Oceanological and Hydrobiological Studies
International Journal of Oceanography and Hydrobiology
Vol. XXXVI, No.4

Institute of Oceanography | (91-103) | University of Gdańsk
ISSN 1730-413X | 2007 | eISSN 1897-3191

DOI 10.2478/v10009-007-0031-4
Original research paper

Received: *March 19, 2007*
Accepted: *September 26, 2007*

A comparative seasonal pollution assessment study on Ennore Estuary with respect to metal accumulation in the grey mullet, *Mugil cephalus*

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Key words: Seasonal variation, Metal accumulation, pollution, environmental contaminants

Abstract

Environmental pollution is currently a major concern due to unorganised and increased industrialization and urban development. In the Tamilnadu state of India, on the Bay of Bengal, lies the Ennore estuary, which is an important industrial centre. The main industries in the region include a fertilizer company, refineries, a rubber factory, paint industry and a thermal power station. Investigations into the seasonal pollution impacts on the distribution of various metals in the waters of the Ennore estuary, situated in North Chennai, Tamilnadu, India, and the concentrations of some of the toxic metals in the tissue of the grey mullet, *Mugil cephalus* L., inhabiting in the same estuary were conducted during the period of April 2004 to March 2006.

The results were compared to those of the Kovalam estuary, which is unpolluted. In the water at one sample site of the Ennore estuary, heavy metal concentrations ranged between: Pb

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0.15-0.23, Hg 0.05-0.06, Cd 0.01-0.03, and Cr 3.33-1.093 ppm for summer and monsoon seasons, and were significantly related to the seasonal rains ($p<0.01$). In contrast, heavy metal concentrations in the Kovalam estuary ranged between: Pb 0.043-0.039, Hg non-detectable–0.001, Cd 0.01-0.098, and Cr 0.063-0.052 ppm, and were not significantly related to the rainy season. The metal concentrations found in the muscle tissues of *Mugil cephalus* L., at the same site on the Ennore estuary were: Fe 11.06 ±0.29, Zn 1.67 ±0.14, Cr 2.98 ±0.15, and Pb 1.26 ±0.11 ppm, which were also significantly related to season ($p<0.01$). The metal concentrations in fish inhabiting the Kovalam estuary were: Fe 2.17 ±0.15, Zn 0.42 ±0.05, Cr 0.63 ±0.04, and Pb 0.31 ±0.04, and showed no significant seasonal difference. The highest metal concentrations found in the tissues of the Ennore estuary fish which lead to the oxidative stress and shorten the lifespan of the fish are postulated to primarily result from anthropogenic activities and effluent discharge from the local industrial activities.

INTRODUCTION

Estuaries are in a state of constant flux and their dynamic nature provides many ecological niches for diverse biota. The health status and biological diversity of Indian estuarine ecosystems are deteriorating day by day through multifarious man-made activities. The dumping of enormous quantities of sewage and industrial effluents into estuaries has resulted in a drastic reduction of shallow water fish populations, increased pollution and ecological imbalance resulting in the large-scale disappearance of numerous flora and fauna (Rajendran et al. 2004).

Heavy metals, as defined by Nieboer and Richardson (1980), are normal constituents of the marine environment. These metals, which normally occur at low concentrations, function in combination with organic molecules, usually proteins. Even at low levels heavy metals are capable of exerting considerable biological effects (Rainbow 1992). All metals are toxic above some threshold bioavailability levels, although Hg, Cu, Cd and Pb are particularly toxic (Bryan 1979). Comparisons of levels of heavy metal pollution in aquatic environments are undertaken by analysis of water, sediments and members of indigenous biota, i.e. biomonitors (Phillips and Rainbow 1993).

It is well known that fish are good indicators of chemical pollution and as a result they have long been used to monitor metal pollution in coastal and marine environments world wide. Fish are widely used as bioindicators of marine pollution by metals (Evans et al. 1993) and their position in the food chain is a useful factor determining mercury contamination (Bernhard 1998).

