

## **SOME HEALTH HAZARD METALS IN COMMERCIALY IMPORTANT COASTAL MOLLUSCAN SPECIES IN BANGLADESH**

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**Abstract:** The present study was conducted to measure the heavy metals (Pb, Cd, Hg) concentrations in the muscles of four edible mollusk species (*Perna viridis*, *Crassostrea crassostrea*, *Sepia officinalis* and *Loligo edulis*) for two seasons. The heavy metal concentrations were analyzed by Atomic Absorption Spectrophotometer (AAS). The wet weight concentration of heavy metals varied for Pb: 0.03 - 0.05 mg/kg, Cd 0.03 - 0.04 mg/kg and Hg 0.01 - 0.023 mg/kg. The trend of metals in mollusk muscles were found as Pb > Cd > Hg. There was no significant variation of Cd and Hg concentrations in mollusk species that were analyzed, but Pb varied ( $F = 22.297$ ;  $p = 0.00$ ), in terms of both seasons and species. Principal Component Analysis and correlation matrix showed significant anthropogenic intrusions of Pb, Cd and Hg in mollusks. There was significant positive correlation between Cd vs Pb (1.00) and Hg vs Cd (0.447) indicates their common origin especially from industries and municipal wastes. The determined concentrations of all metals in the present study were lower than the limits permitted by World Health Organization (WHO) and European Union (EU) guidelines.

**Key words:** Heavy metals, edible, mollusks

### **INTRODUCTION**

Bivalves known as bioindicators of heavy metals pollution (Bryan *et al.* 1985, Goldberg *et al.* 1978, Burns and Smith 1981, Martin and Richardson 1991, Rainbow 1995, O'Connor 1996, 1998, Cantillo 1998, Lauenstein and Daskalakis 1998) in coastal areas because they tend to concentrate (Netpae and Phalaraksh 2009) trace metals. Long life span, high density and sessile (Otchere 2003, Rainbow 2007) nature make this creatures suitable for metal accumulation. These metals mainly originated from anthropogenic and natural sources (Gabr *et et al.* 2008, Yu *et al.* 2011, Muhammad *et al.* 2011, El Nemr *et al.* 2012, Salman 2012). Some heavy metals have adverse effects on aquatic organisms (Islam and Tanaka 2004, Yi *et al.* 2011) when accumulated at high concentrations ultimately causing human health risks when these organisms consumed (de Gieter and Baeyens 2005, Peter and Viraraghavan 2005). In comparison with other types of aquatic pollution, though the effects of heavy metal less visible, but the adverse effects on ecosystem and humans are intense and extensive due to their toxicity and their ability to accumulate in the biota

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(Shanmugam *et al.* 2007, Edem *et al.* 2008). Though some metals are essential for human growth, development, achievement, and reproduction (Hogstrand and Haux 2001), but excessive amount can cause serious illness (Damek-Proprawa and Sawicka-Kapustak 2003, Yi *et al.* 2011). Many researches carried out in the bioaccumulation of heavy metals in aquatic mollusks in different aquatic systems worldwide (Demina *et al.* 2009, Netpae and Phalaraksh 2009, Amisah *et al.* 2009, Shanmugam *et al.* 2007, Liu *et al.* 2007, Turkmen *et al.* 2005, Ohimain *et al.* 2008, Gaber *et al.* 2008, Salman 2006). Mollusks are very popular to coastal tribal people of Cox's Bazar but unfortunately very limited researches were done on heavy metal accumulation in mollusks in this area. This present research was conducted to determine the concentration of heavy metals and to assess the potential health risks at Cox's Bazar area.

### **MATERIAL AND METHODS**

Mollusk samples were collected from two points (Moheskhali Channel and fish landing centre) of Cox's Bazar (Fig. 1).

A total of 4 mollusk species were collected for individual season and then identified (following Quddus and Shafi 1983, Quddus *et al.* 1988, Roy *et al.* 2007, Rahman *et al.* 2009). After collection the mollusks were placed immediately in poly-ethylene bags and then kept into isolated container of polystyrene icebox. Finally, the samples were transferred to the Bangladesh Council of Scientific and Industrial Research (BCSIR) in ice box (Irwandi and Farida 2009, Ismail and Saleh 2012) where the mollusks were first washed with deionized water then sealed in polyethylene bags and kept in a freezer at  $-20^{\circ}\text{C}$  till analysis (Elnabris *et al.* 2012).

The heavy metal contents were determined by AAS using standard analytical procedure. To avoid contamination, glasswares were properly cleaned in acid water and the reagents used were of analytical grade. Deionized water was used throughout the study. Reagents blank determinations were used to correct the background errors. The techniques for samples preparation, standard preparation, analysis for metal analyses have been briefly described below.

