INT. J. BIOL. BIOTECH., 15 (2): 269-275, 2018.

SOME BIOMONITORING STUDIES OF HEAVY METALS IN COMMERCIAL SPECIES OF CRUSTACEAN ALONG KARACHI COAST, PAKISTAN

Noor Us Saher* and Nayab Kanwal

Centre of Excellence in Marine Biology, University of Karachi, Karachi, Pakistan
noorusaher@yahoo.com; nayabkanwal01@gmail.com

ABSTRACT

The present study was conducted to estimate and compare the concentration of (Zn, Fe, Co, Cu, Pb, Cd, Ni and Cr) in the edible muscles of commercially important species of male and female crustaceans (*Portunus pelagicus, Portunus sanguinolentus, Charybdis feriata* and *Scylla serrata*). It also evaluated the bioaccumulation progression of the various elements based on Metal Pollution Index (MPI). It's an attempt to use these organisms as biomonitor species of heavy metals pollution, ensure for the seafood safety. The obtained result showed significant intersexual variations of Zn, Fe, Co, Cu, Pb, Cd, Ni and Cr levels in the edible muscles of each investigated species of crabs. The procured data also, be evidence for the higher MPI factor of essential metals than the toxic and borderline heavy metals in all studied crab species. Moreover, MPI values suggested that *P. pelagicus* and *S. serrata* have a greater competence for metal bioaccumulation than *C. feriata* and *P. sanguinolentus.* The *P. pelagicus* and *S. serrata* are more vulnerable to metal pollution than the other studied species. Therefore, it is suggested that *P. pelagicus* and *S. serrata* can be used as biomonitoring species of metal pollution.

Keywords: Biomonitor, Crabs, Essential metals, Non-essential metals, Metal pollution index.

INTRODUCTION

The biomonitoring studies (National and regional level) provide a useful tool for the assessment of the current status of environmental health and indirectly evaluation of coastal water quality and changes in the environment as exposed due to anthropogenic causes. In addition, the biomonitoring play a conspicuous role in the making and implication of protective measures, for the management and sustainability of specific and vulnerable marine ecosystems as for example an estuarine area of the coast and as well as transitional waters that likely influenced by various anthropic activities (Spada *et al*., 2013). An enormous number of marine organism bioindicators "sentinels" includes in monitoring programs and utilize to detect temporal and spatial variation of chemical pollutants (heavy metal and pesticides) which contribute to the assessment and knowledge of trends in marine contamination (Carro *et al.*, 2004).

The term "heavy metal" is used to explain the trace metals includes both essential and nonessential and are of particular concern around the world because they are environmentally persistent, undergo biogeochemical recycling, and pose ecological risks (Olmedo *et al*., 2013; Gu *et al*., 2015; Liu *et al*., 2015). All aquatic organisms need very low quantities of these metals, but the harmful effects are imposed upon the ecosystem where these metals exceed certain threshold levels (Maanan, 2008).

Heavy metals are characterized as being potentially toxic (As, Cd, Hg, Pb), probably nonessential or borderline (Ni, Cr), and essential (Fe, Co, Cu, and Zn) (Tuzen, 2009; Hedberg *et al*., 2014). Toxic metals can be detrimental if ingested continuously even in low concentrations. Essential heavy metals can also imprinted toxic effects if intake in excess (Tuzen, 2009; Gu *et al*., 2015; Gu *et al*. 2016). The bioaccumulation tendency of metals make them dangerous and in marine organism, the level of bioaccumulation depends on the total quantity and as well as their bioavailability in the environmental medium and the path of uptake, storage and through excretion mechanisms. Chemical compounds such as metals and pesticides accumulate in aquatic organism at any time they are taken up and accumulate faster than excretion from the body or tissues (Chiarelli and Roccheri, 2014).

The term bioindicator (the organisms or group of organisms) used to predict the current situation of a particular system through their interactions. The bioindicator species require a certain specific set of environmental variables for their growth and survival and changes in such specific variables directly or indirectly affect the number, reproduction and behavior of the species that indicated the responses of the species including that changes in the variables are non-suitable for such species.