The main sources of metal pollution in the Ennore estuary are the surrounding industries. Vertical and horizontal distribution of metals in the Ennore estuary are greatly influenced by the hydrodynamic and physiochemical conditions of the estuary. Physicochemical factors are also responsible for physical and chemical partitioning behaviour and speciation of metals within and between different environmental compartments. Consequently, changes in bioavailability of metals may be as important to estuarine organisms as changes in the total metal inputs and their concentrations (Mubiana et al. 2005).

Seasonal variations have a potential to effect metal concentrations in estuarine organisms due to changes in physiochemical variables and some biological factors. Seasonal effects are critical in biomonitoring programmes especially when sampling is undertaken at different times of the year. Zwolsman et al. (1997) showed strong seasonal variations in river flow, dissolved oxygen, pH, temperature, salinity and suspended particulate matter in the Western Scheldt estuary. Consequently seasonal variations in dissolved heavy metals in the estuary were more pronounced than those of the nearby less polluted tidal basin of the eastern estuary (Gerringa et al. 1996, Gerringa et al. 1998). Little is currently known about the influence of seasonal changes on metal content in the Ennore estuary region and the organisms in that area.

The objectives of the current study were:

1. To examine the temporal and spatial distributions of heavy metal concentrations in the grey mullet, *Mugil cephalus,* in the Ennore estuary, in order to evaluate the impact of increased pollution input into the estuary;
2. To determine the influence of season on metal concentrations in fish in the Ennore estuary;
3. To examine relationships among water and fish tissue metal concentrations as they relate to fish diversity, tissue metal accumulation.

MATERIALS AND METHODS

Water samples were collected from April 2004 to March 2006. The study period was divided in to four seasonal groups (based on the North East Monsoon, which brings the maximum amount of rainfall to the east coast of India): summer (April, May and June), pre-monsoon (July, August and September), monsoon (October, November and December) and post-monsoon (January, February and March).

In order to assess whether seasonal variations were related to overall levels of environmental pollution two locations were chosen for sampling, the Ennore estuary (the most polluted site in North Chennai) and the Kovalam estuary (the least polluted site in South Chennai). In the Ennore estuary three sample sites were selected: Station I - the Bar mouth region, Station II - Ennore creek region (about 1.5 km from Station I), and Station III - the left side of the railway bridge. Samples were collected monthly, three times for a season from all sites.

In order to assess metal accumulation (Fe, Zn, Cr, Pb) in the fish, samples of *Mugil cephalus* (of 30-35 cm in length) were collected monthly (based on their availability) during the summer and monsoon seasons from Station III of Ennore estuary and the Kovalam estuary site. 10-15 fish were collected from each site close to the waterline at low tide; and samples were then transported on ice in a cool box to the laboratory. Each individual was dissected with a clean scalpel blade and washed well in mili-Q water. The soft tissues of each individual were placed in a polypropylene vial and dried for 72 hrs at 60°C, after which time the dry weight was determined. Samples were digested through the addition of about 7 ml of concentrated nitric acid and 3 ml of perchloric acid and overnight incubation on a hot plate. After digestion, samples were filtered using Whatman filter paper No. 1, and the filtrate made up to 100 ml with mili-Q water. Metal concentrations in the final solutions were determined using a flame atomic absorption spectrophotometer.

In water samples from both locations, the concentration of metals (Pb, Hg, Cd, Cr, Mn, Fe, Zn, Cu, Ni, K, Na, Ca and Mg) and sulphate were determined with a Perkin Elmer 2380 atomic absorption spectrophotometer (APHA, 1980).

Tissue metal concentrations of *Mugil cephalus* were analysed using the Student's t test (Sokal and Rohlf 1995), and a two way ANOVA was applied to test the effects of season and location on metal concentrations in the estuaries.

RESULTS

The results (Figure 1-3) show that Station III of Ennore estuary had higher concentrations than any other site of all the metals studied.

