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Research Article

BIOACCUMULATION OF METALS IN TISSUE OF MARINE ORGANISMS IN MUTTUKADU CREEK, KOVALAM, SOUTH EAST COAST OF INDIA

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ABSTRACT

Heavy metals were estimated in different types of marine organisms in fin fishes and shell fishes (*Rastrelliger kanagurta*, *Lates calcarifer*, *Ephinephalus malabricus*, *Penaeus monodon* and *Meretrix meretrix*) collected from the Muttukadu creek, Kovalam, south east coast of India. Concentrations of heavy metals (Cu, Cr, Zn, Pb and Cd) in the edible part of marine organisms in fin fishes and shell fishes were analysed by atomic absorption spectrometer. The lowest concentration of Cu, Cr, Zn, Pb and Cd were determined $2.74 \pm 0.23 \mu\text{g g}^{-1}$, $1.03 \pm 0.41 \mu\text{g g}^{-1}$, $4.49 \pm 0.27 \mu\text{g g}^{-1}$, $1.62 \pm 0.25 \mu\text{g g}^{-1}$, $3.70 \pm 0.18 \mu\text{g g}^{-1}$ and highest were observed $9.80 \pm 0.36 \mu\text{g g}^{-1}$, $3.62 \pm 0.28 \mu\text{g g}^{-1}$, $12.48 \pm 0.47 \mu\text{g g}^{-1}$, $7.63 \pm 0.33 \mu\text{g g}^{-1}$, $6.41 \pm 0.64 \mu\text{g g}^{-1}$ respectively. These results indicated that the heavy metal levels in the marine organisms in the studied areas were moderate but unacceptable Pb and Cd from the view of safety of seafood.

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INTRODUCTION

It has been detected that metals are a group of the most important pollutants; they cause environmental degradation in coastal areas. Heavy metals are introduced in to the aquatic ecosystems in various ways. Accumulation of heavy metals in the tissues of aquatic organisms at concentrations many times higher than concentrations in water and it may be biomagnified in the food chain to the level of causing physiological impairment at higher trophic levels and in human consumers. During the past two decades, it has been found out that, the metals and their relevant compounds both inorganic and organic, have released to the environment as a result of variety of anthropogenic activities (Komarnicki, 2005). Specifically the coastal zones with a view point of environmental base, can be considered as geographic space of interaction in between terrestrial and marine ecosystems. This is a very important factor which serves as protoplasm to the survival of many kinds plants, animals and marine species (Castro *et al*, 1999). It is further traced out that coastal pollution has been increasing significantly over the past few years and found expanding environmental problems in many developing countries. Not only this, but also the urban industrial expansion and the activities thereof, are causing permanently the marine pollution

to its highest degree further causing disturbances, permanently, leading to environmental and ecological degradation constituting potential risk to number of flora and fauna species, including humans, through food chains (Boran and Altinok, 2010).

Particularly Bivalve molluscs constitute one of the best biological indicators of coastal pollution, they exhibit symptoms of sedimentary nature, it is observed large part of geographical distribution, generally, there are many individuals at the sampling sites, and easily sampled with final result of accumulation of pollutants (i.e. contaminant levels in their tissue responsible for changes in environmental levels and pollutants accumulate with a little metabolic transformation (Chase *et al*, 2001). Rainbow (Rainbow, 1995) emphasized that mytilids widely spread over cosmopolitan of all marine genera and inhabiting contaminated in high degree of, coastal waters too tolerant of fluctuations in salinity, temperature and other physico-chemical parameters, so, these abilities are suitable to use them as biomonitors. The heavy metals are chosen as suitable pollutants which are widely spread over the environmental contaminants from either natural or anthropogenic sources and are believed to be a threat to the aspect of life itself of many a marine animal besides crustaceans (Lorenzon *et al*, 2001). The contaminated fish and

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crustaceans from watery environment will surely become a public health concern. So it has been determined to take notice of the concentration of heavy metals in fish to be marketed along with shrimps, thus the risk of human consumption can be clearly evaluated (Cid *et al*, 2001).

