

## Variation in heavy metals concentration in the edible oyster *Crassostrea madrasensis*, clam *Polymesoda erosa* and grey mullet *Liza aurata* from coastline of India

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### ABSTRACT

Seven trace metals Zinc(Zn), Iron(Fe), Manganese(Mn), Lead (Pb), Nickel(Ni), Cobalt(Co) and Cadmium(Cd) variation and (or) accumulation in mullet fish *L. aurata*, edible bivalve oyster *C. madrasensis* from Gosthani estuary-east coast and edible clam *Polymesoda erosa* from Mandovi estuary-West coast of India have been studied by Atomic Absorption Spectrophotometry (AAS). Overall trend in bioaccumulation was revealed with the following decreasing order Zn>Fe>Mn>Pb>Ni>Co>Cd. Among all the metals Fe showed highest percentage (79.5%) in clam followed by Zn (65.95) in oyster, which suggest and indicates that bivalve accumulates higher metal concentrations compared to fish. Four metals ie. Fe, Ni, Co and Mn showed high accumulation in gills of Mullet *Liza aurata* which is 10 times higher compared to muscle tissue, where as Pb and Zn were high in Oyster. 85% of the metals studied showed high concentration in gills. This study indicates the factors like feeding habits, habitat, size and regulatory ability plays important role in metal bioaccumulation shown by two different ecological niches.

**Keywords:** Heavy metals, Bioaccumulation, *Crassostrea madrasensis*, *Liza aurata*, *Polymesoda erosa*, AAS

### 1. INTRODUCTION

Heavy metal pollution is one of the serious problems and is of more concern due to nonbiodegradability of the metals resulting in to biomagnifications. Estuarine and coastal waters, sediments and biota act as a ultimate sink and accumulating means for trace elements that are discharged into the aquatic environment. Heavy metals entered in to the water body gets accumulated in the biota or sediments (Jeena et al 2010) and observed the average metal concentrations trend as Cr>Zn>Pb>Ni>Co>Cu thus infecting the humans by direct consumption of water or by consuming the affected organisms like fishes. Goldstein (1990) and Malik (2004) claim that when the level of trace metal concentrations exceeds the stipulated level, it turns out to be toxic. Accumulation of the metals in to biota depends on the physicochemical and biological factors. The ability of aquatic organisms to accumulate pollutants in their tissues makes them useful to study the rate of bioaccumulation in particular habitat. In recent years fishes and bivalves are widely used as an indicator of heavy metal accumulation in the aquatic and marine systems (Chen et al, 2004, Jeena & Abbas, 2010, Jean-Christophe et al, 2004, O'Connor, 1996, Phillips, 1976). Many studies have indicated the enhanced levels of heavy metals in tissues and different organs of fishes and bivalves (Chen et al, 2004, Sultana & Rao D 1998, Tuzen 2003, Uluozlu et al 2009).

In present study, muscle tissues and gills of *Liza aurata* (Mullet fishes), *Crassostrea madrasensis* edible oyster and *Polymesoda erosa* (as a whole organism) were used to analyze their metal concentrations. Sample selection was aimed to include two different ecological niches, the sedentary bivalve (oyster) and clam and the mobile fish (mullet). These two groups of organism were checked for the accumulation of seven metals, viz., Zn, Fe, Mn, Ni, Co, Pb and Cd, to help understand the influence of anthropogenic activities. Samples of mullet fish and oyster were collected from Gosthani estuary in Bheemunipatnam, East coast and clam samples from Mandovi Estuary West coast of India were analyzed for the heavy metals content. Gosthani is a small river which emerges in the Arakku valley, travels over the Andhra Pradesh and finally draining in to Bay of Bengal at Bheemunipatnam. River Mandovi is the largest river in Goa and is known as the lifeline of Goa. The river emerges in the Western Ghats, moving westward and meets the Arabian Sea. Both Gosthani and Mandovi estuary have been studied in various aspects earlier (Rao & Sivani, 2011, Shulkin & Presley, 2003, Attri K & Kerkar, 2011, Parulekar et al, 1986, Alagarsamy, 1998). A lot of fishing activities take place in the estuaries and this fish supplements the protein requirements of the inhabitants within the area, thus the contamination of fish and the aquatic environment by heavy metals is viewed with serious concern. This is the first time ever metal studies for two different niches (fish and bivalves) from Gosthani estuary and clams from Mandovi Estuary.