Water at Ennore Station III of the Ennore estuary had toxic heavy metal concentrations ranging between Pb 0.15-0.23, Hg 0.04-0.06, Cd 0.01-0.03 and Cr 1.1-3.3 ppm in the summer and monsoon seasons, significantly higher ($p<0.01$) than in the Kovalam estuary, where metal concentrations during summer and monsoon seasons ranged between Pb 0.029-0.04, Hg non-detectable–0.001, Cd 0.09-0.1, Cr 0.05-0.06 ppm. No significant seasonal difference in the toxic metal concentrations was seen in the Kovalam estuary samples .When results from the Ennore and Kovalam estuaries were compared a Two Way ANOVA showed a strong effect ($p<0.01$) of site on metal concentration for Station III of the Ennore estuary. Metal concentrations in the Ennore estuary were much higher than in the relatively less polluted Kovalam estuary. In the Station III of Ennore estuary, all the metals studied had concentrations of about twice those in the Kovalam estuary.

Figures 2 and 3 show the seasonal variations of metals (Na, K, Ca, Mg, Fe, Cu, Zn, Mn and Ni) and SO_4 in the waters of the Kovalam and Ennore estuaries. Station III of the Ennore estuary showed highly significant ($p<0.01$) concentrations of all these metals compared to Stations I and II of the Ennore estuary and the Kovalam estuary.

As the metal concentration was significantly related to season at Station III of the Ennore estuary, fish were collected from that site and compared to fish

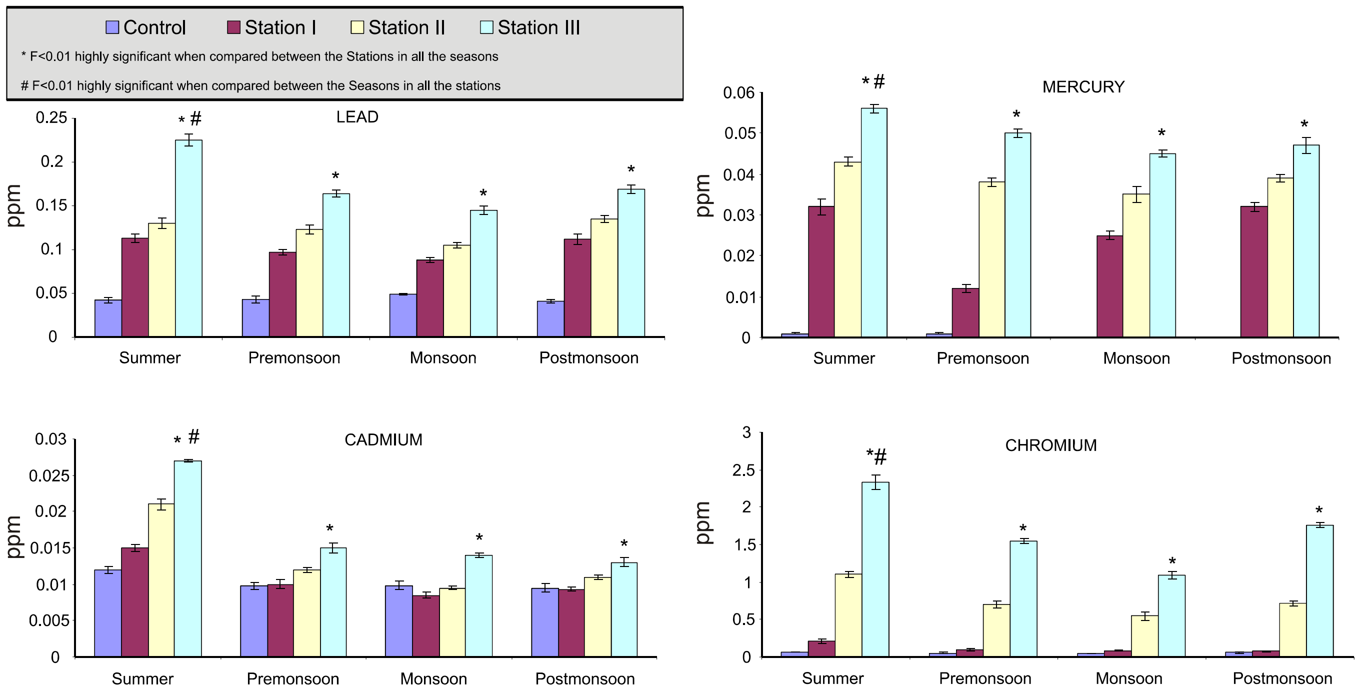

Fig. 1. Seasonal variations of heavy metal (Pb, Hg, Cd, Cr) concentrations in waters of the Kovalam Estuary and three sites of Ennore Estuary.