Pollution creeps into fin and shell fishes through five ways by way of food and non food particles, gills, oral consumption of water and the skin. The use of fin and shell fishes as bioindicators of metal pollution in the aquatic environment and its conformity for human use has been documented toxicologically (Amin *et al*, 2011). Not only this, the sensitivity of crustaceans to heavy metals is also perfectly documented and for all these reasons, the importance of marine shrimp for environmental monitoring the studies as bioindicators of heavy metal pollution has been emphatically cognizable and the same is the verdict of many investigators (Yilmaz and Yilmaz, 2007). Fish is the food stands at the high level of food chain. So it is possible for accumulation of metals in large quantities, accumulation follows the intake and elimination from and to the body (Karadede *et al*, 2004). Pollution monitoring studies in the coastal regions betrayed that there are higher and elevated levels of metals (Henry *et al*, 2004). Hence, the present study is taken up to find out the level of heavy metal concentrations of Cu, Cr, Zn, Pb and Cd in surface seawater and whole body tissues of finfish and shell fish such as *Rastrelliger kanagurta*, *Lates calcarifer*, *Ephinephalus malabaricus*, *Penaeus indicus* and *Meretrix meretrix* from this creek.

MATERIALS METHOD

Study area

The reference site taken for this study is Kovalam coast, which is located 40 km away from Chennai. Kovalam estuary (12° 49' N, 80° 5' E) is situated on the east coast of India and is about 35 km south of Chennai. It runs parallel to the sea coast and extends to a distance of 20 km. The temperature and salinity of this estuary ranged between 25–28°C and 24–26 ppt, respectively. It was chosen as the unpolluted site for the present investigation as it is surrounded by high vegetation and it is from industrial, agricultural, municipal and sewage pollution.

Sample collection

The fin fish and shell fish were collected in the Muttukadu creek with the help of fisherman by using cast net. After collection, fin fish and shell fishes were immediately transported to the laboratory in an ice box. The specimens were thawed at room temperature and their total length and weight were recorded. The fin fishes of *Rastrelliger kanagurta* (length 11.05±0.83 and weight 25.99±2.06), *Lates calcarifer* (length 11.83±1.20cm and weight 40.58±0.82g), *Ephinephalus malabaricus* (length 9.66±0.42cm and weight 21.39±0.74g) and the shell fishes of *Penaeus indicus* (length 7.91±0.35cm and weight 11.28±0.45g) and *Meretrix meretrix* (length 4.94±0.55cm and weight 7.79±0.55g) was collected from the Muttukadu creek (April 2009 to March 2010). The metal content of the tissue samples was based on dry weight. The marine organisms collected from the field were sacrificed in the field and was stored to -4°C and then transported to the laboratory and stored at -20°C in the deep freezer until analysis.

Sample analysis

The fin fishes and shell fishes were dissected to muscle using stainless steel knife and dried in hot air oven at 70°C for 14 h. A porcelain mortar and pestle were employed to grind and homogenize the dry tissue and weighed to 1g using an electronic weighing balance (Metlor Toledo). The weighed samples were digested in 100 ml glass beaker with concentrated nitric acid (20 ml) overnight. It is then mixed with 10 ml of concentrated nitric acid and perchloric acid (4:1) solution followed by hotplate heating at 120°C up to complete dryness. The residue was then dissolved and diluted with 20 ml of a solution of de-ionised water and conc. Nitric acid (4:1) (v:v) and then this solution is filtered through No. 1 Whatman filter paper. The filtered sample solutions were directly introduced into atomic absorption spectrometry (Perkin Elmer AAnalyst 800) for estimation of the heavy metals. The blanks were carried out in parallel with all analysis and the blank values were lower than 0.5% of the sample signals. Analysis of standards (Merck, Germany) was done for every five samples to maintain the accuracy of analysis. International Reference Standards DORM-2 and DORM-3 was run concurrently along with the samples and the obtained values are reported in Table 1. All acids and chemicals used were of analytical reagent grade. Metal concentrations were calculated in µg g⁻¹ dry weight. Laboratory glass wares were kept overnight in 10% nitric acid solution and rinsed with de-ionised water and air dried before use.

Table 1 Recovery values of heavy metal concentrations (mg kg⁻¹ dry weight) measured in certified reference materials DORM - 2 (Dog fish muscle) and DORM - 3 (Fish protein).