The bioindicator organism has to fulfill some basic criteria that mainly include easiness in sampling, temporal and spatial abundance of the organism and the range in which the biological responses can be detected. There are various crustacean species, usually consider as bioindicator for the estimation of toxicological impacts. Some preliminary parameters taken into consideration include percent survival, population growth rate, number of larvae released, number of mature adults and survival numbers of progeny. The brachyuran crabs are high assorted group comprising about 700 genera and approximately 5000 species amongst crustacean fauna and these species are distributed in various habitats and play an important role to maintain the ecosystem. Therefore, any sticky variable that affects the crabs can have a conspicuous effect due to their habitat and the surrounding ecosystems. Few studies have been done to assess the physical condition of the particular ecosystem affected by diverse kinds of pollution using brachyuran crabs as an indicator species (Arya *et al*., 2014).

The toxicological impact differs according to taxa, probably even at the specific level (Zhou *et al.*, 2008). Thus, these metals should be defined with reference to their chemical criteria, typically in terms of their properties as Lewis acids and bases (Kucuksezgin *et al*., 2006). Therefore, these heavy metals as described the normal constituents of the marine environment (Nieboer and Richardon, 1980; Ali *et al*., 2014). The main rationale of monitoring the heavy metals concentration in shellfish is to determine the toxicological risk faced by the allied marine organisms, it is essential to detect the bioindicators that can be effected efficiently and specify the health state of the marine and accurate markers of pollution. For the assessment of the concentration of chemical pollutants and current status of marine ecosystems, the different species of Portunid crabs used as bioindicator organisms. In addition the evaluation of possible risks on human health and even by humans through the ingestion of contaminated edible species also estimated in this study.

MATERIALS AND METHODS

Study site

Pakistan has a coastline of 990 km, Karachi is situated on the southwestern part of Indus delta and northern border of the Arabian Sea. The Karachi has 30 km of coast, receiving a load of heavy metal from the industries and commercial hubs adjacent to the coastal areas (Monawwar *et al*., 1999). It is the densely populated city having more than 30 million population, so 60% of discharge from industries approximately 292 million gallons per day of untreated effluents through Malir and Lyari rivers is mixed into the coastal waters (Beg *et al*., 1975).

Karachi is a big city and the center of industrial activities containing more than seven thousand industries at four different sites, Landhi Industrial Trading Estate (LITE), Sindh Industrial Trading Estate (SITE), the West Wharf Industries (WWI) and Korangi Industrial Area (Monawwar et al., 1999). These industries produced steel, chemicals, oil, paints, pharmaceutical, food and fabric products (Zehra *et al*., 1996; Naqvi *et al*., 1993; Quraishee 1975) and these industries discharge their hazardous waste either directly or indirectly into the coast of Karachi (Ali *et al*., 2014).

Sampling

A total 60 fresh shellfish samples consist of edible Portunid crabs *Scylla serrata* (mud crab), *Portunus reticulatus* (Blue crab), *Portunus sanguinolentus* (Three spotted crab) and *Chaybdis feriatus* (Orange crab) were purchased in Karachi city from the fish harbor of Pakistan. The four crab’s species, each consisting of 15 samples, were normally collected from the Karachi fish harbor March to June 2011. All the crab species were commercially valuable and sale in markets and were cultured in local coastal areas in Karachi, Pakistan. After collection, all the samples were directly transported to the laboratory and freeze until analysis. In the laboratory, crabs were sorted, identified and sexed. The specimen was then blotted dry and its body weight was measured using an analytical balance, then soft tissues from shell were separate out with a sterilized scalper. Then the tissues were oven dried at 70 Cº for 24 hours and ground into fine powder for heavy metals measurement.

Elemental analysis

The heavy metals measurement was performed by using acid digestion. The 2 g of dry sample was digested in a mixture of concentrated Perchloric acid and nitric acid. The analytical detection was performed using an Atomic absorption spectrophotometer-Varian model spectral AA. Each shellfish samples were analyzed in triplicate and analytical blank was run as same way as the samples run. The standards were prepared in the same acid matrix using mono element certified reference solution for AAS standard (Merck).

Metal pollution Index

To compare the total heavy metal concentration in the different sampling species examined in this study, the following formal was used for metal pollution index (MPI)

MPI = (M1 x M2 x M3x…….x Mn) 1/n

where, Mn is the metal concentration expressed in µg/g in dry weight (dw) of investigating species.

Descriptive statistics and regression analysis were calculated among heavy metals to signify the results through Minitab 17.

