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Status of heavy metals in tissues of wedge clam, *Donax faba* (Bivalvia: Donacidae) collected from the Panambur beach near industrial areas

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Abstract

The wedge clam, *Donax faba* are edible bivalves and consumed as seafood. We found out the concentrations of Cu, Pb, Fe, Cr and Ni in the tissues of the wedge clams in coastal site, Panambur located along the south west coast of Karnataka. The concentrations of heavy metals in the samples were determined by atomic absorption spectrophotometer (AAS, GBC 932_{plus}) working with an air acetylene flame. *D. faba* showed seasonal variations of metals accumulation with maximum concentrations during all seasons occurred in our study. The analysis of Fe content showed higher average concentrations for wedge clam collected from the study site. The concentration was in the order of Fe>Cu>Cr>Ni>Pb in our present study. The levels of heavy metals like Cu, Pb and Cr in the whole tissue of *D. faba* were below permissible limits except Ni. High concentrations of metals in the whole tissue of wedge clams collected may be derived from the industrial areas and a variety of anthropogenic activities.

Keywords: *Donax*; Bivalve; Beach; Concentration; Pollution

INTRODUCTION

Benthic organisms are important component of the marine ecosystem and play an important role in maintaining ecological balance. Not only this, they act as a major food source for many fish species, even humans also consume some of the large sized crustaceans and bivalves. Bivalves, crustaceans and polychaetes are dominant among the macrobenthic forms and are considered as environmental indicators. In India, metal pollution is becoming a problem for local management especially in coastal waters and estuaries which are ultimately the repositories of pollutants from anthropogenic activities. Agricultural runoff and industrial effluents into aquatic system contaminate the ecosystem with mixtures of toxic or potentially toxic heavy metals. Due to structures and properties, many of these metals remain in the environment for prolonged periods and continue to pose environment problems and health risks.

Various processes through anthropogenic activities may bring to elevate in concentration of our natural waters [1] regarding heavy metals. They are discharged from mining, factories and municipal sewer systems, run-off from agricultural and urban areas, leaching from former industrial sites and dumps and atmospheric deposition. In the past decades, metal pollution resulting from recreational or industrial activities is becoming a great deal. Due to their potentially harmful effects on human health, the presence of heavy metals can limit the quantity of molluscs which humans can consume. In a case, the presence of heavy metals can affect the bivalves directly by damaging shell growth or killing their larvae, thus affecting their quality and quantity. For example, the presence of pollutant in the environment and bioavailability in the organisms could promote birth rate reduction and/or somatic growth rate reduction in the organism, with a consequent population growth rate decrease, affecting the studied population life cycle with all the future implications at ecosystem levels [2, 3]. Over the past several decades, the increasing use of metals in industry and factory has lead to serious environmental pollution through effluents and emanations [4- 8]. Under particular environmental conditions, heavy metals cause environment damage [9-11].

Bivalves are filter-feeder invertebrates and can accumulate heavy metals from food, water and also from the ingestion of inorganic particulate materials [12, 13]. Heavy metals pollutants are conservative and often highly toxic to biota [14]. The wedge clams, *Donax* spp. (Bivalvia: Donacidae) appear in marine waters in the lower and higher zones of both sandy and muddy beaches. The use of *Donax* spp. had been established to monitor the coastal areas [8, 15-23]. However, a little information on toxic responses of *Donax incarnatus* when exposed to combination of Cu, Cd and Hg had been studied from the Ambalapuzha beach (south west coast of India) [24]. Recently, a few studies on molluscs associated with heavy metal pollution have been done by some researchers [25, 26] in India, but the applications of wedge clam from the south west coast of India for pollution are lacking. Therefore, *D. faba* which are found in coastal site of Panambur as biomonitors of heavy metal contamination and bioavailabilities are focused upon here. Along the Karnataka coast, inshore waters lying adjacent to Karwar and Mangalore receive effluents from several industrial sources including port activities, fertilizer plant, iron ore processing plants, dyes and pigment processing plants, caustic soda plant, petroleum refinery and other sources. Therefore mainly untreated agricultural and municipal wastes and industrial production influence the inshore waters along the south west of India directly or indirectly. Generally, the natural sources of heavy metals in coastal waters are through wind flow and river runoff. The Central Pollution Control Board of India (CPCB)

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identified two cities, Mangalore and Karwar along the Karnataka coast as pollution “hotspots” [27]. In addition to this, the coastal ecosystems on the southern part of the coast have been identified as “stressed” by anthropogenic activity [28].