Reference materials	Heavy metals	Certified values (mg kg ⁻¹)	Observed values (mg kg ⁻¹)	% of recovery
DORM -2	Cu	2.34±0.16	2.49±0.09	106.24
	Cr	34.7±5.5	34.04±0.61	98.10
	Zn	25.6±2.3	24.46±0.45	95.54
	Pb	0.065±0.007	0.068±0.001	104.31
	Cd	0.043±0.008	0.042±0.004	97.21
DORM-3	Cu	15.5±0.63	16.20±0.34	104.54
	Cr	1.89±0.17	1.86±0.01	98.20
	Zn	51.3±3.1	49.37±0.38	96.23
	Pb	0.395±0.060	0.385±0.01	97.57
	Cd	0.290±0.020	0.308±0.01	106.28

RESULT

In the present study, Cu, Cr, Zn, Pb and Cd concentrations were determined in the fishes, crustacean and bivalve. The level of heavy metal concentrations in tissue samples, collected from different marine organisms of the Muttukadu creek, south east coast of India. The highest concentration of Cu (9.80±0.36 µg g⁻¹), Zn (12.48±0.47 µg g⁻¹), Pb (7.63±0.33 µg g⁻¹) were determined in *M. meretrix* other metals of Cr (3.62±0.28 µg g⁻¹) in *P. monodon* and Cd (6.41±0.64 µg g⁻¹) in *R. kanagurta*. However the lowest concentration of Cu (2.74±0.23 µg g⁻¹), Zn (4.49±0.27 µg g⁻¹) and Pb (1.62±0.25 µg g⁻¹) observed in *P. monodon* other metals of Cr (1.03±0.41 µg g⁻¹) in *L. calcarifer* and Cd (3.70±0.18 µg g⁻¹) in *M. meretrix*. Overall, the level of heavy metal accumulation of Cu, Cr, Zn and Pb were higher in the shellfish and Cd was in the finfish (Fig-1 and Table-2). The metal accumulation of Cu, Zn, Pb and Cd were lower in the shell fish and Cr was in the finfish collected from the

Muttukadu creek, south east coast of India and the cause of this elevated concentration is known to be the natural origin.

Table 2 Level of heavy metal concentration (mean±SD) of marine organisms in Muttukadu creek ($\mu\text{g g}^{-1}$)

Heavy metal	<i>Rastrelliger kanagurta</i>	<i>Lates calcarifer</i>	<i>Ephinephalus malabaricus</i>	<i>Penaeus indicus</i>	<i>Meretrix meretrix</i>	FAO/WHO
Cu	5.19±0.50	6.84±1.27	3.80±0.35	2.74±0.23	9.80±0.36	10-100
Cr	1.70±0.34	1.03±0.41	1.50±0.27	3.62±0.28	3.62±0.28	50
Zn	10.12±0.67	11.45±0.34	7.42±0.33	4.49±0.27	12.48±0.47	30-100
Pb	4.59±0.33	3.23±0.28	5.51±0.26	1.62±0.25	7.63±0.33	0.05-6
Cd	6.41±0.64	5.76±0.39	4.47±0.29	5.67±0.25	3.70±0.18	0.05-5.5