RESULTS AND DISCUSSION

The mean heavy metals (Zn, Cu, Fe, Ni, Cr, Co, Pb and Cd) concentration (mg/kg dry weight) of four portunid crab species collected from Karachi coastal waters presented in table 1. The accumulation pattern of heavy metals in edible tissues of different crab species samples in decreasing order contents, were Zn > Fe > Ni > Cu > Co > Cr >Pb> Cd. The concentration of Zn was the dominant metal in crabs among all the studied metals.In statistical analysis, a linear regression of correlation (Table 2) showed high significant correlation between the sexes corresponding to Cu, Zn, Fe,Ni and Cd contents in both soft tissues ($p<0.05$). No significant correlations were determined in Cr, Pb and Co heavy metal in soft tissues of crabs. The level of these metals in shellfish species along the Karachi coast is due to the discharged of industrial and domestic wastes in the Karachi Harbour from Korangi creeks and Gizri. These highly toxic wastes enter in the coastal waters, results, increasing the levels of metals pollution in Karachi coast, thus our results confirm this trend.

Zn concentrations in collected crab species were ranged from 60.29mg kg^{-1} to 461 mg kg^{-1} with mean value 113.71± 71.77 mg kg^{-1}. Significant difference $p > 0.05$ was recorded between the four species. The mean concentration of the Zn was highest in the female of *S. serrata* 461 mg kg^{-1} and lowest in 60.29± 8.26mg kg^{-1} female of *P. sanguinolentus.* Females have a high accumulation of Zn was found in *S. serrata* and *P. sanguinolentus* while in males of *P. pelagicus* and *C. feriata* showed high accumulation than females.

Accumulation of Cu in procured samples showed concentrations ranged from 21.71 to 89.3 mg kg^{-1} with mean value 45.92 ± 42.56 mg kg^{-1}. The concentration of Cu was significantly different species. The highest concentration (89.3± 68.6 mg kg^{-1}) of Cu was found in female of *P. pelagicus.* Zn and Cu are essential metals for organisms and also human health, but in exceed can be posed harmful effect for human health.

Cu is an important metal that plays specific roles in the development and growth of an organism and main component of haemolymph (Van Aardt and Erdman 2004) also essential in the development and maintenance of eye and cytochrome activities (Gupta and Mathur 1983; Chona *et al*., 2015). The highest concentration of Zn was detected in the crab’s tissues than Cu concentration. High concentration was probably due to the major role played in different enzymatic activities by these heavy metals and hence, all the body parts have the ability to accumulate higher concentration of these metals (Kamaruzzaman *et al*., 2012).

The concentration of Ni in crabs was ranged 20.76 to 155.3mg kg^{-1} with mean value 72.71±59.90 mg kg^{-1}. The high concentration of Ni (155.3± 22.4mg kg^{-1}) recorded in males of *P. pelagicus.* The Ni showed high accumulation in male crabs then of females. The accumulation of Fe in crab samples ranged from 44.20 to 98.58 mg kg^{-1} with mean value 72.61± 36.69 mg kg^{-1}. Fe is the most abundant and well known transition element in biologic systems (Hoda *et al*., 2007).

The concentration of Pb was ranged 3.46 to 8.31mg kg^{-1} with mean concentration 4.776±4.187mg kg^{-1}. The highest concentration (8.31mg kg^{-1}) of Pb was observed in males of *C. feriata* and lowest (3.46mg kg^{-1}) observed in females of *P. sanguinolentus*. Pb is a most toxic, bio-accumulative metal had no known beneficial or biological function (Stankovic *et al.* 2012) therefore, its accumulation may pose serious risk to human health. In children’s Pb may induce reduced intellectual performance and cognitive development and in adults increase in blood pressure and cardiovascular diseases (OJEC, 2001).

The concentration of Co ranged 14.71 to 72.8 mg $kg^{-1}$and the concentration of Cr ranged 13.28 to 30.89 mg $kg^{-1}$with high accumulation of Cr was found in male of *P. sanguinolentus* (30.89± 0.787) (Table 1). Cr is an essential trace element in humans, fish, and shellfish, butits carcinogenic nature caused respiratory, gastrointestinal, hematological and reproductive effects through oral exposure (Jooste *et al*., 2015).In excess, however, it can be lethal to shellfish and other life (Akan *et al*., 2009).