### 2. MATERIALS AND METHODS

#### 2.1. Sample selection

Samples of Indian backwater oyster *C. madrasensis* were collected from the intertidal area by hand picking whereas samples of Golden grey mullet *L. aurata* were collected with the help of local fishermen by operating a gill net. Golden grey mullet *L. aurata* is a pelagic species that is in frequent contact with sediments, feeding on small benthic organisms and detritus. Clam samples were collected with the help of local fisherwomen. The sedentary, filter feeder bivalves which can indicate the bioavailability of metals from water column, on the other hand mobile detritus feeder mullet fish can indicate the metal

**Table 1 Concentrations ( $\mu\text{g/g}$  dry wt) of heavy metals (Mean  $\pm$  SD) in fish organs & bivalves and metal permissible level by different authorities**

Group	Species	Organ	Element( $\mu\text{g/g}$ )						
			Fe	Zn	Mn	Ni	Co	Pb	Cd
Fish	<i>L. aurata</i>	Tissue	60.5 $\pm$ 40.5	46.6 $\pm$ 13.5	42.2 $\pm$ 16.7	16.5 $\pm$ 12.4	12.4 $\pm$ 7.4	2.2 $\pm$ .34	0.5 $\pm$ .19
		Gills	754.3 $\pm$ 350.2	252.7 $\pm$ 238	380.9 $\pm$ 176.1	156.1 $\pm$ 136	108.5 $\pm$ 54	1.8 $\pm$ .67	0.7 $\pm$ .33
Bivalve	<i>C. Madrasensis</i>	Full	415.1 $\pm$ 134.7	954.4 $\pm$ 58.7	48.4 $\pm$ 20.7	20 $\pm$ 10.6	20.4 $\pm$ 18.6	2.2 $\pm$ .73	0.8 $\pm$ .52
		<i>P. aerosa</i>	Full	711.7 $\pm$ 206.3	91.7 $\pm$ 10.2	53.9 $\pm$ 27.6	14 $\pm$ 9	26 $\pm$ 8.5	1.1 $\pm$ .5
WHO (1989)			50	150				0.2	0.2
FAO(1992)			30-100					0.5-0.6	0.2
MFR (1985)			100					2	1
NHMRC (1983)			<100						< 2
EC (2001)									0.05-1
FDA (2001)			40			80		1.7	4

**Table 2 Metal percentage in fish and bivalves**

	Cd	Pb	Co	Ni	Mn	Zn	Fe
Fish Tissue	0.278	1.421	6.814	12.011	21.854	23.524	34.094
Fish Gills	0.029	0.106	6.729	10.134	21.681	12.273	49.045
Oyster	0.049	0.151	1.392	1.390	3.109	65.953	27.954
Clam	0.048	0.113	3.025	1.143	6.751	9.407	79.510

**Table 3 Metal concentrations ( $\mu\text{g/g}$ ) in different oyster species from different area**

Species	Zn	Fe	Mn	Co	Ni	Pb	Cd	Location	Reference
<i>Crassostrea madrasensis</i>	1305 (882-254)	414 (199-597)	48.3 (27.4-86.5)	16 (4.2-31.5)	20 (5.1-22.7)	13.5 (ND-16.3)	10.4 (ND-13.2)	Gosthani estuary, East coast of India	Present Study
<i>C. ngulata</i> (wet wt)	-	-	-	-	-	3.14	0.228	Spain Atlantic sea and Mediterranean sea	Schumacher & Domingo(1996)
<i>C. angulata</i> ( dry wt)	-	-	-	-	-	0.02- 41.4	0.4 – 3.8	"	"
<i>C. virginica</i>	-	-	-	-	-	ND-7.9	0.44-3.5	Mississippi coast USA	Lyte & Lyte (1990)
<i>C. angulata</i>	-	-	-	-	-	0.08 -1.24	0.69-4.07	Huelva estuary Spain	Lopez Artiguez et al (1989)
<i>Pinctada radiate</i>	-	-	-	-	-	-	0.24-5.57	Arabian gulf (Arabian Saudi coast)	Sadiq and Alam (1989)
<i>C. verginica</i>	-	-	-	-	-	3.3 - 66.6	1.5-7.5	San Andres Lagoon, Mexico	Vazquez et al (1990)
<i>C.gigas</i>	4183	-	-	-	2.55	17.12	11.8	North western sea of Japan	Shulkin et al (2003)

bioavailability from water column. *L.aurata* and *C.madrasensis* are two abundant species in the estuary and are most preferred sea food diet of the locals and to the other markets. Whereas the clam *polymesoda erosa* is very common in markets and is of public interest.

