFERENCES

Agrawal SK (2005) Environmental monitoring. publishing corporation. (5-12PP) New Delhi – 110002.

Amini-Ranjbar Gh and Miraki Gh (2006) Evaluation of quantity and quality of the Chabahar bay. Sistan And Baluchestan University, Y.pp: 7-9.

Ballan-Dufrancais C, Jeantet AY, Geffard A, Amiard JC and Amiard-Triquet C (2001) Cellular and tissular distribution of Copper in an intrasedimentary bivalve, the Baltic clam *Macoma Balthica*, originating from a clean or a metal-rich site. *Aquat Sci.* 58,1964-1974.

Babukutty Y and Chacko J (1995) Chemical partitioning and bioavailability of Lead and Nickel in an estuarine system. *Environ Toxic Chem.* 14,427-434.

Besada V, Andrade JM, Schultze F and Gonzalez J (2009) Heavy metals in edible seaweeds commericialised for human consumption. *J of Marine Syst.*75, no (1-2):305-313.

Boening W (1999) An evaluation of biomonitors of heavy metals pollution in marine waters. *Environ Mon and Assess* 55,459-470.

Chang F, Li, G, Haws M and Niu T (2007) Element concentrations in shell of *pinctata margaritifera* from French Polynesia and evaluation for using as a food supplement. *Food Chem.* 104,1171-1176.

Caihuan K and Wang W (2001) Bioaccumulation of Cd, Se, and Zn in an estuarine oyster *Crassostrea rivularis* and a coastal oyster *Saccostrea glomerata*. *Aqua Toxic.* 56,33–51.

Caussy D, Gurzau DM, Neagu C and Ruedel H (2003) Ecotoxicology and lessons from case studies of metals: investigating exposure, bioavailability and risk. *Environ Saf,* 56,45-51.

Censi P, Spoto SE, Saino FM, Sprovieri S, Mazzola A and Nardone G (2006) Heavy metals in coastal water systems: Acase study from the northewestern Gulf of Tailand. *Chem*, 64,1167-1176.

Coles SL, RC Defelice, RC, Eldidge LC and Carlton JT (1997) Biodiversity of marine communities in Pearl Harbors, Ohau, Hawaii with observation on intropuced exotic spicies. (111p). Hawaii Biological Survey Bioshep Musem Technice Report Nomber10.

Connor TP and Lauestein. GG (2005) Status and trends of copper concentrations in mussels and oysters. *Mar Chem*. 97,49-59.

Cravo A, Foster P and Bebianno. MJ (2004) Minor and trace elements in the shell of *Patella aspera*. *Environ Inter.* 28,295-302.

Darvish A (2007) The survey of heavy metals (Cd, Cu, Ni and Hg) in water and soft tissue of *Saccostrea cucullata* in intertidal zone of Hormoz islans, Persian Gulf, M. S. thesis, Environmental Engineering, Azad Univ., of Ahvaz, Iran.

Delman O, Demirak A and Balci 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 Chem.* 26,157-162.

Elfwing T and Tedengreen M (2002) The effects of Copper on the metabolism of three species of tropical oysters, *Saccostrea cucullata, Crassostrea lugubris* and *Crassostrea belcheri*. *Aquacul.* 204,157-166.

EL Nemr A, Sikaily EL and A. Khaled (2007) Total and leachable heavy metals in muddy and sandy sediments of Egyptian coasts along Mediterranean sea, Estuar. *Coast Shelf Sci.* 192,151-168.

Fowler SW, Villeneuve JP, Wyse, E., Jupp B and De Mora S (2007) Temporal survey of petroleum hydrocarbons, organochlorinated compounds and heavy metals in benthic marine organisms from Dhofar, southern Oman, *Mar Pollut BulleT.* 54,339–357.

Gillikin DP, F Dehairs, W Baeyens, J Naves, A Lorrain and L Andre (2005) Inter and intra annual variations of Pb/Ca ratios in clam shells (*Mercenaria mercenaria*): a record of anthropogenic lead pollution? *Mar Pollut Bullet.* 50,1530-1450.

Gochfeld, M (2003) Cases of mercury exposure, bioavailability and absorption. *Ecotoxic and Environ Safety*. 56,174-179.

Hamed AH and AM Emara (2006) Marine molluscs as biomonitors for heavy metal levels in the Gulf of Suez, Red Sea. *J of Mar Sys*. 60,220–234.

Karsten J, E Gjengedal and HJ Mobbsa(2008) Trace element exposure in the environment from MSW landfill leachate sediments measured by a sequential extraction technique. *J of Hazard Mater.* 153,751–758.

Kowalski Z (1994) Treatment of Cchromic tannery wastes, *J of Hazard Mater* . 37,137-144.

Lauenstein GG, AY Cantillo and TP Connor (2002) The status and trends of trace element and organic contaminants in oysters, *Crassostrea irginica,* in the waters of the Carolinas, USA., *The Sci of the Tot Environ*. 285,79-87.

Liu JH and CSW Kueh (2005) Biomonitoring of heavy metals and trace organics using the intertidal mussel *Perna viridis* in hong kong coastal waters. *Mar Pollut Bullet.* 51,857-875.

Maanan M (2008) Heavy metal concentrations in marine mollusks from the Moroccan coastal region. *Environ Pollut* 153,176-183.

Mashinchian Moradi A (2001) The kinetics of uptake and release of polycyclic aromatic hydrocarbons in the green mussels *Perna viridis* for biomonitoring of marine pollution. P,31-42, Univ., Putra Maly.