Generally, marine molluscs provide a cheap source of protein for human consumption. It had been reported that there was high protein for *Donax incarnatus* [29] found in Mirya Bay (India). In addition to this, other *Donax* species like *D. cuneatus* contained the polyunsaturated omega-3 fatty acids which are helpful in pronouncing less inflammatory responses towards bronchial asthma, lupus erythematosus multiple sclerosis, psoriasis and kidney diseases and also inhibit the development of cancer cells [30]. However, from the toxicological point of view, information on the wedge clams is not available in India. Our present study aimed to investigate whether these metals are within the permissible limits for human consumption. Estimation of heavy metals concentration is desirable for assessment of environmental pollution.

MATERIALS AND METHODS

Characteristics of the study area

Karnataka state consists of three coastal districts. They are Dakshina Kannada, Udupi and Uttara Kannada districts. The coastal area, which is blessed with 14 perennial rivers, is increasingly subjected to urban development. Sampling site, Panambur (12^{0}57′ N, 74^0 48′ E) is a sandy beach near New Mangalore Port located about 15 km north of Mangalore city (Dakshina Kannada) (Figure 1). This site is located in the industrial belt where Managalore Chemicals and Fertilizers Limited (MCF), Mangalore Refinery and Petrochemicals Limited (MRPL), Kudremukh Iron Ore Company Limited (KIOCL), dye and pigment processing plants and others are available. Netravati is one of the largest rivers in Karnataka, which originates in the Western Ghat and joins the Gurpur River to form a common estuary before entering the Arabian Sea, south of New Mangalore harbour. These two rivers are the main sources of sediments and flood water discharges into the coastal sea.