## 2.2. Methodology

Analyses were carried out using 18 fish samples with size varying from 8-16 cm total length and 18 oyster samples of size varying from 3-5 cm shell length and 12 clam samples. Samples were cleaned of sand particles and mud by washing thoroughly with distilled water. Dorsal muscle tissue and gills were analyzed in each fish sample. Oysters and clams were used as a whole organism. All the samples were dried at 60°C for 72 hrs till the wet tissues reached to a constant weight. All the samples were crushed to powder using mortar and pestle. Acid digestion of the samples was carried out as per Schumacher M & Domingo (1996) method. Following acid digestion, all the samples were analyzed for seven metals using Perkin Elmer 3110 Atomic Absorption Spectrophotometer (AAS) with air acetylene fuel mixture.

## 3. RESULTS AND DISCUSSION

### 3.1. Trace metals contents in organs of fish and bivalves

Knowledge about heavy metals concentration in fish is important with respect to nature management and human consumption. Among seven metals only Pb and Cd are very toxic to human beings and has no biological role, but excessive consumption of these kinds of metals affect the human beings. Average levels of seven metals ( $\mu\text{g/g}$  dry wt.) in muscle and gill tissues of mullet fish (*L.aurata*), oyster collected from the Gosthani Estuary & Clams from Mandovi estuary are shown in Table 1. The average concentration of heavy metals in the fish organs and bivalves studied are in the following order of gills> Oyster>Clam>Muscle tissue. Highest concentrations of Mn, Co, Ni and Fe were found in the fish gills, where as Zn, Cd and Pb were high in oyster samples. The concentrations of the studied metals was in following decreasing order Fe>Zn>Mn>Ni>Co>Pb>Cd in fish and bivalves. Fe exhibited the highest concentrations in fish gills (754.3 $\pm$ 350.2  $\mu\text{g/g}$ ) and clams (711.7 $\pm$ 206.3  $\mu\text{g/g}$ ) except oyster where Zn was high (954.4 $\pm$ 58.7  $\mu\text{g/g}$ ), followed by Cd. Among all metals, the level of Pb and Cd was generally the lowest. Similar findings were reported by many researchers (Dural et al 2007, Tepe et al 2008, Turkmen et al 2008). Wide variation of metals concentration in fish and oyster was observed being in the same environment. Among bivalves, oyster showed high concentrations than clam except Fe & Mn (Table 1). Metal percentage in bivalves and fish has been shown in Table 2. Present study showed more concentration of metals in *C. madrasensis* compared to other oyster species from different area (Table 3). Followed by oyster, clams showed the high level of Cd, indicating higher concentrations of Cd in bivalves than fishes. High levels of metal in gills followed by oyster, clams and fish muscle can be due to filter feeding nature of bivalves, less enzymatic activity and also bivalves were used as whole organism for the metal analysis. It is observed that Fe concentration was the highest in both the study areas and both groups. With exception of Zn in oyster, the mean concentrations of metals in the muscle, gills, oyster and clams showed great variations, this may be related to the differences in ecological needs, metabolic activities, physiological role and feeding habits as well as regulatory ability

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**Table 4 Metal concentration (µg/g) in different mullet species from different area**

