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COMPARISON OF UPTAKE OF HEAVY METALS AMONG THE SPECIES *Villorita cyprinoides* AND *Perna viridis*

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ABSTRACT

Heavy metal pollution due to increased industrial activity is of particular concern as they have deleterious effects on biota through mobilization and accumulation in various trophic levels. The present study mainly focused on the water pollution by heavy metals in Cochin estuary. In this paper, literatures on heavy metal sources, level and spatio-temporal distribution in water, sediments and organisms were reviewed for a comprehensive understanding of the status of heavy metal pollution in Cochin backwater. The methods include collection of surface water from the study area and their subsequent heavy metal analysis. The study revealed that the concentrations of heavy metals were high in the biota. The use of aquatic organisms as bio-indicators for trace metal pollution is very common these days. Aquatic organism can be exposed to heavy metals through uptake of water, ingestion of sediment particles and via food chain. Bivalves have a tendency to bioaccumulate heavy metals at higher concentration more than the sediment or water in their environment. Bivalves are filter- feeders and feed through their gills. They play an important role in aquatic food chains because they represent the primary consumers in the ecosystem.

KEYWORDS: Cochin Estuary, Heavy Metals, Water Pollution, Industrial Pollution, Bivalves

Heavy metals are regarded as serious pollutants of aquatic ecosystems, because of their environmental persistence, toxicity, and ability to be incorporated into food chains Heavy metals from mining, smelting, agriculture, petrochemical industry, printing, aquaculture, electronic industry and municipal waste discharged to the aquatic environment can be bioaccumulated by the organism and biomagnified through food chain [Ciji and Nandan, 2014]. The sediments have a great capacity to accumulate and integrate heavy metals and organic pollutants even from low concentrations in the overlying water column.

Various urban and industrial activities facilitate the disposal of chemicals and heavy metals into the water bodies which has made the coastal marine ecosystem highly vulnerable to adverse impacts [Mohan and Omana, 2007]. This has caused deleterious impact on the aquatic organisms [Jayakumar et.al., 2001]. Metal contamination in aquatic systems is a matter of serious concern from human health point of view as many of the organisms forms an integral part of human diet. Therefore a better understanding of the status of heavy metal pollution is inevitable for a sustainable development of the coastal marine ecosystem.

The coastal zone of Kerala is unique with the presence of a large number of perennial or temporary backwaters (Kayals). It is endowed with rich biological and genetic diversity [Thomson, 2001]. The estuary is characterized by high productivity, acts as a nursery ground for many species of marine and estuarine fin fishes, molluscs and crustaceans. The low lying swamps and tidal creeks dominated by sparse patches of mangroves give shelter to juveniles of many important species. The areas of the backwater with fine sediments and rich organic matter supports abundant and diverse benthic fauna [Menon et.al., 2000]. The booming city of Cochin and 60% of the chemical industries of Kerala located in the vicinity of the backwater discharge nearly 0.105 Million m^3/d of effluents, [Anonymous, 1998]. Therefore Cochin backwater is greatly affected due to pollution and toxicity problems particularly from heavy metals, pesticides, PCBs.

Molluscs such as oysters, mussels, cockle and clams have been widely employed as biomonitors for heavy metals pollution due to their ability to accumulate metals without harming themselves. Particularly bivalve molluscs have been considered as a potential biomonitor for metallic contamination in aquatic ecosystems [Kumar et.al., 2011]. They are sedentary and sessile filter-feeders, having a wide geographical distribution. Because of their abundance in coastal water and their ability to accumulate several classes of pollutants, they have been chosen as a suitable organism for mussel watch monitoring programs [Heng et.al., 2004]. Marine organisms are characterized by a greater spatial ability to accumulate some metals when compared with bottom sediments [Subathra and Karuppasamy, 2008]. Filter feeding bivalves capable to accumulate heavy metals in their tissues and numerous studies were attributed. Mussels can accumulate cadmium (Cd) in their tissues at levels up to 100,000 times higher than the level observed in the water in which they live [Manahan, 2000]. The shellfish is widely used because they are an important source of protein for coastal

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communities. The use of bivalves as biomonitors of heavy metal pollution has been widely reported. This is due to their characteristics from ecological and biological points of view which are advantageous for biomonitoring [Kaladharan et.al., 2005] [Avelar et.al., 2000].

The present study was conducted as a preliminary survey on sediment pollution of estuarine wetlands in Ezhikkara and Ulliyannoor, part of Cochin estuary. The aims of this study were to: 1) determine the concentration levels of some heavy metals copper (Cu), lead (Pb) and cadmium (Cd) in the estuarine surface sediment; 2) evaluate the heavy metals pollution in the water and the bivalve species – *Perna viridis* and *Villorita cyprinoides*.