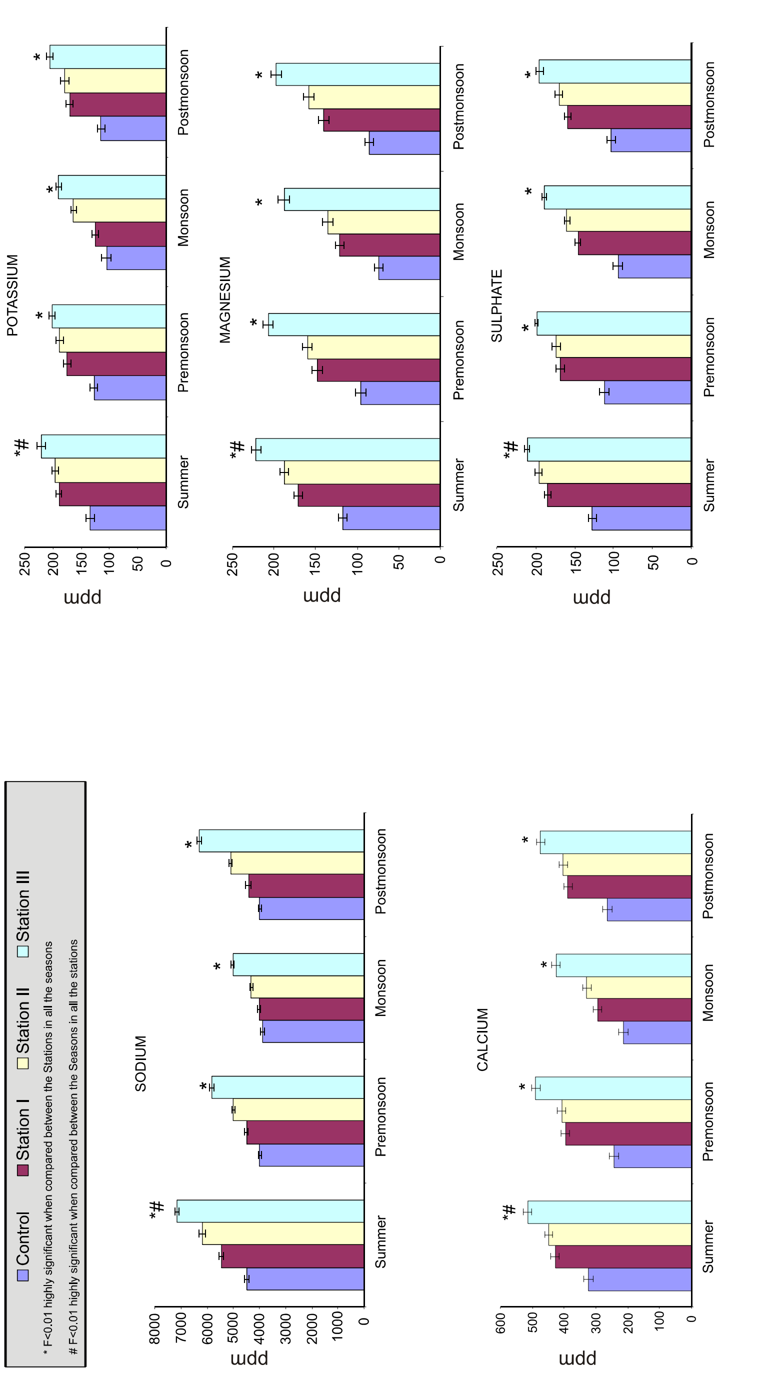

Fig. 2. Seasonal variations of metal (Na, K, Ca, Mg) and SO_4 concentrations in waters of the Kovalam Estuary and three sites of the Ennore Estuary.

