

## Bioaccumulation of heavy metals in flying fishes along southeast coast of India

\*Jayaprabha, N., Balakrishnan, S., Purusothaman, S., Indira, K., Srinivasan, M. and Anantharaman, P.

Centre of Advanced Study in Marine Biology, Faculty of Marine Sciences, Annamalai University, Parangipettai - 608 502, Tamil Nadu, India

### Article history

Received: 30 September 2013

Received in revised form:

8 January 2014

Accepted: 9 January 2014

### Keywords

Bioaccumulation

Metals

Flying fish

ICP-OES

Southeast coast of India

### Abstract

Bioaccumulation of metals (iron, zinc, copper, lead and cadmium) in the muscles of flying fishes, *Hirundichthys coromandelensis* and *Cypselurus spilopterus* collected from the landing centres of Pondicherry and Cuddalore, Southeast coast of India was studied. Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES) was used for the assessment of present condition of the coastal ecosystem and compiling the baseline data for future monitoring, with respect to metal accumulation of different species. In the present study, tissue samples of fishes revealed that the metals have different levels of accumulation viz. Fe ( $7.80 \pm 2.26 \mu\text{g g}^{-1}$ ); Zn ( $3.30 \pm 2.55 \mu\text{g g}^{-1}$ ); Cu ( $0.33 \pm 0.06 \mu\text{g g}^{-1}$ ), Pb ( $0.24 \pm 0.10 \mu\text{g g}^{-1}$ ) and Cd ( $0.05 \pm 0.02 \mu\text{g g}^{-1}$ ). The concentration of heavy metals was in order of, Fe > Zn > Cu > Pb > Cd. The result shows that the metal accumulation in tissues of both the fishes indicates a similar variation between two stations. The concentration of metals was more in Cuddalore than in Pondicherry. Concentrations of toxic metals such as Pb and Cd were well below the permissible limits proposed by the World Health Organization. Cd showed the lowest concentrations in all fish samples followed by Pb.

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### Introduction

Metals are non-biodegradable and are considered as major environmental pollutants causing cytotoxic, carcinogenic and mutagenic effects in animals (More *et al.*, 2003). The pollution of the aquatic environment with heavy metals has become a worldwide problem during recent years, because they are indestructible and most of them have toxic effects on organisms (Mac Farlane and Burchett, 2000). Among the environmental pollutants, metals are of particular concern due to their potential toxic effects and ability to bioaccumulate in aquatic ecosystems (Censi *et al.*, 2006). Metals especially in coastal environment is common and prevalence due to the fast development of industries, urbanization and human population. Heavy metal pollution has gained serious attention from public and scientific community because of their toxicity to aquatic organisms and ultimate effect on human. There are essential and non essential metals, but when their limits exceed in the body of organism, they become more toxic (Thiyagarajan *et al.*, 2012). Heavy metals including both essential and nonessential elements have a particular significance in exotoxicology, since they are highly persistent and all have the potential to be toxic to living organisms (Storelli *et al.*, 2005). Heavy metals like iron, copper and zinc are essential for regulation of metabolic

activities but become toxic at higher concentrations, whereas, lead and cadmium have no documented role in living organism (Canli and Atli, 2003).

Heavy metals can be accumulated by marine organisms through a variety of pathways, including respiration, adsorption and ingestion (Zhou *et al.*, 2001; Turkmen *et al.*, 2008). Seafood especially marine fish are vulnerable to the effects of chemical contaminants including heavy metals which bioaccumulate and biomagnifies along the aquatic food chain (Kennish, 1998; Agusa *et al.*, 2007). These toxic elemental contaminants cause unhealthy effects to the fish and are transferred into human metabolism through consumption of contaminated fish that leads to serious deterioration of human health status (Virtanen, 2007; Raja *et al.*, 2009; Alinnor and Obiji, 2010). Fish are the major part of the human diet because of having low risk of coronary heart disease, hypertension and cancer. Fishes were considered as better specimen for the investigation of pollutant loads than the water samples because of the significant levels of metals they bioaccumulate. Fish are often at the top of aquatic food chain and may concentrate large amounts of some metals from the water (Mansour and Sidky, 2002). Thus, fish not only indicates the pollution status of aquatic ecosystem but have significant impact on the food web (Qiao-qiao *et al.*, 2007). Consumption of heavy metal

\*Corresponding author.