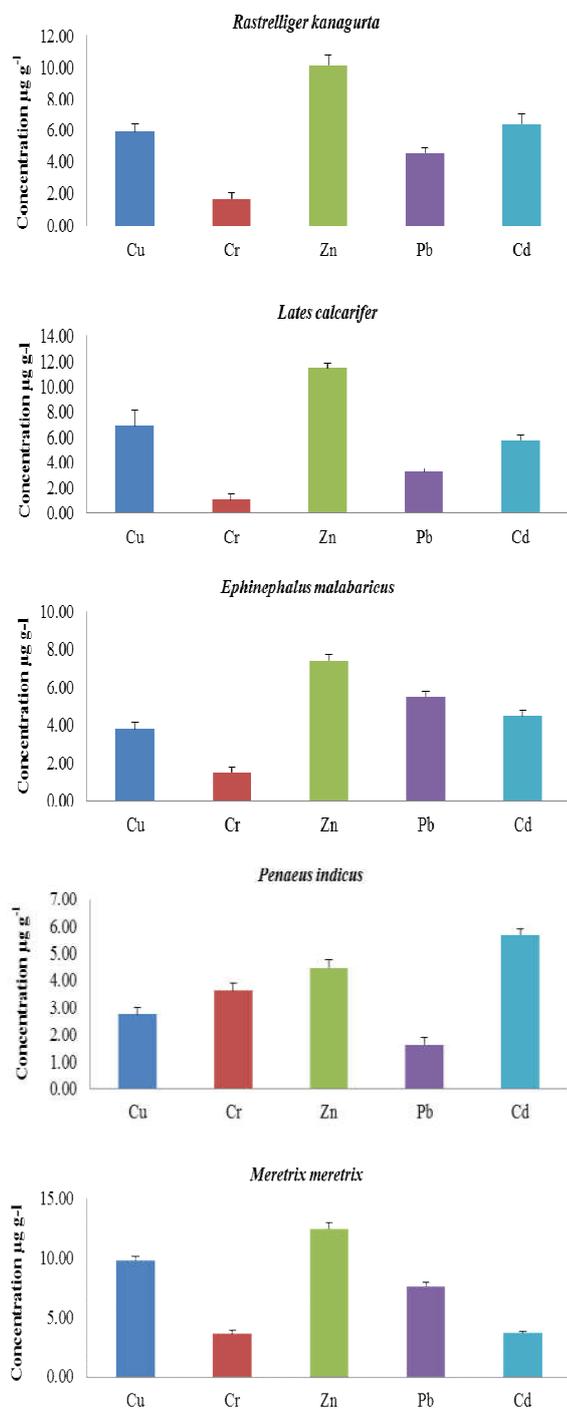


Figure 1 Heavy metal concentration of water and bioaccumulation in marine organisms Muttukadu creek, Kovalam.

DISCUSSION

The accumulation of metals by the fish depends on the location, feeding behaviour, trophic level, age, size; duration of exposure to metals and homeostatic regulation activities of fish [Sankar *et al*, 2006; Kargin, 1996) has listed multiple factors that influence metals accumulation in fish such as season, physical and chemical properties of water. Knowledge of metals concentration in fish is important to management for various purposes such as risk of taking fish as part of diet and metals pollution control strategies. Most of fish are at top in aquatic food chain and have potential to accumulate high metals content even in mild polluted conditions. Therefore, metals concentration in fish could be used as an index to estimate level of pollution especially in aquatic bodies (Karadede-Akin and Unlu, 2007) even in the lake system. Although, fish muscle was reported have lowest metal concentrations compare to bone, gill and liver, this study focus metals in fish muscle since, people eat fish muscle and not others. Various studies on metal concentrations in fish samples has done previously (Fidan *et al*, 2007; Chi *et al*, 2007; Ahmad and Shuhaimi-Othman, 2010; Laar *et al*, 2011). This study found Cu and Zn concentrations were almost as low as from unpolluted lake likes Eber (Fidan *et al*, 2007). However, Sn and Pb concentrations were higher than fishes from Eber Lake and Taihu Lake (Chi *et al*, 2007). Both metals were probably contributed to the lake through various sources, such as agriculture activities likes oil palm and rubber plantations nearby catchment besides mining activities. A large scale of oil palm plantation was developed by Sime Darby Company nearby the catchment and the use of chemical fertilization could introduce Pb, Zn and Cu to the catchment. Excessive fertilizer may be transported to the catchment through surface run off. The catchment has several dredged out small and big ponds that could transport these metals in to the catchment. Previous studies also indicated that different contents of heavy metals in different fish species might be a result of different ecological needs, metabolism and feeding patterns (Ayse, 2003). Romeo *et al*. (Romeo *et al*, 1999) pointed out that Cd, Cu and Zn contents in edible muscles of pelagic fish species were lower than for benthic fish species. Similarly, this study showed that the Cd, Cu and Zn contents in muscles of *H. molitrix* and *A. nobilis* (pelagic fishes) were lower than those in *C. auratus* and *C. carpio* (benthic fishes). The Pb and Cr contents in muscles showed no correlation with fish species.

