Heavy metal levels in two food fish species from Negombo estuary, Sri Lanka: Relationships with the body size

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Abstract

The present study was carried out to assess the levels of eight metals viz. Pb, Cu, Cd, Ni, Cr, Mn, Hg and Zn in two species of food fish, Etroplus suratensis and Ambassis commersoni from Negombo estuary. The levels of Hg were analyzed by cold vapour atomic absorption spectrometry whereas the other metals were analyzed by flame/graphite furnace atomic absorption spectrometry. Total metal levels in muscle, gill and liver tissues of the fish showed a wide range. Accumulation of most metals in the tissues of the fish followed the increasing order, muscle < gills≤ liver. Of the different metal levels detected in the fish, only the level of Cd in liver tissue of 6% of E. suratensis and 12% of A. commersoni exceeded the level specified for human consumption by European Union. Interspecies comparisons revealed that mean level of Zn in the muscle tissue of *A. commersoni* was two fold higher than that of E. suratensis. The levels of metals in the muscle tissue of both fish species were positively correlated with the body weight or body length of the fish. Based on the levels of Hg, Pb, and Cd in the fish, edible muscle of both fish species collected from the estuary was found to be safe for human consumption. However, heavy consumption of bigger size fishmay pose a health risk to the consumers as there were strong positive correlations between body weight or body length and metal levels in muscle tissues of both species of fish.

Introduction

Metals are natural components of the aquatic ecosystems. Heavy metals such as copper and zinc are essential for life whereas some metals including mercury, lead and cadmium are biologically non-essential metals which can be toxic to biota even at very low levels. High concentrations of some essential trace metals may become toxic at concentrations which exceed the required limits (Wright and Welbourn 2002). The metal levels in many aquatic ecosystems have been increased due to anthropogenic activities which raises the concerns on metal bioaccumulation and related human health hazards. Hence there is a growing need for monitoring heavy metal pollution and taking necessary steps to manage and remediate ecological damage.

Fish are useful as sentinel species and bioindicators of metal pollution because they can help to understand the risk to the aquatic ecosystem and to humans (Peakall and Burger 2003). Many of the heavy metals are concentrating at different levels in different organs of the fish body. Moreover, biological factors of the fish such as age and body size could play a role in the bioavailability of metals (Peakall and Burger 2003). The use of wild and cultured food fishes as biomonitors of metal pollution in aquatic ecosystems is becoming popular throughout the world (Norrgren et al. 2000; Mansour and Sidky 2002; Bervoets and Blust 2003; Marcovecchio 2004; Begum et al. 2005; Demirak et al. 2005; Storelli et al. 2006; Agarwal et al. 2007; Ploetz et al. 2007). However, only a few studies have been carried out to monitor metal pollution in food fish species in Sri Lanka. It is important to determine the heavy metal levels in commercial food fish in order to evaluate the possible risk of fish consumption.

Negombo estuary is one of the most productive brackish water bodies in Sri Lanka (Silva 1996). The estuary is a very significant fishery centre. It also serves as a shrimp nursery and nutrient source for the coastal sea. In addition, the estuary serves as an anchorage for about 15% of Sri Lanka's marine fishing fleet including multiday boats and fiberglass boats powered by outboard motors. The estuary has been indiscriminately exploited for commercial, residential and industrial development and as a sink for industrial and domestic waste (CEA 1994). With the rapid industrialization and urbanization in the area, the estuary is becoming polluted with various hazardous chemicals including heavy metals. However, less attention has been given to study heavy metal pollution in Negombo estuary. Most of the people in the Negombo area fulfill their protein requirements using fishery resources in the estuary. However, no baseline data are available with respect to the levels of heavy metals in fishery resources in the estuary Etroplus suratensis (Pearl spot, Iri Koraliya) and Ambassis commersoni (Common glass fish, Katilla) are two resident fish species in the Negombo estuary E. suratensis is considered as an omnivorous species where as *A. commersoni* is a carnivorous fish (Pethiyagoda 1991). They are consumed by the people in the area as a protein source. The objectives of the present study were (i) to determine the total levels of eight metals viz. Pb, Cu, Cd, Cr, Ni, Mn, Hg and Zn in the

muscle, gills and liver of *E. suratensis* and *A. commersoni* inhabiting the estuary and to assess whether edible parts are safe for human consumption (ii) to evaluate relationships (if any) between the accumulated metal levels in the edible tissues and body size of the fish. In addition, levels of the metals in the abiotic components of the estuary viz. water and sediments were determined in order to identify sources of heavy metals.