Email: [prabhamb12@gmail.com](mailto:prabhamb12@gmail.com)

Tel: +91 7639761445

contaminated fish can result in hazardous effects on human health. Fishes have been widely used as bio-indicators of pollution by metals. Muscle tissue of fish is most frequently used for analysis because it is a major target tissue for metal storage and is the main edible part of the fish (Bhupander Kumar *et al.*, 2011). Comparative study regarding the metal accumulation in fishes along Southeast coast region of India is limited. Hence, the present study is taken up to find out the metal concentrations (Fe, Zn, Cu, Pb and Cd) from two species of flying fishes collected from Pondicherry and Cuddalore, Southeast coast of India.

## Materials and Methods

Flying fish *C. spilopterus* (240 mm - 280 mm) and *H. coromandelensis* (160 mm - 220 mm) were collected from Pondicherry (Lat 11°46'03"N; Long 79°49'45"E) and Cuddalore (Lat 11°42' N; Long 74° 46'E) landing centres, along the Southeast coast of India (Fig. 1) and 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 muscles (below the dorsal fin) were removed and analyzed for the concentration of iron, copper, zinc, lead and cadmium. Metal concentrations from the fish tissues were determined by following the method of Dhaneesh *et al.* (2012a, b). Muscle tissues were dissected 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 1 g ( $\pm 0.01$  g) using an electronic weighing balance (Denver, USA). 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. Concentrations of metals were determined by using Inductively Coupled Plasma Optical Emission Spectrometer (Perkin Elmer, Optima 2100DV).

Pearson correlation coefficient was employed for the better understanding of relationship between the concentrations of various metals using statistical package of SPSS 16.0 for windows. One way ANOVA was employed to understand the variation in the concentration of metals with respect to different species. Group linkage clustering technique was used for studying the similarities between metals at each

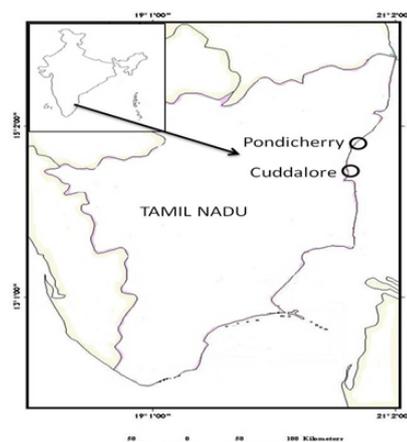


Figure 1. Image showing the study area station using statistical package of Primer 6.

## Results

In the present study, concentrations of Iron, Zinc, Copper, Lead and Cadmium ( $\mu\text{g g}^{-1}$  d.w.) were determined in the tissues of *H. coromandelensis* and *C. spilopterus* collected from Pondicherry and Cuddalore landing centers along Southeast coast of India. Concentration of iron, zinc, copper, cadmium and lead in muscle tissue of these fishes were presented in Table 1. Metal concentrations and the corresponding mean standard deviations (expressed as  $\mu\text{g g}^{-1}$  dry weight) were measured in tissues for both fishes (Figs. 2 & 3). The order of metal accumulation in fishes was as: Fe > Zn > Cu > Pb > Cd. The level of metals in the flying fishes are in the range of, Fe ( $7.80 \pm 2.26 \mu\text{g g}^{-1}$ ); Zn ( $3.30 \pm 2.55 \mu\text{g g}^{-1}$ ); Cu ( $0.33 \pm 0.06 \mu\text{g g}^{-1}$ ); Pb ( $0.24 \pm 0.10 \mu\text{g g}^{-1}$ ) and Cd ( $0.05 \pm 0.02 \mu\text{g g}^{-1}$ ). There were significant differences between the concentration of Fe, Zn, Cu, Pb and Cd in fishes from different stations. Among the two areas studied, Cuddalore showed higher concentration of Fe ( $7.80 \pm 0.81$ ), Zn ( $3.30 \pm 1.98$ ), Cu ( $0.33 \pm 0.06$ ), Pb ( $0.24 \pm 0.01$ ) and Cd ( $0.05 \pm 0.02$ ) than Pondicherry. Among the two fish species, Iron and lead was found to be higher in *H. coromandelensis* whereas zinc, copper and cadmium was higher in *C. spilopterus* (Table 1).