This procedure was also used for destruction of organic matter. Precaution was to be taken to avoid losses by volatilization of elements. At first samples were homogenized. Then the samples were weighed accurately a suitable quantity (10 to 20 g) of the homogenized samples in a tared silica dish. After that the samples were dried at  $100^{\circ}\text{C}$  in a laboratory oven. These dishes were then placed in the muffle furnace at ambient temperature and slowly raised temperature to  $450^{\circ}\text{C}$  at a rate of no more than  $50^{\circ}\text{C}/\text{h}$ . The samples were ignited in a Muffle furnace at  $450^{\circ}\text{C}$  for at least 8 hrs. or overnight. After ashing was completed and cool, then the dishes were removed from furnace. Then the

ashes were dissolved in diluted nitric acid (Afthan *et al.* 2000). The solutions were returned to a hot plate and continued heating, adding additional acid as necessary until digestion was completed. Then the samples were filtrated into a 100 ml volumetric flask using Whatman No. 44 filter paper and washed the residue. Each sample solution was made up to the mark with distilled water.

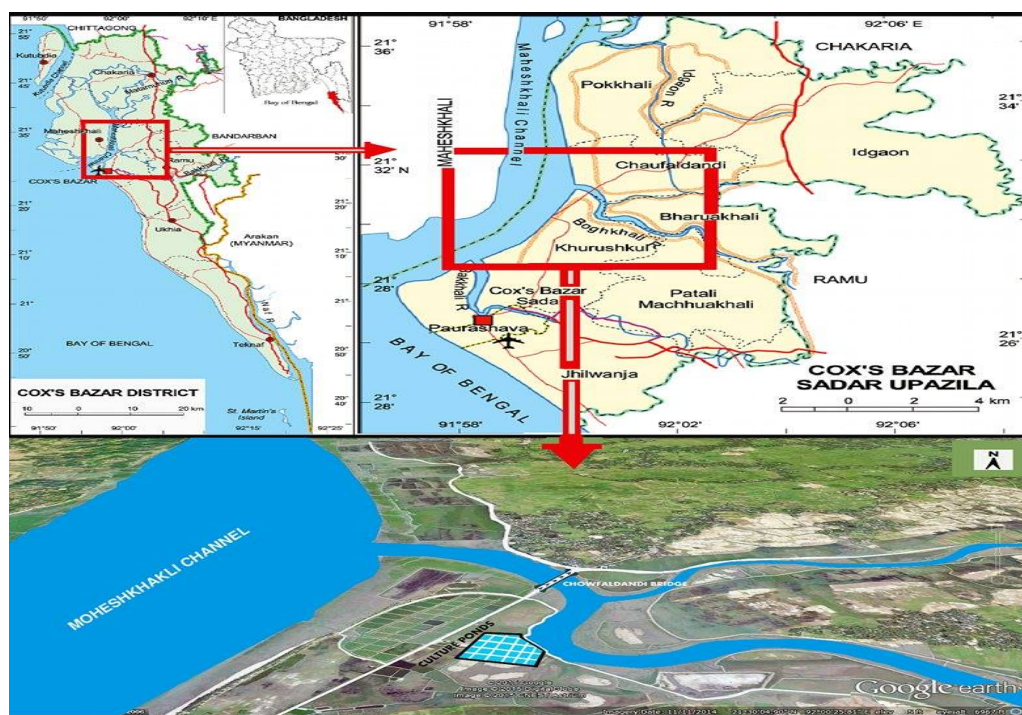


Fig. 1. Map showing sampling location.

Every metal standard solution was prepared for calibration the instrument for each element being determined on the same day as the analyses were performed due to possible deterioration of standard with time. All samples were prepared from chemicals of analytical grade with distilled water. 1 gm of metal cadmium, copper, lead, nickel were dissolved in  $\text{HNO}_3$ ; 1 g of cobalt, iron, manganese, zinc, aluminum were dissolved in  $\text{HCl}$ ; 2.8289 g  $\text{K}_2\text{Cr}_2\text{O}_7$  (= one g chromium) was dissolved in water and made up to 1 liter in volumetric flask with distilled water, thus stock solution of 1000 mg/l of Cd, Cu, Pb, Ni, Co, Fe, Mn, Zn, Al and Cr were prepared (Cantle 1982). Then 100 ml of 0.1, 0.25, 0.5, 0.75, 1.0 and 2.0 mg/l of working standards of each metal except iron were prepared from these stock using micropipettes in 5 ml of 2N nitric acid. One hundred ml of 2.0, 2.5, 5.0, 10.0 and 20.0 mg/l of working standards of iron metal were prepared from iron stock solution. Reagent blank was prepared in

the same manners of sample preparation without sample to avoid reagents contamination.

Finally, the atomic absorption instrument was set up carefully. In the meantime, flame condition and absorbance were optimized for the analyte. Then blanks (deionized water), standards, sample blank and samples were aspirated into the flame in AAS (Model: iCE 3300, Thermo Scientific). The calibration curves were found for concentration vs. absorbance. Data were statistically analyzed using fitting of straight line by least square method. For more accuracy, a blank reading was also taken and necessary corrections were made during the calculation of various elements concentration.

One way analysis of variance (ANOVA) was done to show the variations in concentration of heavy metal in terms of seasons and mollusks. According to Dreher (2003), Principal Component Analysis (PCA) was performed on the original data set (without any weighting or standardization). Pearson's product moment correlation matrix was done to identify the relation among metals to make the result strong obtained from multivariate analysis.