The accumulation of Cd in Portunid crabs was ranged 0.03 to 0.66mg kg^{-1} with mean concentration 0.241±0.533 mg kg^{-1}. The highest accumulation (0.66mg kg^{-1}) was found in female of *P. pelagicus* and lowest (0.03mg kg^{-1}) in males of *C. feriata*.

Cd absorption makes up a risk to human health because it may accumulate in the human body as inducing skeletal damage, kidney dysfunction and reproductive deficiencies (OJEC, 2001). High levels of Cd are often associated with human activity (industrial emissions and the application of fertilizer and sewage sludge to farm land) (Jovic *et al.*, 2011).

The male crabs are the common inhabitants of low saline area, have tendency to accumulate higher metal concentrations compared to the female crab body, as females generally exhibit the variation in seasonal migratory behavior, especially in reproduction purposes in which exchange of metallic ions into the clean environment take place.Thus, the male crabs shows high accumulation heavy metals than the female crabs (Ololade *et al.*, 2008).

From Table 1. *P. pelagicus* showed great aptitude for bioaccumulation of Co, Cu, Cd and Fe in soft edible tissues. *S. serrata* manifested the most bioaccumulation capacity of Zn and Ni. *P. sanguinolentus* had stronger ability for bioaccumulation of Cr. *C. feriata* showed great ability to accumulate the Pb in their edible tissues.

Table.1 mean St.dev of Essential, toxic and bordered line metals in different *Portunid* species found along Karachi coast.

Species	Gender	Essential metals				Toxic Metals		Borderline metals	
		Co	Cu	Fe	Zn	Cd	Pb	Cr	Ni
S. serrata	F	31.40 ±13.28	51.85 ± 6.61	59.16 ±18.76	461 ± 422	0.340 ± 0.142	3.46 ± 1.19	13.28 ± 10.47	58.7 ± 50.9
	M	29.62 ±25.1	49.77 ± 23.5	73.5 ± 5.0	128.3 ± 73.9	0.145 ± 0.180	3.93 ± 2.11	29.2 ± 38.3	74.2 ± 65.2
P. pelagicus	F	72.8 ± 66.8	89.3 ± 68.6	98.6 ± 30.5	146.3 ± 86.2	0.66 ± 0.89	4.28 ± 1.020	15.96 ±10.78	113.4 ± 78.2
	M	14.71 ± 3.4	24.75 ± 4.1	71.9 ± 9.85	70.6 ± 16.9	0.00 ± 0.00	4.550 ± 1.344	22.81 ± 0.477	155.3 ± 22.4
P. sanguinolentus	F	41.29 ± 20.8	26.29 ± 9.86	53.8 ± 21.2	60.29 ± 8.26	0.23 ± 0.636	3.350 ± 0.557	30.12 ± 0.316	20.76 ± 23.49
	M	27.4 ± 33.1	21.71 ± 10.81	44.20 ± 17.76	61.17 ± 25.19	0.12 ± 0.302	5.33 ± 6.06	30.89 ± 0.787	42.24 ± 22.09
C. feriata	F	42.06 ±20.55	22.05 ± 6.98	74.95 ± 13.16	85.72 ± 14.37	0.00 ± 0.00	4.287 ± 1.146	28.24 ± 2.267	80.4 ± 39.2
	M	40.9 ±32.1	32.81 ± 26.05	77.33 ± 9.59	119.9 ± 49.7	0.03 ± 0.10	8.31 ± 8.61	29.66 ± 2.483	64.7 ± 31.9

Table 2. Regression analysis in Toxic, borderline and essential metals.

Categories	Metals	R2	F	P
Toxic Metals	Cd	15.03	3.30	0.02***
	Pb	7.38	1.49	0.22
Border line Metals	Ni	27.72	7.16	0.00*
	Cr	8.87	1.82	0.15
Essential metals	Co	11.07	2.32	0.08
	Cu	28.14	7.31	0.00*
	Fe	20.14	4.71	0.005**
	Zn	15.73	3.49	0.02***