According to Pearson Correlation Coefficient, the metals were significantly correlated to each other at each sampling area at 0.05 levels. One way ANOVA revealed that the concentration of metals was significantly varied with respect to two species ( $p < 0.05$ ). Dendrograms are drawn to study the similarities between metals at each station using group linkage clustering technique based on Bray Curtis coefficient of similarity. Cd and Cu grouped at the highest level of similarity (85.93%) at Pondicherry and Cuddalore. Cd, Cu and Pb at Pondicherry and Cuddalore, were successively grouped at 100%

Table 1. Table showing the comparative level of concentration of heavy metals in flying fishes in Cuddalore and Pondicherry

Species	N	Length (cm)	Total weight (g)	Concentration of metals ( $\mu\text{g g}^{-1}$ )									
				Pondicherry					Cuddalore				
				Fe	Zn	Cu	Pb	Cd	Fe	Zn	Cu	Pb	Cd
<i>H. coromandelensis</i>	30	16-22	50-120	5.41 ± 1.48	3.00 ± 0.32	0.26 ± 0.05	0.20 ± 0.05	0.02 ± 0.00	7.80 ± 0.81	3.28 ± 2.55	0.28 ± 0.05	0.24 ± 0.01	0.02 ± 0.00
<i>C. spilopterus</i>	30	24-28	160-210	5.07 ± 2.26	2.15 ± 1.01	0.26 ± 0.052	0.19 ± 0.10	0.02 ± 0.00	5.88 ± 1.35	3.30 ± 1.98	0.33 ± 0.06	0.17 ± 0.05	0.05 ± 0.02

Number of specimens (n), average length and total wet weight (Mean ± SD)

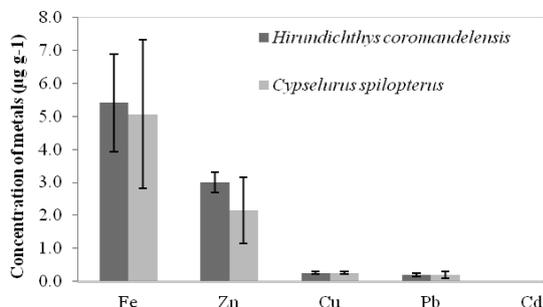


Figure 2. Comparison of mean concentrations ( $\pm$  SD) of Fe, Zn, Cu, Pb and Cd ( $\mu\text{g g}^{-1}$  dry wt) between tissues of fishes collected from Pondicherry. Bars represent mean and vertical lines the SD.

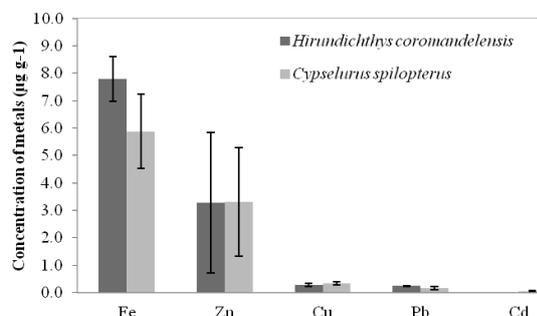


Figure 3. Comparison of mean concentrations ( $\pm$  SD) of Fe, Zn, Cu, Pb and Cd ( $\mu\text{g g}^{-1}$  dry wt) between tissues of fishes collected from Cuddalore. Bars represent mean and vertical lines the SD.