Heavy metal accumulation could increase in the presence of dissolved organic carbon (Chandrashekar *et al*, 2004). Lead is found to be higher in locations that were located near industrial areas (Praveena *et al*, 2008). Generally, crustaceans accumulate some metals in direct proportion to the increase in the bioavailability from water and food chains (Rainbow *et al*, 1990). The level concentrations of heavy metals (essential and nonessential) were measured in different marine biota including cephalopoda, bivalve, crustacean and fish. Bioaccumulation of heavy metal concentration in *Penaeus merguensis* were observed Zn 2.31 mg g⁻¹, Cu 1.166 mg g⁻¹, Pb 0.022 mg g⁻¹, Cd 0.002 mg g⁻¹ from coastal waters of Gresiko, Indonesia (Soegianto *et al*, 2008). Metal accumulation of *Penaeus monodon* stated that Zn 24.2-35.7 mg g⁻¹, Cu 12.2-21.3 mg g⁻¹, Pb 0.8-1.3 mg g⁻¹, Cd 0.2-0.4 mg g⁻¹ Bangladesh coastal waters (Hossain and Khan, 2001). The level of heavy metal concentration in edible and soft tissues of *Penaeus*

indicus indicated that Zn 19-27 mg g⁻¹, Cu 20-45 mg g⁻¹, Pb 0.03-0.14 mg g⁻¹, Cd 0.04-1.47 mg g⁻¹ in Egypt (Ahdy, 2007). The results presented on metal contents in the examined species of *Macrobrachium felicinum* were observed Zn 2.51 mg g⁻¹, Cu 20.5 mg g⁻¹, Pb 0.350 mg g⁻¹, Cd 0.08 mg g⁻¹ in Taylor creek, Nigeria (Opuene and Agbozu, 2008). Heavy metal concentration of *Metapenaeus ensis* were Zn 15.8 mg g⁻¹, Cu 28.0 mg g⁻¹, Pb 0.135 mg g⁻¹, Cd 0.001 mg g⁻¹ in the pearl river estuary, South China (Ip *et al*, 2005). The heavy metal accumulation of *Penaeus semisulcatus* were Zn 53.7 mg g⁻¹, Cu 32.2 mg g⁻¹, Pb 19.1 mg g⁻¹, Cd 3.47 mg g⁻¹ in Yumurtalik coast of Iskenderum, Gulf of Turkey (Cogun *et al*, 2005), in *Parapenaeus longirostris* were Zn 81-100 mg g⁻¹, Cu 100-125 mg g⁻¹, Pb 0.5-2.6 mg g⁻¹, Cd 0.4-0.9 mg g⁻¹ in North East Algeria (Abdennour *et al*, 2000), in the species of *Penaeus setiferus* Zn 107 mg g⁻¹, Cu 17.3 mg g⁻¹, Pb 7.73 mg g⁻¹, Cd 6.1 mg g⁻¹ in Gulf of Mexico (Vazquez *et al*, 2001). The results reveal that these organisms show more or less the same order of distribution for each of the metals studied. The average concentrations of heavy metals exhibited the following decreasing order: cephalopoda > bivalve > crustacean > fish reported by Ahdy *et al*. [Ahdy *et al*, 2007]. In the present study the *P.indicus* accumulated higher proportions of Cd > Zn > Cr > Cu > Pb in Muttukadu creek.

Molluscs, especially bivalves, can accumulate metals both in soft tissues and shells, numerous studies showing how the chemical composition of the shell may reflect human impact on ecosystems [Vazquez *et al*, 2001]. Although, generally, metals concentrate mainly in the soft tissue, there are also situations in which shells accumulate high levels of metals. Shells may be an indicator of changes in environmental pollution, presenting a lower variability compared with soft tissue and providing a historical record of metal content in the body throughout its life cycle. For mussels (*Mytilus edulis*) from the North Sea and Baltic Sea the following threshold values corresponding to natural background concentrations of heavy metals have been proposed: Cu 2.0 µg g⁻¹ f.w.; Cd 0.4 - 0.8 µg g⁻¹ f.w.; Pb 0.4 - 1.0 µg g⁻¹ f.w.; Ni 0.8 - 1.0 µg g⁻¹ f.w.; Cr 0.4 - 0.6 µg g⁻¹ f.w.; Hg 0.04 µg g⁻¹ f.w.; Zn 24 - 40 µg g⁻¹ f.w. [Vazquez *et al*, 2001]. Investigations on the bioaccumulation of metals in mussels from different marine regions have reported various areas of variation, generally comparable to values observed at the Romanian seaside. In some cases, extreme values in mussels from highly contaminated locations were reported (Shulkin *et al*, 2003). The heavy metal accumulation were observed in the mussels tissue of *Mytilus edulis* Ni 0.18 µg g⁻¹, Pb 0.59 µg g⁻¹, Zn 50.60 µg g⁻¹, Cu 2.32 µg g⁻¹ in Dalian Bay (Liu *et al*, 1995), Cr 2.18 µg g⁻¹, Ni 3.31 µg g⁻¹, Pb 0.94 µg g⁻¹, Zn 104.9 µg g⁻¹, Cu 22.08 µg g⁻¹ in Qingdao Bay (Fung *et al*, 2004), Cr 10.93 µg g⁻¹, Ni 0.25 µg g⁻¹, Pb 0.22 µg g⁻¹, Zn 16.49 µg g⁻¹, Cu 2.52 µg g⁻¹ in Weihai Bay (Liang *et al*, 2004) and Cr 0.19 µg g⁻¹, Ni 0.47 µg g⁻¹, Pb 1.34 µg g⁻¹, Zn 46.22 µg g⁻¹, Cu 10.98 µg g⁻¹ in Yantai Coast of northern yellow sea of China (Zhang *et al*, 2012). In the present study accumulation of metals in bivalve shells from the Muttukadu creek presented variations depending on the heavy metals and the species, generally highest concentrations being observed in Zn and lowest concentration observed in Cr.