Species	Zn		Fe		Mn		Ni		Pb		Cd		Co		Location	Ref.
	G	M	G	M	G	M	G	M	G	M	G	M	G	M		
<i>Liza aurata</i>	203 (51.4 - 366)	72.2 (29.3 -135)	683 (174 - 1160)	67.4 (20.9 - 190)	364 (119 - 546)	42.1 (22 - 89.5)	157 (26.3 - 308)	16.5 (2.99 - 38.4)	230 (31.7 - 416)	20.2 (1.92 - 26.5)	7.44 (0.02 - 10)	4.02 (2.3 - 6.1)	115 (37.5 - 150)	12.3 (2.9 - 26.8)	Present study	
<i>Liza parsia</i>	44.74	25.5	-	-	28.88	10.28	-	-	-	-	-	-	-	-	Sunderban mangrove wet land N.E. India	Saha et al 2006
<i>Mugil cephalus</i>	73.49	6.56	-	-	-	-	-	-	9.43	1.22	0.51	0.16	-	-	Gosthani estuary, A.P. India	Sultana & Rao 1998
<i>Mugil cephalus</i>	719.75	24.35	-	-	-	-	-	-	16.98	3.12	2.23	0.51	-	-	Vizag harbour East coast of India	Sultana & Rao 1998
<i>Mugil sps</i> (Organ not specified)	26.9	-	-	-	-	-	-	-	0.08	-	0.047	-	-	-	Victoria bay Brazil	Joyeux et al, 2004
<i>L. aurata</i> (Organ not specified)	6.07	-	-	-	2.33	-	-	0.07	0.03	-	0.03	-	-	-	Southern Atlantic coast	Usero et al, 2003

G - Gill  
M - Muscle

(Jing et al, 2006). The mean concentrations of Fe in the muscles, Gills, oyster and clams were 59.9µg/g, 754 µg/g, 415.1 µg/g and 711.7µg/g, respectively. The adsorption of metals on to gill surface could be an important influence in total metal levels of the gill (Canli & Furness 1993). *L.aurata* is a detritus feeder and the uptake of detritus associated contaminants by fish may occur by respiratory and dietary routes, whereas the dermal route is usually minimal contributors of exposure, due to often effective barrier provided by the external epithelium. Also the gills are metabolically active organ and due to ion exchange property of the mucous layer covering this organ compared to fish muscle which are metabolically less active (Bhattacharya et al, 2006).

### 3.2. Variation of Trace metals in fish muscle, gills, oyster and clam

#### 3.2.1. Lead

Pb ranged from 1.7-2.8 ,0.7-3.1 ,1.1-3.2 and 0.5 -2.1 µg/g in Tissue, Gills, Oyster and Clam respectively. Pb concentrations were reported with the range of 1.4 to 8.9 and 1.9 to 7.9 µg/g in fish and oyster respectively in SE gulf of California coast (Soto-Jimenez et al, 2008) and 1.96-2.03 µg/g in fish gills from Malaysia coast (Fathi et al, 2012). Hence the present study values are within the range of earlier observed values. Tepe et al (2008) observed Pb concentrations from 0.43 to 31.10 µg/g in bivalve mollusk from Chile. Present study, Pb concentrations in fish and bivalves were within the literature range. Pb is highly toxic to aquatic organisms, especially fish resulting in to delayed embryonic development, suppressed reproduction, and inhibition of growth, increased mucous formation etc. The levels of lead in muscle tissue, oyster and clams were within the FDA, (1993a) limits and do not constitute any threat upon the consumption of these species (Table 4). In Mandovi estuary Pb concentrations in sediments were observed to be varying from 20-150 with average value of 49.5µg/g (Shynu R et al, 2012) and 4.5 to 46.5 µg/L (Alagarsamy, 1998).

#### 3.2.2. Iron & Manganese

Average concentration of Fe was 754.3±350.2, 711.7±206.3, 415.1±134.7 and 60.5±40.59 µg/g in Gills, Clam, Oyster and tissue respectively. High concentration was observed in gills followed by clam. In gills Fe concentration was almost 10 times higher than the tissue (Table 1) followed by clams; oyster and muscle tissue. Fe had significantly higher concentrations in fish organs and bivalves than other metals. In Mandovi Estuary high concentration of Fe (0.8 mg/g) & Mn (14 mg/g) was observed in sediments (Shynu R et al, 2012). Phillips D, 1976 studied Fe to be normally high in gills. Mn average concentrations were 42.2±16.7, 380.9±176.1, 48.4±20.7 and 53.9±27.6 µg/g in Tissue, Gills, Oyster and clam respectively. Mn observed to be high in gills followed by clams, while muscle tissue was least accumulated organ. Among bivalves, clams showed high concentrations of Fe and Mn than that of oyster which might have contributed by their habitat (Van den Broek et al 2002). Fe concentrations in muscle were similar to those reported by Chen et al, (2004), where as Mn values were towards higher side. High Mn(9.4%) and Fe concentrations (79.5%) in clams collected from Mandovi estuary might have contributed by mining activity as the Fe and Mn ore transportation by barges has become a common activity in the estuary which ultimately contributes for high content of these two elements in the estuarine and coastal regions off Goa (Parulekar et al, 1986). Mn concentrations in the fish and bivalves were below the limit of 0.7 mg/g as stated by Charbonneau C & Nash (1993). USFDA (United States Recommended Dietary Allowance) recommend daily Fe and Mn allowance for healthy life as 4 mg. Fe and Mn in fishes would also be important nutritional trace elements for the general public.