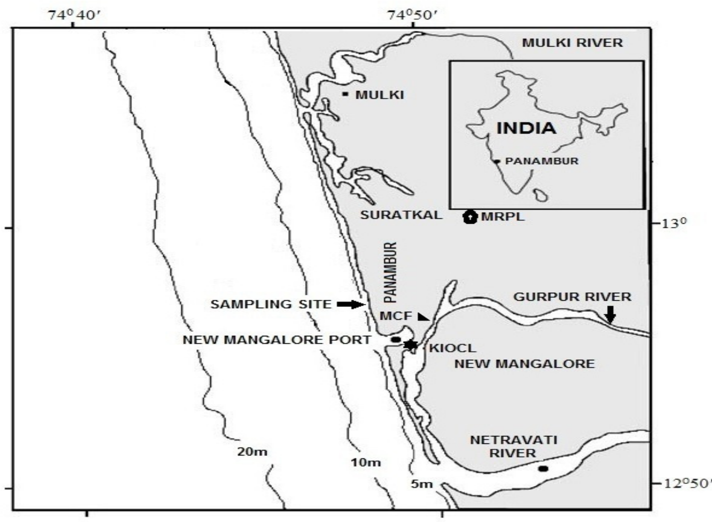

Fig 1. Location of sampling sites along the Panambur beach

Analysis of heavy metals

Samples of wedge clam, *Donax faba* were collected at monthly for a period of 12 months from April 2009 to May 2010 from the intertidal sandy beach at Panambur located in Dakshina Kannada. The individuals were individually measured for shell length (maximum antero-posterior distance accurately to 0.01 mm using vernier calliper. 10 individuals of the wedge clam, *Donax faba*, ranging in size between 20 mm to 23 mm were pooled monthly. The tissues of the wedge clams were washed with distilled water and removed with the help of a stainless steel knife and dried at a constant temperature of 60°C for 2 days and homogenized. 0.5 g of homogenized tissue was digested in triplicate. Digestion of the sample was carried out by adding 8 ml of nitric acid and 1 ml of perchloric acid. Afterwards, the sample was diluted with double glass-distilled deionised water and the distillate was filtered. The volume was made upto 25.0 ml and kept in precleaned plastic vials. The metals were detected on an Atomic Absorption Spectrophotometer (GBC 932 $_{plus}$) in an air acetylene flame. The concentrations of the heavy metals were expressed in µg/g dry weight.

Statistical analysis

Statistical analyses were performed using SPSS 16.0 statistical package. Data were expressed as mean. The data on values of heavy metals were subjected to cluster analysis (CA) which can identify relatively similar, that is homogeneous groups of seasonal patterns associated with metal concentrations,

RESULTS AND DISCUSSION

The monthly concentrations of heavy metals in the tissues of *Donax* species are shown in Figure 2 and 3. In the tissue samples of *Donax faba* from the Panambur beach, the concentration of Cu ranged from 12.30 to 38.60, Pb from 0.05 to 22.60, Fe from 172.15 to 1632.50, Cr from 3.00 to 12.15 and Ni from 7.70 to 24.55 µg/g dry weight. The mean values of heavy metal concentration in the tissue samples of clam at the site are given in Table 1. The mean concentrations of Cu, Pb, Fe, Cr and Ni metals were 21.84 µg/g, 4.22 µg/g, 501.11 µg/g, 6.23 µg/g and 5.76 µg/g respectively at the Panambur beach. The data presented in Figure 3 show that among the concentrations of heavy metals in all tissue sources, the concentration of Fe was higher than other metals in the tissues of the wedge clam. High concentration of Fe observed in the whole tissue of wedge clams collected from the coastal area adjacent to the industrial areas in Dakshina Kannada may be derived from the industrial areas and a variety of anthropogenic activities. Heavy metals and high nutrient discharging from the industrial areas through drain pollute the subbasin. The subbasin receives discharge of municipal sewage, industrial effluent and agriculture runoff.

Some values of heavy metals detected in this edible species were low as compared to the maximum acceptable limits (Table 2). The present study shows that the mean tissue concentrations of heavy metals, Cu, Pb and Cr in *Donax faba* collected from the study site along the Karnataka coast were found to be below the MAFF, BOE, NHMRC and EEC permissible concentrations for seafood [31-35]. But the Ni levels in the wedge clam exceeded beyond the WHO permissible level for sea food [36] (Table 2). In molluscs' tissues, the bioconcentration of heavy metal is influenced by numerous factors. It is observed that the concentrations of heavy metal accumulated by marine organisms are not only depending on the water quality but also salinity, seasonal factor, food intake, temperature, spawning and individual variation [12, 22, 37, 38].

The findings of the present study revealed that the concentrations of Cu and Ni at the Panambur beach were higher than the concentrations reported by Edward et al.,[38] except the concentrations of Pb and Fe for the same species, *Donax faba*. The concentration was in the order of Fe>Cu>Cr>Ni>Pb at the Panambur beach in our present study. Element concentrations in molluscs at the same location differ between different species and individuals due to species-specific ability/capacity to regulate or accumulate heavy metals [39, 40]. As benthic organisms, bivalves show bigger sensitivity for metallic pollutants. Such organisms represent the most reliable tool to monitor heavy metal contaminations [8, 16, 19, 23, 41-43]. In the present study, the concentrations of heavy metals were higher during the south west monsoon in all the samples. This condition might be due to the increased inputs of land derived metals due to rainfall as indicated by Asha et al.,[15].