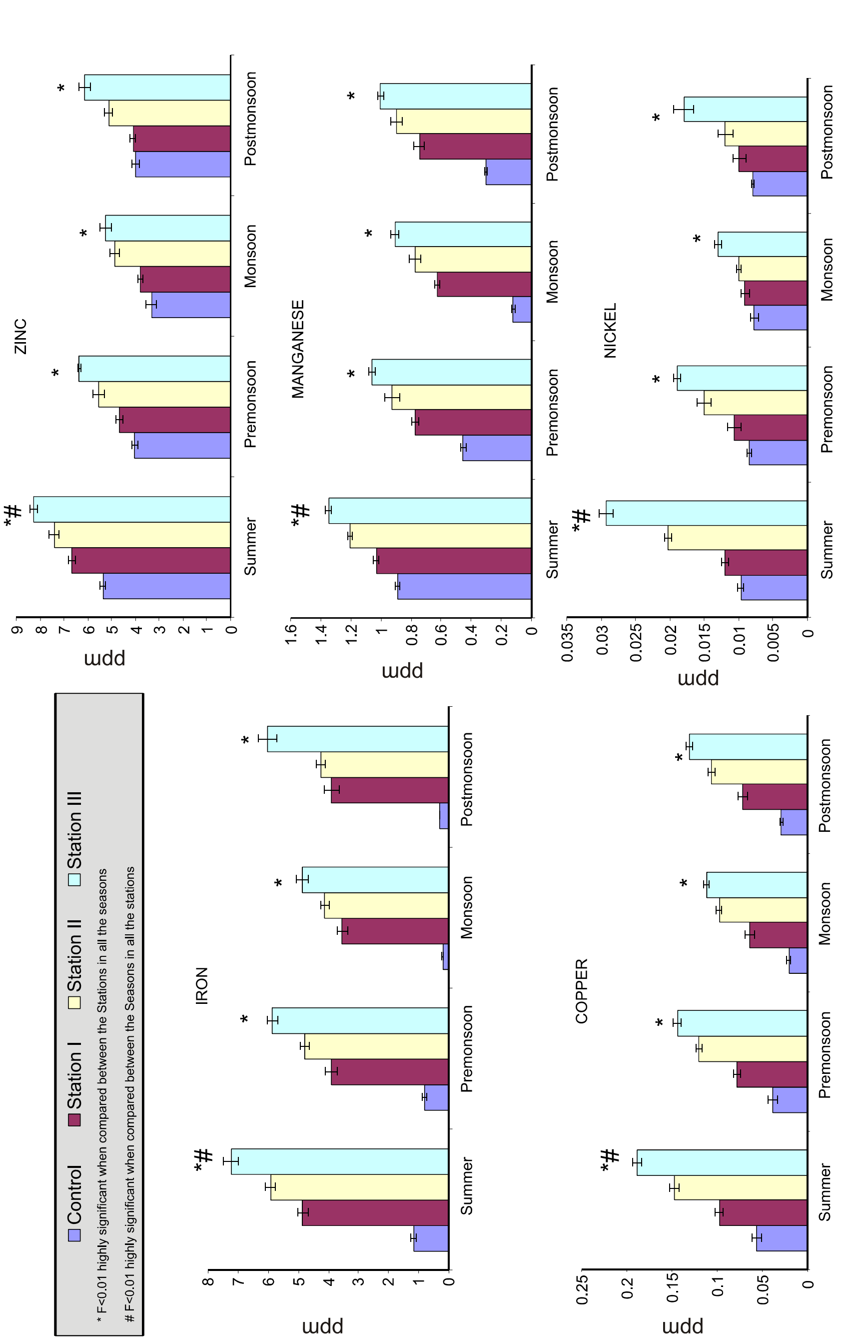

Fig. 3. Seasonal variations of metal (Fe, Cu, Zn, Mn, Ni) concentrations in the waters of the Kovalam Estuary and three sites of the Ennore Estuary.

collected from the Kovalam estuary for tissue heavy metal concentration studies. Figure 4 shows the metal concentrations (Fe, Zn, Cr and Pb) in the muscle tissue of *Mugil cephalus* collected from Station III of the Ennore estuary and the Kovalam estuary during the summer and monsoon seasons.

The concentrations of the heavy metals found in the muscle tissue of *Mugil cephalus* collected at Station III of the Ennore estuary were Fe 11.064 ±0.29, Zn 1.67 ±0.14, Cr 2.978 ±0.15, and Pb 1.258 ±0.11 ppm. These values are significantly higher ($p<0.01$) than those seen in the samples from the Kovalam estuary (Fe 2.165 ±0.15, Zn 0.424 ±0.05, Cr 0.625 ±0.04, and Pb 0.307 ±0.04).