#### 3.2.3. Zinc & Cadmium

High levels of Zn and Cd were observed in oysters. Higher accumulation of Zn and Cd in estuarine oysters than the coastal specimens have been reported earlier by Ke & Wang (2001). According to the results (Table 1), concentrations of Zn in the gills, muscle tissue, oyster and clam were 252.7, 46.6, 954 & 97.9 µg/g respectively. This high level of Zn in oyster is a consequence of high levels of calcium found in the tissues, demonstrating a competition for their uptake (Chen YC et al, 2004). Moreover, ecological factors such as season, location/environment of development, nutrient availability, temperature and salinity of the water, may contribute to variations in the metal concentrations in fishes. Ranges of Zn concentrations reported earlier in the muscles of Malaysian marine fish were 15.4-60.1 µg/g (Akan et al, 2009). Another study conducted in Langkawi Island showed that all species had higher concentrations of Zn than of other metals and that the concentrations in muscles ranged from 34.3-49.4µg/g (Irwandi & Farida, 2009). The Zn concentrations in the fish muscles detected by the present study are similar to those reported by Agusa et al, (2005) and are within the literature range.

#### 3.2.4. Cobalt and Nickel

In this study, the concentrations of cobalt were the highest in the gills and ranged from 2.9 -26.9µg/g in muscle, 4.2 to 85 µg/g in oyster, 11 to 33.6 µg/g in clam's and 41 to 209 µg/g in gills. Whereas Ni varied from 26.3-308 µg/g in gills, 2. 99-38.4 µg/g in muscles and 5.1-22.7 µg/g in oysters.

### 3.3. Statistical analysis

The analysis of variance also showed that the mean concentrations of metals in the organs of each species were significantly different ( $p < 0.05$ ,  $n = 18$ ) in fish and oyster species except for Cd. Strong positive correlation between Cd and Mn ( $r=0.88$ ,  $n=18$ ) in gills, between Ni and Cd ( $r=0.67$   $n=18$ ) in Oyster and between Zn and Ni ( $r=0.79$   $n=18$ ) in clam indicates the strong association between them. Positive correlation of Mn between Gills and tissue ( $r=0.50$   $n=18$ ), which might be due to ion



exchange property of the mucous layer covering these organs compared to bivalves and also due to complexing ability of Mn ions. Metal percentage in all the samples was in following decreasing order. Fe>Zn>Mn>Ni>Co>Pb>Cd. Percentage decreasing order was same as the metal concentration order. Cd and Pb showed the least percent.

#### 4. CONCLUSIONS

Results of the present study showed the wide variation in metal concentrations within the same organism depending on the route of absorption. Metal concentration showed the following decreasing order Fe>Zn>Mn>Ni>Co>Pb>Cd. whereas group wise order is as follows gills> Oyster>Clam>Muscle tissue. Highest concentrations of Fe, Mn, Co and Ni were found in the gills of fish, where as Zn, Cd and Pb were high in oyster samples. Correlation analysis proved that the mean concentrations of metals in the organs of each species were significantly different ( $p < 0.05$ ) in fish and oyster species. Among all the metals Fe showed highest percentage (79.5) in clam followed by Zn (65.95) in oyster, which suggest and indicates that bivalve accumulates higher metal concentrations compared to fish which supports the studies by Stephen de Mora et al, 2004. Least percentage was of Cd and Pb ranging from 0.02 to 0.27% and 0.1 to 1.42% respectively. Even though Fe, Zn and Mn were high in bivalves and fish it may not be harmful, but it can act as an important nutritional trace elements for the general public. Although 85% of the metals studied showed high concentration in gills than in fish muscle it may not cause harm as general public did not normally consume the gills.

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