MATERIALS AND METHODS

Arrangement of Sampling Sites

Cochin backwaters extend from Cochin to Alleppey, with an area of 256 km^2. It has two permanent openings, one at Cochin, which forms the main entrance (450 m wide) to Arabian sea and another opening, further north at Azhikode. Six rivers (Achenkovil, Pampa, Manimala, Meenachil and Muvattupuzha in the south and a branch of Periyar in the north) are discharging about $2\times10^{10}m^3/y$ of fresh water, >60% of which is during summer monsoon (June - Sep). Among six, Periyar and Muvattupuzha are the major rivers emptying to the system. The present study was conducted at Ezhikkara panchayat and Aluva. Four stations were selected with 100 m apart in both the regions.

Sampling Strategies and Sample Processing Methods

Water samples were collected using Niskin water sampler and filtered through 0.45µm pore size filter paper. The filtrate was then acidified with 1N HCl or 1 N HNO_3. Dissolved metals were extracted with APDC (Ammonium pyrrolidinedithio carbamate) - DDDC (Diethylammoniumdiethyldithiocarbamate)-Chloroform or APDC-MIBK. Sediment sample for heavy metal analysis were collected using Van Veen Grab and dried thoroughly. Finely powdered samples were then digested with mixed acids such as HNO_3 and HCl. The samples were washed in a tap water using a plastic brush to remove any dirt attached to the shell surface. The shells and flesh of each bivalve were separated, dried in the oven at 60°C for 2 days, then grounded using a mortar and pestle and stored in the desiccators. Tissue samples were dried in oven and metals were extracted with HNO_3 and $HClO_4$. Metal concentration in all the samples were analyzed by either AAS (Atomic Absorption Spectrophotometer) or ICP-AES.

The two species of bivalves were collected with the help of divers. The species *Perna viridis* was exposed to 0.1, 1 and 5 mg/l concentration of Pb, Cu and Cd for byssogenesis study. The species was kept under laboratory condition and was fed with algae before been exposed to the metals.

RESULTS

Concentration Levels of Heavy Metals in Bivalves

The concentration levels of heavy metals varied from species to species and size of the bivalves (Fig 3). The range of heavy metals concentration in all the species was as follows: Cu: 8.4±0.48µg/g to 11.2 µg/g, Pb: 10 µg/g to 64 µg/g, Cd: 1.4 to 7.1µg/g. Pb had concentration levels ranging between 63.59±2.42 to 70.35±2.47 µg/g while Cd 4.2±0.67 to 13.92±1.24 µg/g was detected in high concentration in the shells of all organisms. The high concentration level of these toxic metals could be attributed to discharge of municipal waste at the shores as well as from the paints or oils from industries. These metals are toxic and are not metabolized in the bivalves hence are easily excreted from the flesh.

The test results showed that the byssus production in Perna viridis in 0.1, 1 and 5mg/l treatments was significantly different from the controls. The length of byssus range between 0.7 to 0.4cm. At higher concentration the byssus was absent.

The concentration level for water and sediment samples were within acceptable limits set by USEPA. Cd level was 0.4 to 0.8 for the sediment samples in both sites, while in water samples, were below detection limit. Cd is a rare metal and its occurrence is majorly due to anthropogenic activities like mining and from industrial waste. Table1 shows the concentration of heavy metals in the water samples.

During the monsoon season the lead concentration in water samples was 0.012±0.002 mg/L in Ezhikkara, while in sediment samples, the concentration levels were 20.0±2.82 µg/g at Ezhikkara and 10.0±1.50 µg/g at Aluva (Ulliyannoor). The concentration of lead in the water sample from Ulliyannoor was between 3.88-4.42mg/l.

The concentration of copper in water samples 0.0043±0.00071 mg/L from both the sites, while sediment ranged from 13.2±0.14µg/g at Ezhikkara and 19.0±1.34

µg/g at Ulliyanoor. Table 2 shows the concentration of heavy metals in the sediment.

Table 1: Concentration of Heavy Metals in Water Sample during Monsoon Season

Sample	Lead (Pb) mg/l	Copper (Cu) mg/l	Cadmium (Cd) mg/l
A1	0.014	0.0043	Bdl
A2	0.012	0.0024	Bdl
A3	0.010	0.0026	Bdl
A4	0.010	0.0035	Bdl
B1	4.42	0.0042	Bdl
B2	4.23	0.0035	Bdl
B3	4.02	0.0026	Bdl
B4	3.88	0.0026	Bdl

Bdl: Below Detection Level

Table 2: **Concentration of Heavy Metals in Sediment Sample during Monsoon Season**