Level of significance: 0.05***, 0.01**, 0.001*

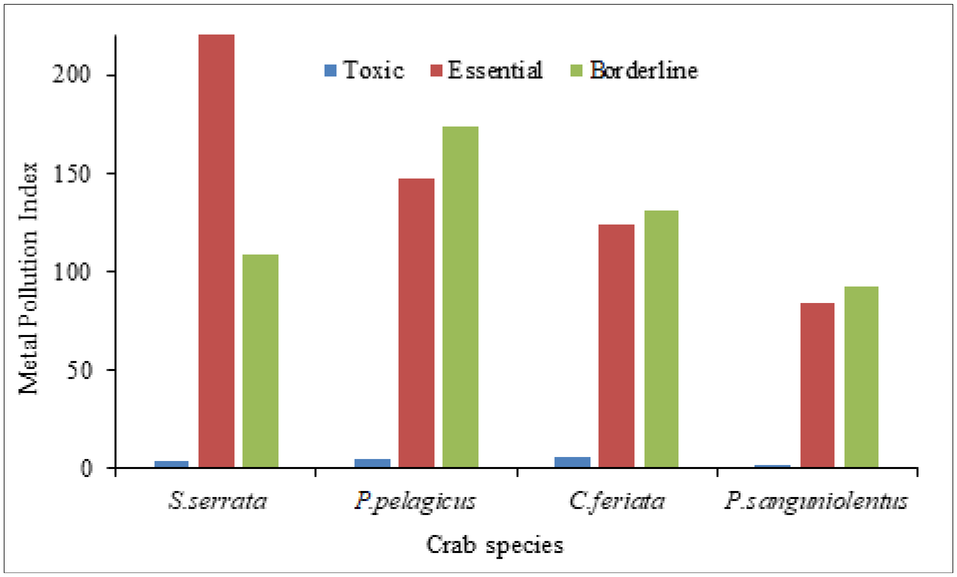

Fig. 1. Metal pollution Index of toxic, essential and borderline heavy metals in four Portunid crabs species.

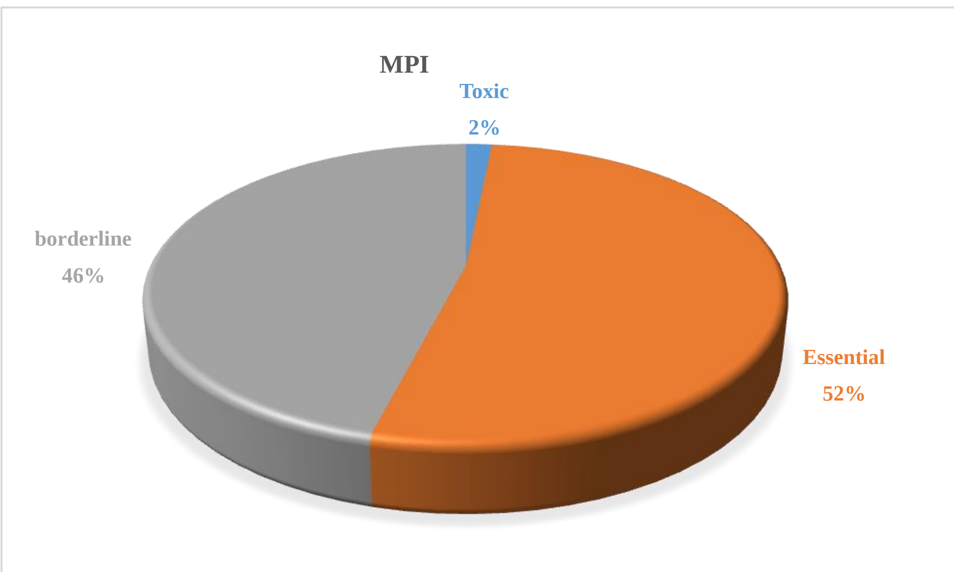

Fig. 2. Overall Metal Pollution Index of Essential, Toxic and borderline heavy metals.

Metal pollution index

The MPI of toxic, borderline and essential heavy metals present (Fig. 1), showed that the high MPI of toxic metal was recorded in *C. feriata* (6.313), MPI in borderline metals was high in *P. pelagicus* (173.32) and in Essential metals was high in *S. serrata* (221.075). Therefore *P. sanguienolentus* showed low values of MPI in toxic, bordered line and essential metals. On the other hand, the MPI was higher in essential metals than of borderline and toxic metals (Fig. 2).