Materials and Methods

Sampling and processing of water and sediments for metal analysis

Water and sediment were sampled from twelve sampling locations (Figure 1) representing north, east, west and south regions of the estuary in 2003. Immediately after, they were transferred to the laboratory, water samples were filtered through 0.45 μ m membrane filters and filtrate was acidified to pH < 2 with analytical grade concentrated HNQ. As sampling through direct aspiration could not be performed using graphite furnace due to high background absorbance, for the metals except mercury, metal extraction was done using the method as described by Lo (1982) with 1% ammonium pyrrolidine dithiocarbamate and 1% sodium diethydithiocarbamate. Another set of water samples collected from each sampling site was processed for Hg analysis using hydride vapour generation system.

The sediment samples were digested using the method described by Cook et al (1997). Except for mercury, the other seven metals in water or sediment samples were analysed using flame or graphite furnace atomic absorption spectrometry as appropriate. Mercury was analysed using cold vapour atomic absorption spectrometry. Standard solutions were prepared using spectrochemical grade metal solution from BDH. Three replicates were obtained for each measurement with background correction. Recovery tests were done to examine the reliability of the test methods. Percentage recoveries obtained for the metals in the sediments ranged from 70 % to 97%.

Sampling and processing of fish for metal analysis

Fish samples were collected in 2003 from seven locations of the estuary namely Thalahena, Settappaduwa, Pitipana, Kepungoda, Kurana, Liyanagemulla and Kadolkele. In the laboratory, the total length and weight of the fish were recorded. Muscle, liver and gill tissue were homogenized separately and divided into two portions for wet and dry ashing. For determination of Pb, Cu, Cd, Cr, Mn, Ni and Zn the samples were ashed according to the method described by Jorhem (1993). Digested fish samples were analyzed using graphite furnace atomic absorption spectrometry. Wet ashing was carried out for Hg analysis according to the method given by Association of Analytical Chemists (AOAC 2002). Hg in the digested fish samples was analysed using cold vapour atomic absorption spectrometry. A known volume of standard solution was added into the analyte and samples

were processed according to the same procedures. Percentage recoveries obtained for the metals were in the range of 89% to 96%.

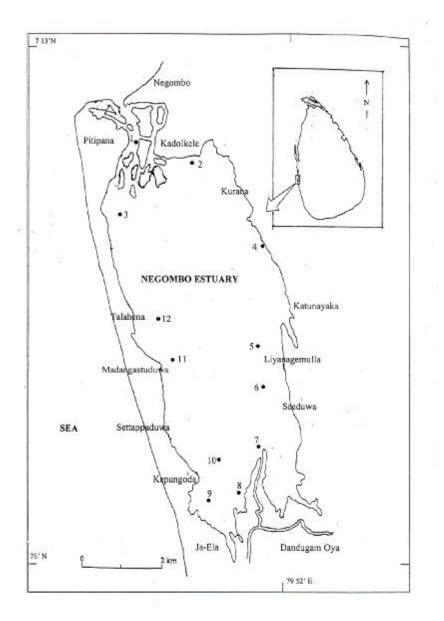


Figure 1. Locations of the sampling sites of the Negombo estuary

Data analysis

Differences in the level of each metal in water, sediment and the two species of fish collected from the estuary were tested by one way analysis of variance (ANOVA). Where differences were significant, mean values were compared by Tukey's test. Students' t test was used in the inter species comparisons. Correlations between body weight of the fish and metal content in the muscle tissue of the two species were assessed by Pearson's correlation test. The relationships between the metal level and body length or body weight were also determined by linear regression analysis. Log transformed data were used in the data analysis. Statistical analysis was carried out as described by Zar (1996). For analysis of body size relationships further, the allometric approach (Weathery and Gill 1987) was applied in which slope of log-log regression of the relationships between total metal burden and body weight or length when compared with slope =1 (for body weight) or slope =3 (for total length), is a good predictor for isometric or allometric increase of these metals with increasing body weight or length. P<0.05 was considered as statistically significant.