Although seasonal effects were seen on the metal concentrations in the water of the Ennore and Kovalam estuaries, the fish tissue metal concentrations do not show much of a seasonal effect, however the metal concentrations in the water samples and the fish tissues collected from Station III of Ennore estuary were much higher than those from the Kovalam estuary.

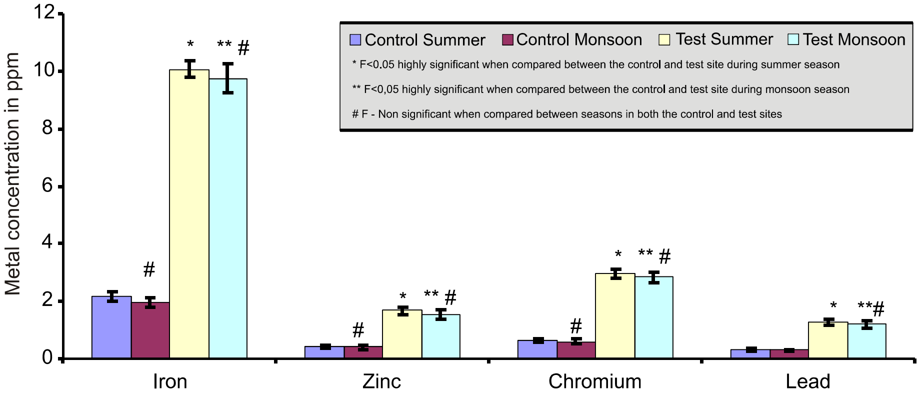

Fig. 4. Concentrations of toxic metals (Fe, Zn, Cr and Pb) in fish muscle tissue from the Kovalam Estuary and Station III of the Ennore Estuary during summer and monsoon seasons.

DISCUSSION

Estuarine environments are vulnerable to stress and if a key species disappears due to heavy metal pollution no other species can replace it. A vast number of chemicals, as well as large amount of nutrients, are released into the environment daily and transported via rivers and lakes into estuarine and marine environments. Hence there is need to monitor environmental changes, with quantitative laboratory and field investigations integrated to link ecological metrics with metal body burden in sentinel aquatic organisms.

Heavy metal contamination of the environment is recognised as a serious pollution problem. Variability in metal concentrations of marine organism depends on many factors, both environmental and purely biological (Phillips 1995). Fish are widely used as sentinels of contamination as sufficient background knowledge enables the use of certain species as bioindicators of heavy metal pollution (Pastor et al. 1994, 1996). In particular, mercury has received much attention due to its well known toxic effects. Metal analyses of water samples and fish tissue have revealed that metal accumulation is inversely proportional to ecosystem changes (Birge et al. 2000).

The results presented here indicate that metal distribution in the waters of the Kovalam estuary are around the normal range, as prescribed by ISI (Anon 1986). However, metal concentrations in the waters of the Ennore estuary were found to be very much higher than the normal range as prescribed by ISI, and were significantly higher than those of the Kovalam estuary. This may be attributed to the discharge of effluents from industries situated in the vicinity of the Ennore estuary.

Samples from Station III at Ennore estuary showed significantly higher levels of metal pollution compared to the other sample sites at that location. This may result from its location making it the first point receiving anthropogenic inputs from the Buckingham canal and the Kotralayar River.

In the Ennore estuary, the concentrations of metals were observed to be significantly higher during summer than during the monsoon. These seasonal low values may be attributed to fresh water input following rain as well as due to the release of surplus water from the Poondi reservoir in to the sea via Ennore creek, while the higher values in summer are due to evaporation raising the metal concentrations (Murthy and Rao 1987). In an earlier study, lower metal concentrations were observed during winter and higher concentrations during the summer season (Caccio and Millero 2003).