The use of Portunids crab species as bioindicators turned out to be significant for the evaluation study of metal contamination level in the coastal area. Still, no single species are universally acceptable for Biomonitoring, as is factual of any marine organism used in these studies. It is suggested that when the Biomonitoring approach is planned, use those species which found and perform significant roles at different trophic levels, in combination with hydrographic and chemical measurements. It is important that any Biomonitoring study should be specific and comprehensive to the requirements of the exact site so that all issues need of potential concern can be reliably observed.

REFERENCES

Akan, J. C., F.I. Abdul –Rahman, O. A.Sodipo and P.I. Akandu (2009). Bioaccumulation of some heavy metals of six fresh water fishes caught from Lake Chad in Doron Buhari, Borno State, Nigeria. *Journal of Applied Science in Environmental Sanitation,* 4(2): 103-114.

Ali, S., F. Begum, S.A. Hussain, A.S. Khan, H. Ali, T. Khan, G. Raza, K. Ali and R. Karim (2014). Biomonitoring of heavy metals availability in the marine environment of Karachi, Pakistan, using oysters (Crassostreasp). *International Journal of Biosciences,* 4(7): 249-257.

Arya, S., J.N. Trivedi and K.D. Vachhrajani (2014). Brachyuran Crabs as a Biomonitoring tool: A Conceptual Framework for Chemical Pollution Assessment. *International Research Journal of Environment Sciences,* 3(1): 49-57.

Beg, M.A., M. Ali and A.H.K. Yousuf Zai (1975). Industrial effluents their nature and disposal in Karachi region. Part 1: Survey of the Pollution Lyari River. *Proceeding Pakistan Academy Science*, 12: 125- 131.

Carro, N., I. García, M. Ignacio and A. Mouteira (2004). Possible influence of lipid content on levels of organochlorine compounds in mussels from Galicia coast (Northwestern, Spain). Spatial and temporal distribution patterns. *Environment International,* 30 (4): 457- 466.

Chona C.E.V., C. G. Ramos and M.C. Ablan-Lagman (2015). Heavy Metal Levels in Mud Crabs (*Scylla* spp.) from East Bataan Coast. DLSU Research Congress. De La Salle University, Manila, Philippines March 2-4, 2015

Chiarelli, R. and M.C. Roccheri (2014). Marine Invertebrates as Bioindicators of heavy metal pollution. *Journal of Metallurgy*, 4: 93- 106.

OJEC (2001). Commission Regulation N° 466/2001 of 8 March 2001. Setting maximum levels for certain contaminants in foodstuffs. *Official Journal of the European Communities*, L77: 1-13.

Gu, Y.G., H.H. Huang and Q. Lin (2016). Concentrations and human health implications of heavy metals in wild aquatic organisms captured from the core area of Daya Bay's fishery resource reserve, South China Sea. *Environmental Toxicology and Pharmacology*, 45: 90–94.

Gu, Y.G., Q. Lin, X.H. Wang, F.Y. Du, Z.L.Yu and H.H.Huang (2015). Heavy metal concentrations in wild fishes captured from the South China Sea and associated health risks. *Marine Pollution Bulletin*, 96: 508–512.

Gupta, B.N., and A.K. Mathur (1983). Toxicity of heavy metals. *Indian Journal of Medical Science,* 37:236-240.

Harris, R.R. and M.C.F. Santos (2000). Heavy metal contamination and physiological variability in the Brazilian mangrove crabs *Ucides cordatus* and *Callinectes danae* (Crustacea: Decapoda). *Marine Biology*, 137(4): 691-703.

Hedberg, Y.S., C.Lidén and I.O. Wallinder (2014). Correlation between bulk- and surface chemistry of Cr-tanned leather and the release of Cr(III) and Cr (VI). *Journal of Hazardous Materials*, 280: 654–661.

Hoda, H.H., A.M.Ahady, A. Abdullah and T.T. Fatty (2007). Assessment of heavy metals an non-essential content of some edible and soft tissues *Egyptian Journal Aquatic Research,* 33(1): 85-97.

Jooste, A., Marr, S. M., Addo-bediako, A., & Luus-powell, W. J. (2015). Sharptooth catfish shows its metal: A case study of metal contamination at two impoundments in the Olifants River, Limpopo river system, South Africa. *Ecotoxicology and Environmental Safety,* 112: 96–104.

Jović, M., A. Stanković, L. Slavković -Beskoski, I. Tomić and S. Degetto, *et al.*, (2011). Mussels as a bio-indicator of the environmental quality of the coastal water of the BokaKotorska Bay (Montenegro). *Journal of the Serbian Chemical Society*, 76 (6): 933-946.