Results

Metal levels in water and sediments

The levels of eight metals in water at 12 sampling sites of the estuary are presented in Table 1. The concentration (in $\mu g L^1$) of the metals in water showed a wide range: Pb, not detected-5.7; Cu, not detected-2.5; Cd, not detected -2.1; Mn, not detected-0.9; Ni, not detected -7.4; Cr, 0.9-1.2; Hg, below the detection limit; and Zn, 40.4-180.4. The detection limits (in $\mu g L^1$) of the techniques for Pb, Cu, Cd, Mn, Ni, and Hg are 0.5, 0.5, 0.2, 0.2, 0.5, and 10 respectively.

The sampling sites 1, 2 and 3 were located in the north region of the estuary whereas the sites 7, 8, 9 and 10 were located in the south region (Figure 1). The sites 4, 5 and 6 were located in the east region and the sites 11 and 12 were in the west region. The results indicate that the levels of Pb, Cu and Cd in water were comparatively low in the south region of the estuary. The levels of these metals in the west and east regions were similar or lower than the levels in the north region. No prominent site related differences were found with respect to the levels of Cr in water. In addition, the levels of Pb and Cu in water appear to be high in the east side of the estuary compared to the west side.

The sediment associated metals in the estuary (in mg kg¹ dry weight) showed irregular distribution of metal contamination reflecting many metal inputs (Table 2). The ranges of the concentrations of all eight metals (in mg kg⁻¹) in sediments were: Pb, 4.8-20.1; Cu, 5.3-24.2; Cd, 0.03-0.22; Mn, 121.9-792.5; Ni, 9.2 -34.7; Cr, 19.2-73.5; Hg, 0.3-2.2 and Zn, 119.5-207.4. The highest level of Cd in the sediments was found at the sampling site 1 located in the North region whereas levels of Pb and Ni were high in the site located in the south region (site 7). Cu and Hg levels were the highest in the sediments collected from the east region (sites 5 and 4 respectively). Mn, Cr and Zn levels were high in the sampling sites located in the west region of the estuary. In general, Mn and Zn levels in the sediments were very high at all sites compared to the level of the other metals.

Metal levels in fish

Total metal levels in muscle tissue (in mg kg¹ wet weight) of the two species are presented in Table 3. The ranges of the concentrations of metals in the edible muscle tissues were: Pb 0.01-0.08; Cu 0.02 -0.37; Cd 0.002-0.048; Mn 0.05-0.51; Ni 0.01-0.31; Cr 0.02 -0.28; Hg 0.03-0.33 and Zn 0.03 - 3.02 in *E. suratensis* and Pb 0.004-0.06; Cu 0.01-0.25; Cd 0.001-0.030; Mn 0.01-0.32; Ni 0.01-0.21; Cr 0.01-0.24; Hg 0.04-0.26 and Zn 0.07-2.56 in *A. commersoni*. Accumulation of most metals in the tissues of these two species followed the increasing order, muscle < gills liver. Inter species comparison of metal levels in muscle tissue of the two fish species revealed that the levels of Pb, Cu, Mn, Ni and Cr were significantly lower in the muscle tissue of*A. commersoni* in comparison to those in*E. suratensis*. However, mean level of Zn in the muscle tissue of *A. commersoni* was two fold higher than that of*E. surentensis*. No significant species-wise differences were found between the two species in relation to the levels of Cd and Hg in the muscle and levels of all eight metals in gills and liver tissue of the fish.

Relationships between heavy metal levels and body weight

The correlations between the levels of selected eight metals in the muscle tissue of both fish species and the body size (body weight and total length) are presented in Table 4. The results show that the accumulation of each metal in the edible muscle is positively correlated with the body length or body weight of the fish. Most of these correlations were highly significant (P < 0.001).

The relationships between body weight or body length and metal level in the muscle tissue of the fish were curvilinear. Regression analysis of the log metal levels and log body weight or log body length of the fish showed linear relationships with significant correlations in most cases. Figures 2 and 3 present the linear relationships between log metal levels in the muscle tissue and log body weight or log total length of the fish with respect to an essential metal, Cu (Figure 2) and for non-essential metal, Hg (Figure 3). Slope of loglog regression of the relationship between Cu level in muscle tissue of E. suratensis and total body weight was 1.049 which was not significantly different from 1 indicating that Cu level increases in direct proportion to an increase in body weight showing isometry. The slope of the log-log regression of the relationship between Cu level of this fish and total length was 2.89 which was not significantly different from 3 also indicating the isometric relationship. Other metals in E. suratensis and all the eight metals in A. commersoni also showed increase in metal concentration with increase in body length or body weight. However, the slopes of the regression lines between the other metals and body weight or total length were significantly different from 1 (for body weight) or 3 (for total length) showing positive allometric relationships.