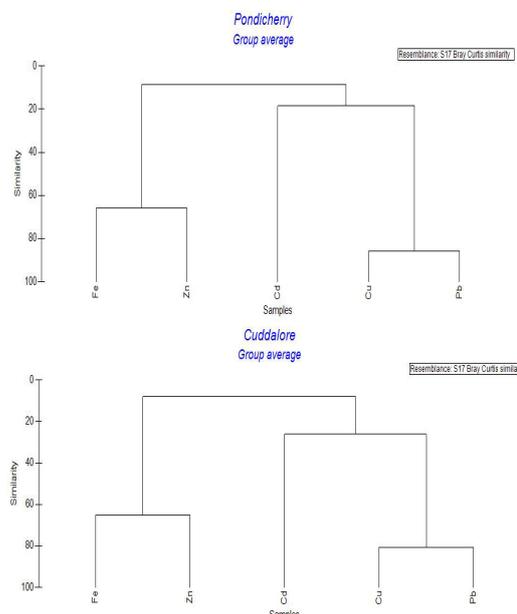


Figure 4. Bray Curtis similarity index versus trace metals in Pondicherry and cuddalore

level similarity. Further at Pondicherry, cluster of Fe and Zn were formed at the next level of similarity (65.88%) and at Cuddalore, cluster of Fe and Zn were successively grouped at next level of similarity (64.98%). Followed by this, Zn and Pb were grouped between 18.28% and 25.91% similarity at Pondicherry and Cuddalore (Fig. 4).

**Discussion**

The study of heavy metal concentrations in fishes was important with respect to human consumption of fish. Several studies shows heavy metal concentrations in tissue of coastal fishes may vary considerably among the different species. This was possibly due to differences in metabolism and feeding patterns of the fishes. Fish is among the dominant bio indicator species used for acute toxicity assay of pollutants such as heavy metals since much attention has been drawn due to the wide occurrence of metal pollution in aquatic system. The rapid

development of industries and agricultures have promoted the increase of environmental pollution although heavy metals in aquatic system can be naturally produced by slow leaching from rocks and soil into water which occur at low levels. Cd and Pb are among the aquatic metal pollutants which usually present at significant level in water system which may pose high toxicities on the aquatic organisms (Zhou *et al.*, 2008). Fe, Zn and Pb levels in the fishes collected from Cuddalore was higher than the levels reported by Thiyagarajan *et al.* (2012),  $0.24 \pm 0.42$ ,  $1.83 \pm 0.44$  and  $0.07 \pm 0.01$  respectively, but Cd and Cu levels are lower than the reported level ( $0.35 \pm 0.71$  and  $3.26 \pm 0.87$ ). In Pondicherry, level of metal concentration was higher than the levels reported by Thiyagarajan *et al.* (2012), Cd ( $0.02 \pm 0.01$ ), Cu ( $0.03 \pm 0.0$ ), Fe ( $1.14 \pm 0.54$ ), Pb ( $0.12 \pm 0.12$ ) and Zn ( $2.92 \pm 0.49$ ). According to Vijayakumar *et al.* (2011), the metal accumulation of Cu, Cd and Zn was  $0.42 \pm 0.09$ ,  $0.35 \pm 0.06$  and  $20.1 \pm 0.13$ , respectively in *Rastrelliger kanagurta*,  $0.43 \pm 0.12$ ,  $0.43 \pm 0.18$  and  $25.4 \pm 0.09$ , respectively in *Kathala axillaris* and  $0.61 \pm 0.15$ ,  $0.42 \pm 0.11$  and  $26.2 \pm 0.16$ , respectively in *Sardinella longiceps*. These levels were higher than the metal level present in the flying fishes.

This investigation showed that different fish species contained different metal levels in their

tissues. The levels of metals found in tissues of the *C. spilopterus* were generally higher than those found in *H. coromandelensis*. This may be due to variations in feeding habits, habitats, size, age and behavior of these species. A number of studies have shown that various factors such as season (Kargin, 1996), length and weight, physical and chemical status of water (Jeziarska and Witeska, 2001) can play a role in the tissue accumulation of metals. The observed variability of heavy metal levels in different species depends on feeding habits (Romeoa *et al.*, 1999), ecological needs, metabolism (Canli and Furness, 1993), age, size and length of the fish (Linde *et al.*, 1998) and their habitats (Canli and Atli, 2003). Bioaccumulation of metal within an organism results from interactions between physiological factors (growth, weight loss, absorption and accumulation), chemical factors (metal concentration, speciation and bioavailability) and environmental factors (temperature and food concentration) (Casas *et al.*, 2006). Species differences in heavy metal bioaccumulation could be ascribed to differences in feeding habits and behaviour of the species (Kotze *et al.*, 1999; Ibrahim *et al.*, 1999 a,b; Mormede and Davies, 2001; Coetzee *et al.*, 2002; Canli and Atli, 2003; Ali and Abdel-Satar, 2005; Canbek *et al.*, 2007). The mean concentrations of heavy metals analysed in the muscle of the flying fishes were well below the maximum permitted concentrations proposed by FAO (1983). Food and Agricultural Organization limits, 0.5 mg/kg for Cd and Pb, 30 mg/kg for Cu and Zn (FAO, 1983 and for Fe 100 µg/g (WHO, 1989). For an average adult (60 kg body weight), the provisional tolerable daily intake (PTDI) for Pb, Fe, Cu and Zn are 214 µg, 48, 3 and 60 mg, respectively (Joint FAO/WHO 2000).