Kamaruzzaman, B.Y., B. Akbar John, B.Z. Maryam, K.C.A. Jalal and S. Shahbuddin (2012). Bioaccumulation of Heavy Metals (Cd, Pb, Cu and Zn) in *Scylla serrata*(Forsskal 1775) Collected from Sungai Penor,Pahang, Malaysia. *Pertanika Journal of Tropical and Agricultural Science*, 35 (1): 183 – 190.

Kucuksezgin, F., A. Kontas, O. Altay, E. Uluturhan and E. Darılmaz (2006). Assessment of marine pollution in Izmir Bay: Nutrient, heavy metal and total hydrocarbon concentrations. *Environment International*, 32 (1): 41–51.

Liu, J.L., X.R. Xu, Z. H. Ding, J. X. Peng, M. H. Jin, Y.S. Wang, Y.G. Hong and W.Z. Yue (2015). Heavy metals in wild marine fish from South China Sea: levels, tissue- and species-specific accumulation and potential risk to humans. *Ecotoxicology*, 24:1583–1592.

Maanan, M. (2008). Heavy metal concentrations in marine molluscs from the Moroccan coastal region. *Environmental Pollution* 153, 176-183.

Monawwar, S., M. Qasim and G.H. Kazi (1999). Heavy metal level in fish and shellfish of Karachi coastal area. *Pakistan Journal of Marine Biology*, 5 (2): 143- 154.

Naqvi, I.I. (1993). Determination of Trace metals in Karachi coastal waters using Atomic Absorption Spectroscopy. *Journal of the chemical society of Pakistan,* 15 (1): 1-6.

Neiboer, E., and D.H.S. Richardson (1980). The replacement of nondescript term 'heavy metals' by a biologically and chemically significant classification of metal ions. *Environmental pollution*, B1: 3-26.

Olmedo, P., A.F. Hernandez, A.Pla, P.Femia, A.Navas-Acien and F. Gil (2013). Determination of essential elements (copper, manganese, selenium and zinc) in fish and shellfish samples. Risk and nutritional assessment and mercury– selenium balance. *Food and Chemical Toxicology,* 62: 299-307. doi: 10.1016/j.fct.2013.08.076.

Ololade, I.A., L. Lajide, I.A.Amoo and N.A.Oladoja (2008). Investigation of heavy metals contamination of edible marine seafood. *African Journal of Pure Applied Chemistry,* 2:121–131.

Quraishee, G.S. (1975). Influence of the Indus River on the marine environment. In: International Conference on Management of the Environment. Pakistan Academy of Science, Islamabad. 111-112.

Spada, L., C. Annic chiarico, N. Cardellicchio, S. Giandomenico and A. Di leo (2013). Heavy metals monitoring in the mussel *Mytilus galloprovincialis* from the Apulian coast (Southern Italy). *Mediterranean Marine Science,*

Stankovic, S., M. Jovic, A.R. Stankovic and L. Katsikas (2012). Heavy metals in seafood mussels. Risk for human health. p. 311-373. In: *Environmental Chemistry for a Sustainable World. Volume 1: Nanotechnology and Health Risk.* Lichtfouse, E., Schwarzbauer, J., Robert, D. (Eds). Springer,Netherlands.

Tuzen, M. (2009). Toxic and essential trace elemental contents in fish species from the Black Sea, Turkey. *Food Chem. Toxicology*, 47: 1785–1790.

Van Aardt, W.J. and R. Erdman (2004) Heavy metals (Cd, Pb, Cu, Zn) in mud fish and sediments from three hard-water dams of the Mooi river catchment, South Africa. *Water*, 30:211-218.

Zehra, I., H, Naz and I.I. Naqvi (1996). A Preliminary study for establishing *Pernaviridis* (Mollusca, Bivalvia) as a biological monitor for pollution in Karachi coastal waters. *Journal of Marine Sciences*, *Pakistan,* 5(1): 31-40.

Zhou, Q., J. Zhang, J. Fu, J. Shi and G. Jiang (2008). Biomonitoring: An appealing tool for assessment of metal pollution in the aquatic ecosystem. *Analytical chemical acta*, 606: 135–150.

(*Accepted for publication January 2018*)