Sample	Pb (mg/g)	Cu (mg/g)	Cd (mg/g)
A1	22.82	13.34	0.8
A2	20.82	13.12	0.62
A3	18.64	13.2	0.45
A4	17.18	13.14	0.4
B1	11.50	20.34	0.81
B2	10.00	20.00	0.73
B3	11.00	19.00	0.4
B4	8.50	18.78	0.4

Table 3: Concentration of Heavy Metals in the Species

Sample		Pb(µg/g)		Cu(µg/g)		Cd(µg/g)	
		Tissue	Shell	Tissue	Shell	Tissue	Shell
P.viridis	**S**	10	63.59	8.4	-	1.4	4.2
	M	25	65.93	8.8	-	1.7	5.1
	L	64	70.35	11.2	-	7.1	13.92
V.cyprinoides	**S**	20	63.04	8.4	-	1.10	5.4
	M	22.4	66.54	8.4	-	1.21	6.2
	L	62.3	72.82	10.2	-	1.02	8.47

S –small, M – medium, L - large

BioSediment Accumulation Factor (BSAF)

Biota-Sediment accumulation factor is a ratio that measures efficiency of metal bioaccumulation in the flesh of the bivalves and the sediment. It is calculated as a ratio between concentrations of heavy metals in an organism CB, to the concentration of heavy metals in sediment CS. It is expressed as: BSAF= CB/CS. The BSAF was determined for Villorita and Perna. Table 4 represents the BSAF levels among the two species. Fig 1 gives a comparison of BSAF Levels

Table 4: BSAF Levels

Sample	Perna	V. cyprinoides
Pb	1.02	3.17
Cu	0.46	0.85
Cd	8.87	1.75

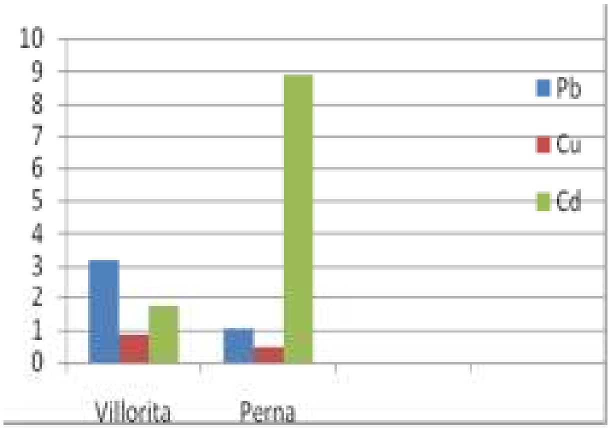

Figure 1: Comparison of Bio-sediment Accumulation Factor among the Species

Bioaccumulation Factor

Bioconcentration involves uptake of chemical pollutants by absorption from water that will result to the concentration of in the organisms being higher than in water .This is a ratio of the chemical concentration in an organisms CB, to the total dissolved chemical concentration in the water CWT and it is expressed as: BCF = CB/ CWT. The bioaccumulation factor for Cu and Cd was high in the bivalves, while concentration of Cu and Cd was low in the water samples compared to the bivalves. This means that the organisms can bioaccummulate high concentration of these metals compared to water. Table 5 shows the bioaccumulation factor of heavy metals.

Table 5: Bioaccumulation Factor of the Heavy Metals-Pb, Cu, Cd

Sample	Perna viridis	Villorita cyprinoides
Pb	-	31500
Cu	12000	5600
Cd	-	-

DISCUSSION

The high concentration levels in the shells could be because of the structural nature of the shells. They are made up of calcium carbonate in form of aragonite and calcite. The aragonite is isostructural to lead carbonate while calcium (9.7nm) has similar ionic radii with Cd (9.8nm), therefore the toxic metals have a tendency of replacing the calcium in the shells especially under low pH [Jung and Zauke, 2008] [Goldberg, 1975] . This study has showed that the concentration levels of the heavy metals in the bivalves are high. Mussels were found to be good bioaccumulators of Cu, Pb and Cd. Heavy metals bioaccumulation in bivalves is dependent on the body part. The concentration levels were higher in the flesh samples for Cu while Pb, and Cd were higher in the shell samples. These bivalves could pose a health risks if consumed because the levels of heavy metals concentration were high compared to WHO and FAO. The bivalves were also found to be better accumulators of heavy metals compared to water and sediment. The study suggests that the byssogenesis production in the species has potential to be used in biomarker studies.

CONCLUSION

Cochin backwaters are showing a high level of heavy metal contamination. Northern part of the estuary is found to be the most polluted compared to other part of the estuary. Heavy metal levels in the bivalve exceeded the safety limit. Therefore it is necessary to give more attention to accumulation of heavy metals in organisms as far as the seafood industry and public health is concerned. More precise environmental protection measures should be taken to control the discharge heavy metals from anthropogenic sources.

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