## RESULTS AND DISCUSSION

The specimens of four mollusk species (*Perna viridis*, *Crassostrea crassostrea*, *Sepia officinalis* and *Loligo edulis*) were analyzed for heavy metals (Pd, Cd and Hg) determination. During winter season, the highest concentration of Pb was found in *S. officinalis* (0.051) mg/kg. The lowest concentration was recorded in *P. viridis* (0.03) mg/kg (Table 1). In case of Cd, the maximum concentration documented in *S. officinalis* (0.041) mg/kg. Minimum amount was recorded in *L. edulis* (0.03) mg/kg. The highest value of Hg was recorded in *S. officinalis* (0.023) mg/kg while lowest value was in *C. crassostrea* (0.01) mg/kg. During rainy season, maximum amount of Pb was recorded in *S. officinalis* (0.045) mg/kg. Minimum concentration was measured in *P. viridis* (0.03) mg/kg (Table 1). The highest value was recorded in *L. edulis* (0.045) mg/kg and the lowest value recorded in *P. viridis* (0.03) mg/kg. The maximum concentration was found in *C. crassostrea* (0.02) mg/kg while the minimum concentration was recorded in *P. viridis* (0.01), *S. officinalis* (0.01) and *L. edulis* (0.01) mg/kg (Table 1).

In aquatic habitat, the relationship among metals in mollusks provide prevalent information about sources and pathways of variables. The result of correlations between heavy metals comply with the results found by PCA and CA that affirm some new associations between parameters. There was significant positive correlation between Cd vs Pb (1.00) and Hg vs Cd (0.447) that revealed their common origin especially from industries and municipal wastes (Table 2)

**Table 1. Heavy metal concentrations (mg/kg) in mollusks during Winter and Rainy season**

Name of species	Heavy metals	Winter season	Rainy season
<i>Perna viridis</i> (L.)	Pb	0.03	0.03
	Cd	0.04	0.03
	Hg	0.011	0.01
<i>Cressostrea crassostrea</i>	Pb	0.05	0.04
	Cd	0.04	0.04
	Hg	0.01	0.02
<i>Sepia officinalis</i>	Pb	0.051	0.045
	Cd	0.041	0.04
	Hg	0.023	0.01
<i>Loligo edulis</i>	Ld	0.05	0.035
	Cd	0.03	0.045
	Hg	0.02	0.01

**Table 2. Correlation matrix of heavy metals in mollusks**

		Lead	Cadmium	Mercury
Correlation	Lead	1.000		
	Cadmium	1.000	1.000	
	Mercury	0.447	0.447	1.000

**Table 3. Component matrix of two factors model with strong to loadings in mollusks**

Parameters	Component	
	PC 1	PC 2
Lead	0.966	-0.257
Cadmium	0.966	-0.257
Mercury	0.662	0.750
Eigen value	2.306	0.694
% Total variance	76.874	23.126
Cumulative %	76.874	100.000

Eigen values used as the extraction method to find out the principal components in PCA analysis. The components considered as principal components whose Eigen values was greater than 0.6 were taken into account. Two PCs were extracted by using correlation matrix which reflects the processes influencing the heavy metals composition having 100.0% of total sample variance (Table 3). The total variance of the PCs were 76.874 and 23.126% for PC 1 and PC 2, respectively. PC 1 is strongly correlated with Pb, Cd, Hg and PC 2 with Hg. The source of PC 1 and PC 2 can be considered as mixed source from anthropogenic inputs particularly from industrial effluents and agricultural activities in the study area.

The concentration of toxic metals (Pb, Cd, Hg) in 4 mollusk species were found to be below the WHO permissible concentrations given for seafood (WHO 1972, 1987). The concentration of Pb varied between (0.03 - 0.051) mg/kg/wet wt. that far below the limit set by the EU is 0.3 mg/kg/wet wt. (EU 2006). More or less similar result was found by (Staniskiene *et al.* 2006, Copat *et al.* 2012). Lead is extensively responsible for the reduced cognitive development and intellectual performance in children and augmented blood pressure and cardio vascular disease in adults (Commission of the European Communities 2001). The amount of Cd (0.03 - 0.04) mg/kg/wet wt. recorded in all mollusk species (*P. viridis*, *C. crassostrea*, *S. officinalis* and *L. edulis*) exceeded the limit (0.02 mg/kg) set by the (EU 2006). Cadmium can amass in the human body that might causes prostate cancer and breast cancer (Saha and Zaman 2012), kidney dysfunction, skeletal damage and reproductive deficiencies in human (Commission of the European Communities 2001). In the present study, estimated Hg concentration ranged between (0.01 and 0.023) mg/kg/wet wt that was far below the European dietary limit of 0.5 mg Hg/kg (Commission of the European Communities 2001). Mercury has acute toxicity that mercury may prompt changes in the normal development of the brain of newborns and at higher levels may induce neurological alterations in grown ups (Commission of the European Communities 2001). Mercury toxicity can affect the kidney, the developing fetus and it is a potential human carcinogen (Occupational Safety and Health Administration 2004).

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