In the present study lowest concentration (2.74±0.23) of Cu were observed in *P. monodon* and highest concentration (9.80±0.36) were observed in *M. meretrix*, though the level of

Cu did not exceed the acceptable levels for food samples (FAO, 1983; WHO, 1985). Accumulation of chromium in fish is not a serious problem since the fish are able to eliminate it from the body. Chromium readily penetrates gill membranes by passive diffusion and concentrates at high levels in various organs and tissues, in the present investigation the lowest concentration observed (1.03±0.41 µg g⁻¹) in *L. calcarifer* and highest concentration observed (3.62 ±0.28 µg g⁻¹) in *P. monodon*. The levels of Cr recorded in fish samples were lower than the safe limits in fish food. Zinc, an essential micronutrient for both animals and humans, has been a cofactor for nearly 300 enzymes in the marine organisms. The recommended daily allowance is 10 mg/day in growing children and 15 mg/day for adults (NAS-NRC, 1974). In the present investigation the lowest concentration observed (4.49±0.27 µg g⁻¹) in *P. monodon* and highest concentration observed (12.48±0.47 µg g⁻¹) in *M. meretrix*. The levels of Zn presents in marine organisms were within the permissible limits. Lead is a non-essential toxic metal that can affect humans when ingested or inhaled in high doses. In shell fish and fin fish, it can cause deficits or decreases in survival, growth rates, development and metabolism, in addition to increased mucus formation (Burger *et al*, 2002). In the present study, the minimum accumulated level of lead was (1.62±0.25 µg g⁻¹) in *P. monodon* and maximum were (7.63±0.33 µg g⁻¹) in *M. meretrix*, it is more than the safe limits. Cadmium is a serious contaminant, a highly toxic element, which is transported in the air. The source of Cd in humans is through food consumption. Severe toxic symptoms resulting from Cd ingestion are reported between 10 to 326 mg (Kalay *et al*, 1999). The present investigation the lowest concentration of Cd were (3.70±0.18) in *M. meretrix* and highest were (6.41±0.64) in *R. kanagurta*, its showed higher level of food safe limits (FAO, 1983; WHO, 1985).

CONCLUSION

The present investigation revealed that the wild marine organisms, Cu, Cr and Zn concentrations in fin fishes and shell fishes samples are lower than the standard limit of seafood safety criteria and Pb and Cd concentrations are over the limit. These results indicated that the heavy metal levels in the marine organisms in the studied areas were moderate but unacceptable Pb and Cd from the view of safety of seafood. It may be due to discharge of industrial wastes from the industrial areas are discharged into the coastal water through pipelines, while the municipal wastes of Chennai city are discharged into drains adjoining the Muttukadu creek, which empties into the Chennai coastal areas. Such work may create aquatic pollution, resulting in severe ecosystem modifications and depletion of fishery resources. Furthermore, it is very necessary and important to further study toxicological and ecological effect of Pb and Cd in the Muttukadu creek understand the potential for risk to human and environmental health.

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