The effect of season on metal concentrations showed similar patterns in both the Ennore and Kovalam estuaries, with high values recorded during summer. However, there were also some interactions between the site and the season (Two way ANOVA) meaning that although the patterns were similar, the strength of the influence of season was different in the two systems. This is demonstrated by the metal concentrations, which showed a greater difference between monsoon and summer values in the Ennore estuary than in the Kovalam estuary. The influence of seasonal changes on the metal concentrations of the estuaries can also be explained in terms of changes in river flow and changes in the geochemistry of the dissolved metals. In the Western Scheldt estuary chemical speciation studies have shown that the distribution of heavy metals is strongly influenced by salinity, dissolved organic carbon and dissolved oxygen (Van Eck and De Rooij 1993, Paucot and Wollast 1997, Zwolsman et al. 1997, Gerringa et al. 1998).

In the summer seasons, when the fresh water inflow in the estuary is largely anoxic, dissolved metal concentrations tend to be very low and the metal partitioning in those conditions favours adsorption to suspended particles and the sediments (Mubiana et al. 2005). Generally increased dissolved oxygen level lead to the oxidation of the bottom sediments, which when resuspended bring metals in to the water column causing secondary pollution. However, the seasonal trends in the more polluted Ennore estuary being very similar to those in the Kovalam estuary suggest that the seasonal changes in metal concentrations observed in fish are probably due to changes in the water quality influencing levels of metal exposure. Earlier reports suggest that metal accumulation can be increased in the presence of dissolved organic carbon (Penttinen et al. 1995, Stuijfzand et al. 1999, Winch et al. 2002, Guo et al. 2001).

Many anthropogenic sources were reported to contribute to heavy metal pollution in Kolleru Lake and the bioaccumulation of heavy metals in fish helped in assessing the aquatic pollution. Large fractions of Zn, Cd, and Cu were associated with the mobile fraction of the sediment and showed greater bioaccumulation in fish inhabiting in the lake (Chandra Sekhar et al. 2004).

The high metal concentrations in the tissues of fish inhabiting the Ennore estuary are probably related to a high influx of metals as a result of pollution from the surrounding industries thereby increased bioavailability to the fish.

Nammalwar (1992) reported that the concentrations of Hg, Cd, Cu, Zn, Ni, Pb and Fe in various tissues of *Liza macrolepis* inhabiting the Ennore estuary were found to be above the permissible safe levels. Padmini and Kavitha (2005 a) reported that the brain tissue of *Mugil cephalus* inhabiting the Ennore estuary is subjected to severe stress as it is surviving in highly contaminated conditions.

Our previous studies have indicated that the *Mugil cephalus* in the Ennore estuary is subjected to cytogenetic damage (Padmini et al. 2006). It has been reported that there is a gender specific interaction between Se and Cu uptake that may contribute to decreased female reproductive condition in wild yellow perch (Pyle et al. 2005).

Rajathy and Azariah (1996) reported that the levels of Fe, Zn, Mn and Cu in water and sediment samples showed seasonal fluctuations in the Ennore estuary. Measurement of trace metals in *Mugil cephalus* at contaminated sites shows that this fish accumulates metals in response to contamination. This contamination may cause oxidative stress in these fish, which in turn can lead to decreased reproduction, susceptibility to infection and sudden death of fish in large numbers (Padmini et al. 2004, Padmini and Sudha 2004). It has been reported that *Mugil cephalus* surviving in the polluted Ennore estuary are subjected to severe oxidative stress causing considerable DNA fragmentation, potentially leading to cell death (Padmini and Kavitha 2005b).

In conclusion, regardless of seasonal variations in metal concentrations in the Ennore estuary, an accumulation of metals in fish is observed which may be explained in terms of long term overloading the water with pollutants leading to increased environmental contamination. This investigation is aimed at revealing differences in the accumulation pattern of heavy metals in fish inhabiting sediments that are characterized by varying metal bioavailability. We suggest that effluent discharge should be minimal during the summer season in order to minimise the negative impacts of these pollutants.

For the better management of the estuary, detailed metal speciation studies should be considered alongside biomonitoring programmes in order to assess the potential risk of the metal pollution in this fragile ecosystem.

ACKNOWLEDGEMENTS

This study is a part of the DST Project Ref. No DST: SP/SO/AS-10/2003 funded by the Ministry of Science and Technology, Government of India.

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