Iron was found to be most abundant metal in muscle tissue of both species studied. Fish is the major source for iron in adults and children and deficiency of it causes anemia. The concentration of Fe in fish muscles from this study varies between  $7.80 \pm 2.26 \mu\text{g g}^{-1}$ . Zinc, an essential micronutrient for both animals and humans, has been a cofactor for nearly 300 enzymes in all marine organisms. As a constituent of many enzymes, Zn is responsible for certain biological functions, for which a relatively high level is required to maintain them (Heath, 2000). A deficiency of zinc is marked by retarded growth, loss of taste and hypogonadism, leading to decreased fertility. In the present study, Zn concentration was higher in fish collected from Cuddalore ( $3.28 \pm 1.98 \mu\text{g g}^{-1}$ ) than in Pondicherry ( $3.00 \pm 0.32 \mu\text{g g}^{-1}$ ), but it is below the maximum permissible limit. Although zinc is an important dietary supplement for humans,

excessive zinc intake can cause a broad spectrum of physiological problems (Fosmire, 1990). Excessive intakes of zinc occur only with the inappropriate intake of supplements (Solai *et al.*, 2010). As copper is an essential part of several enzymes and necessary for the synthesis of haemoglobin, most marine organisms have evolved mechanisms to regulate concentrations of this metal in their tissues. High concentration of Cu was in Cuddalore ( $0.28 \pm 0.06 \mu\text{g g}^{-1}$ ). The level of copper in both the fishes was within the permissible limits. The copper contents in the samples were much less than the FAO-permitted level of 30 µg/g (FAO, 1983). Excessive intake of copper may lead to liver cirrhosis, dermatitis and neurological disorders (Fairweather-Tait, 1988).

Cadmium is a serious contaminant, a highly toxic element transported in the air. The proposed limit values for human consumption of fish reach approximately 0.5 mg/kg (FAO, 1983). In the present study, the highest value of cadmium was documented from Cuddalore ( $0.05 \pm 0.02 \mu\text{g g}^{-1}$ ) and it was below the permissible limit. Cadmium injures kidneys and cause symptoms of chronic toxicity, including impairment of kidney function, poor reproductive capacity, hypertension, tumours and hepatic dysfunction (Waalkes, 2000). Although Cd is a toxic element that would deposit in human body and is danger to human health (Pourang, 1995). Lead is a non-essential toxic metal that can affect humans when ingested or inhaled in high doses. In 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 maximum level of lead was  $0.24 \pm 0.01 \mu\text{g g}^{-1}$ . It is well below the maximum permissible limit of FAO, 1983 (0.5 mg/kg).

## Conclusion

The present study indicated that most of the metal concentrations found in the muscles of these two fish species were below the prescribed limit values for human consumption. Further investigation is recommended on spatial and seasonal variability of essential and toxic metals in other consumable fish species for safeguarding human health. Among the two areas selected for study, Cuddalore was highly polluted with Fe, Zn, Cu, Pb and Cd. This may be due to the industrial activities and anthropogenic introduction of pollution also resulted from local people. This study has enlightened the fact the persistent pollutants like metals should be regularly monitored and any variation from the normal distributional pattern can furnish an idea about the

proper management of the coastal area. A competent monitoring programme is an essential adjunct to any attempt of managing the coastal areas in an ecologically sound and sustainable manner.

### Acknowledgment

The first author would like to thank the DST-INSPIRE programme for their financial support.

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