Mil-Homens M, RL Stevens, I Cato and F Abrantes (2007) Regional geochemical baselines for Portuguese shelf sediments. *Environ Pollut*. 148,418–427.

Morelli E, ML Marangi and L Fantozzi (2009) A phytochelatin-based bioassay in marine diatoms useful for the assessment of bioavailability of heavy metals released by polluted sediments, *Environ Intern.* 35,532–538.

Nicholson S and PKS Lam (2005) Pollution monitoring in southeast Asia using biomarkers in the mytilid mussel *Perna viridis* (*Mytilidae*: bivalvia). *Environ Intern.* 31,121-132.

Orescanin V, I Lovrencic, L Mikelic, D Barisic, Z Matasin, S Lulic and D Pezelj (2006) Biomonitoring of heavy metal and arsenic on the east coast of the middle Adriatic Sea using *Mytilus galloprovinciallis. Nucleus Instrum and Meth Res.* 86,495-500.

Pote J, Haller J Loizeau, AG Bravo, V Sastre and W Wildi (2008) Effects of a sewage treatment plant outlet pipe extension on the distribution of contaminants in the sediments of the Bay of Vidy, Lake Geneva, Switzerland. *Biores Techno.* 99,7122–7131.

Rainbow PS (2006) Biomonitoring of trace metals in estuarine and marine environments. *Aust J of Ecotoxic.* 12:107–122.

Saed K (2001) Ecotoxicology of heavy metals (Cd, Pb, Zn and Cu) in flat tree oyster *Isognomon alatus* from Sepang River Malaysia. Ph. D thesis. Univ., Putra Malay.

Santos IR, EV Silva, CEGR Schaefer, MR Albuquerque and LS Campos (2005) Heavy metal contamination in coastal sediments and soils near the Brazilian Antarctic station, King George Island. *Mar Pollut Bullet.* 50,185–194.

Shulkin VM, BJ Presley and VI Kavun (2003) Metal concentrations in mussel *Crenomytilus grayanus* and oyster *Crassostrea gigas* in relation to contamination of ambient sediments. *Environ Intern.* 29,493-502.

Stanly YM, KS Lam, M Martin and W Calwell (2008) The use of selected genotoxicity assays in green mussels *Perna viridid* a validation study in Hong kong coastal water. *Mar Pollut Bullet.* 83:1521-1529.

Szefer P, K Frelek, K Szefer, CB Lee, BS Kim and J Warzocha (2002) Distribution and relationships of trace metals in soft tissue, byssus and shells of *Mytilus edulis* trossulus from the southern Baltic. *Environ Pollut.* 120,423–444.

Tam NFY. and YS Wong (2000) Spatial variation of heavy metals in surface sediments of Hong Kong mangrove swamps, *Environ Pollut.* 110,195-205.

Topcuo KC, GN Balkis and C Kirbaolu (2003) Heavy metal monitoring algae from the Turkish coast of the Black sea, 1998-2000. *Chem*. 52, no. 10:1683-1688.

Valls M, VD and Lorenzo (2002) Exploiting the genetic and biochemical capacities of bacteria for the remediation of heavy metal pollution. *Fems Microbiol.* 26,327-338.

Vlahogianni M, M Dassenakis, MJ Scoullos and A Valavanidis (2007) Integrated use of biomarkers (superoxide dismutase, catalase and lipid peroxidation) in mussels *Mytilus galloprovincialis* for assessing heavy metals' pollution in coastal areas from the Saronikos Gulf of Greece, *Mar Pollut Bullet.* 54, no. 9:1361-1371.

Wcislo E, D Ioven, R Kucharski and J Szdzuj (2008) Human Health Risk Assessment Case Study: An Abandoned Metal Smelter Site in Poland, Chemosphere. *J of Geochem Explor.* 96,223–230.

Wen C, CM Kao, CF Chen CD Dong (2007) Distribution and accumulation of heavy metals in sediments of Caohsiung harbour, Taiwan. *Chem*. 66,1431-1440.

YapCK, A Ismail, SG Tan and H Omar (2002) Correlations between speciation of Cd, Cu, Pb and Zn in sediment and their concentrations in total soft tissue of green-lipped mussel *Perna viridis* from the west coast of Peninsular Malaysia. *Environ Intern*. 28,117–128.

Yap CK, A Ismail, SG Tan and A Rahim (2003) Can the shell of the green-lipped mussel *Perna viridis* from the west coast of Peninsular Malaysia be a potential biomonitoring material for Cd, Pb and Zn? *Estuarine, Coast and Shell Sci.* 57,623-630.

YiY, Z Wang, K Zhang, G YU and X Duan (2008) Sediment pollution and it's effect on fish through food chain in the Yangtze river, *Intern J of Sed Res.* 23,338-347.

Yin X, L Xia, L Sun, H Luo and H Wang (2008) Animal excrement: A potential biomonitor of heavy metals contamination in the marine environment. *Science of The Total Environmnet* 399, no.1-3:179-185.

Zelika B, S. Lukic and DPezelj (2003) Biomonitoring of heavy metal and arsenic on the east coast of the middle Adriatic Sea using *Mytilus galloprivinsialos. Mar Pollut Bullet.* 56,176-183.

Zorita I, M Zarragoitia, M Soto and P Gajaraville (2006) Biomarkers in mussels from a copper site gradient (Visnes, Norway) an integrated biochemical, histochemical and histological study. *Aquat Toxic.* 78,109-116.