Table 1: Comparison of heavy metal levels found in wedge clam tissues in several field studies. All concentrations are shown as µg/g dry wt (D) and µg/g wet weight (W)

Species	Location	Cu	Pb	Fe	Cr	Ni	Reference
Donax faba (D)	India	21.84	4.22	501.11	6.23	5.76	Present study
D. faba (D)	Malaysia	7.23	12.60	654.00	-	3.65	Edward et al.[18]
D. trunculus (D)	Mauritania	11.80	-	663.00	-	-	Roméo and Gnassia-Barelli[23]
D. trunculus (D)	Israel	17.40	6.22	-	-	-	Hornung and Oren[21]
D. trunculus (D)	Egypt	12.57	-	570.07	-	-	Abdallah and Abdallah[44]
D. trunculus (D)	Morocco	10.52	0.90	-	1.42	1.60	Idardare et al.[41]
D. trunculus (D)	Mauritania	8.88	-	-	-	-	Sidoumou et al.[49]
D. trunculus (D)	Spain	175.00	3.60	-	1.20	1.20	Usero et al.[42]
D. trunculus (W)	Italy	2.80	-	21.40	-	-	Mauri and Orlando[46]
D. rugosus (D)	Mauritania	20.60	-	-	-	-	Roméo *et al.*[48]
D. trunculus (D)	Turkey	1.93	0.57	324.29	-	2.01	Özden et al.[47]
D. deltoides (D)	Australia	6.80	-	-	-	1.16	Haynes et al.[19]
D. serra (D)	South Africa	-	-	181.00	0.70	3.00	Van As et al.[50]
D. serra (D)	South Africa	-	-	59.00	0.24	-	Van As et al.[50]
D. serra (W)	South Africa	1.18	0.03	84.00	0.26	0.43	Watling and Watling[53]
D. serra (W)	South Africa	1.20	0.06	79.00	0.82	0.23	Watling and Watling[53]
D. serra (W)	South Africa	1.10	0.14	81.00	0.60	0.38	Watling and Watling[54]
D. serra (W)	South Africa	1.00	0.06	231.00	0.80	0.33	Watling and Watling[54]
D. serra (W)	South Africa	0.90	0.03	81.00	0.60	0.28	Watling and Watling[54]
D. serra (D)	South Africa	3.50	1.90	236.00	0.90	1.20	Watling and Watling[51]
D. serra (W)	South Africa	1.60	0.03	72.00	-	-	Watling[52]
D. serra (W)	South Africa	0.82	0.34	81.00	0.65	0.47	Watling[52]
D. serra (W)	South Africa	0.64	0.34	42.00	0.17	0.21	Watling[52]
D. serra (W)	South Africa	1.29	0.76	79.00	-	-	Watling[52]
D. serra (W)	South Africa	1.18	0.03	84.0	0.16	0.43	Watling[52]

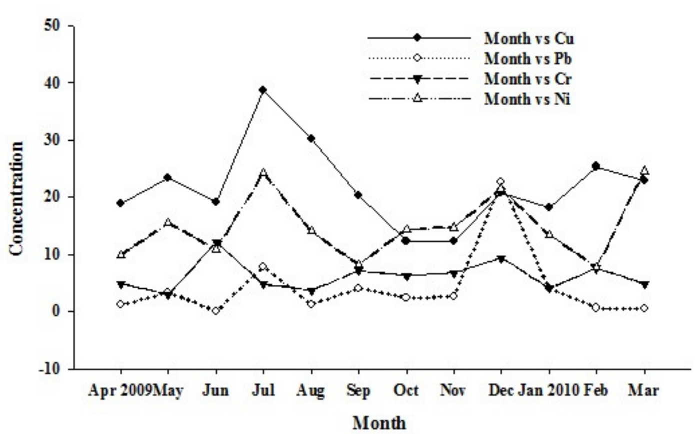

Fig 2.Trends of distribution of heavy metal (Cu, Pb, Cr and Ni) concentrations (μg/g dry weight) in wedge clams at the study site.

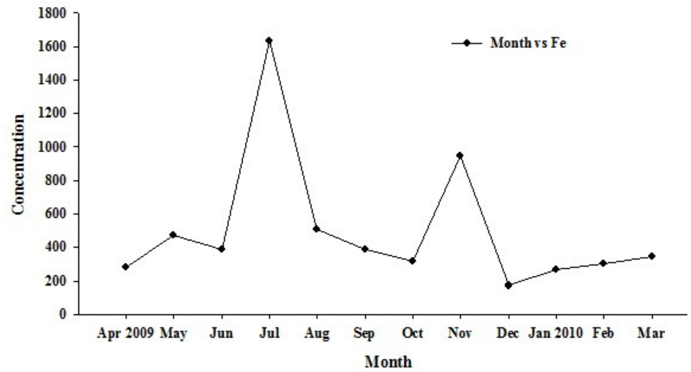

Fig 3. Trends of distribution of heavy metal, Fe concentration (μg/g dry weight) in wedge clams at the study site.

Table 2. Maximum acceptable limits (µg/g) of some heavy metals

Metal	µg/g	Reference
Cu	100	MAFF, 1956; BOE, 1991
	350	NHMRC, 1987
Pb	25	BOE, 1991
	50	Great Britian-Parliament, 1979
Fe	-	-
Cr	8	EEC, 1979
Ni	2	WHO, 1987

The hierarchical cluster analysis showed two major groups (A and B) (Figure 4). Group A with 2 two sub groups namely A1 (April, August, May, January, February and March) and A2 (June, September, July, November, October) separated at correlation coefficient 0.40. Group B separated from Group A at correlation coefficient 1 and contained only December. Temporal variations in marine heavy metals are not absolutely determined by seasonal effects but also the nature and frequency of discharge. On the other hand, heavy metals accumulation pattern in *Donax* is not only under metal enrichment effect, but it also affected by sex differences [22]. The toxicity of heavy metals is related primarily to the dissolved, ionic form of the metal rather than the total concentration of the metal.

The industrial wastes from the industrial areas are discharged into the coastal water through pipelines, while the municipal wastes of Mangalore city are discharged into drains adjoining the Gurpur River, which empties into the coastal sea off Mangalore. Such work may create aquatic pollution, resulting in severe ecosystem modifications and depletion of fishery resources. As shown in Table 1, heavy metal concentrations were found different as seasons and contamination levels of sites examined in several species of *Donax.* Significant correlations ($p<0.05$) were found for each of Zn and Fe in *Paphia textile* and of Co in *Donax*

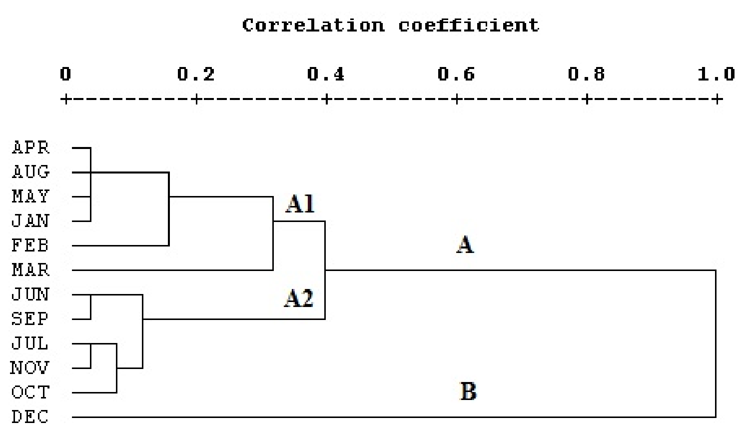

Fig 4. Dendrogram of temporal clustering of monitoring periods

trunculus relative to their concentrations in surficial sediments [44]. Usero et al.,[42] also reported there is a significant correlation for concentrations of heavy metals (Cu, Pb, Zn and Hg) in *D. trunculus* and *Chamelea gallina* relative to their concentrations in surface sediments. Similarly, a fair correlation between concentrations of pollutants in animal tissue and in sediments was observed [45]. The pattern of accumulation of heavy metals can cause toxic reactions in food chain.

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