Metal	Metal levels ($\mu g L^{-1}$)											
	Site 1	Site 2	Site 3	Site 4	Site 5	Site 6	Site 7	Site 8	Site 9	Site 10	Site 11	Site 12
Pb	5.2	5.7	3.6	1.1	1.4	1.0	_*	_*	_*	_*	_*	_*
Cu	2.5	2.1	0.6	1.1	1.7	1.6	_*	_*	_*	_*	_*	1.0
Cd	1.4	2.1	0.6	0.9	1.9	0.9	_*	_*	_*	_*	1.5	1.1
Mn	0.8	_*	_*	0.8	0.8	0.8	0.7	0.7	0.6	_*	0.8	0.9
Ni	2.2	_*	7.4	2.1	2.1	1.7	1.4	1.5	1.15	1.1	1.3	2.1
Cr	1.0	1.2	1.1	1	1.1	0.9	1.0	0.9	0.9	1.0	1.0	1.0
Hg	_*	_*	_*	_*	_*	_*	_*	_*	_*	_*	_*	_*
Zn	120.1	130.1	40.4	110.1	50.1	141.2	112.0	81.1	84.2	61.1	180.4	41.2
	1_0.1	100.1		110.1	0001	1.1.2	112.0	0111	02	0111	100.1	

Table 1. The levels of selected metals in water at 12 sampling sites of Negombo estuary.

*Below the detection limits: Pb, Cu & Ni 0.05 μ g L⁻¹, Cd & Mn 0.02 μ g L⁻¹, Hg 10 μ g L⁻¹

Metal	Metal levels (mg kg ¹)											
	Site 1	Site 2	Site 3	Site 4	Site 5	Site 6	Site 7	Site 8	Site 9	Site 10	Site 11	Site 12
Pb	5.1	4.8	5.6	10.7	15.0	13.9	20.1	7.4	12.5	5.9	16.5	12.1
Cu	6.6	5.3	6.2	11.8	24.2	16.4	16.7	12.6	9.4	6.1	6.2	13.4
Cd	0.22	0.17	0.11	0.07	0.09	0.04	0.03	0.07	0.03	0.05	0.04	0.05
Mn	121.9	137.5	298.5	284.1	276.2	498.8	276.1	331.4	303.4	305.9	792.6	670.4
Ni	15.5	24.9	13.5	10.6	17.1	19.2	30.7	17.3	9.2	14.7	29.7	34.7
Cr	36.8	58.6	33.8	26.3	28.2	32.4	47.5	38.9	19.2	46.8	55.9	73.5
Hg	0.6	0.6	0.5	2.2	0.7	0.6	0.3	1.4	0.3	0.5	0.51	0.75
Zn	120.5	119.5	133.6	145.6	201.5	172.8	161.1	141.1	148.3	126.5	207.4	176.5
Zn	120.5	119.5	133.6	145.6	201.5	172.8	161.1	141.1	148.3	126.5	207.4	

Table 2. The levels of selected metals in sediments at 12 sampling sites of Negombo estuary.

Metals	Metals levels in mg kg^1 (wet weight)							
	Muscle	Gills	Liver					
Etroplus s	uratensis (n=150)							
Pb	$0.03 \pm 0.001 (0.01 - 0.08)^1$	$0.05 \pm 0.004 (0.02 - 0.08)^2$	$0.07 \pm 0.004 (0.04 - 0.09)^2$					
Cu	$0.13 \pm 0.006 (0.02 - 0.37)^1$	$0.23 \pm 0.03(0.10 - 0.41)^2$	$0.41 \pm 0.05(0.15 - 0.74)^2$					
Cd	$0.011 \pm 0.001 (0.002 - 0.048)^1$	$0.027 \pm 0.002 (0.017 - 0.042)^2$	$0.027 \pm 0.003 (0.011 - 0.06)^{2*}$					
Mn	$0.12 \pm 0.007 (0.05 - 0.51)^1$	$0.11 \pm 0.007 (0.06 - 0.17)^1$	$0.42 \pm 0.03 (0.16 - 0.66)^2$					
Ni	$0.05 \pm 0.004 (0.01 - 0.31)^1$	$0.09 \pm 0.005 (0.05 - 0.12)^2$	$0.11\pm0.01(0.09-0.20)^2$					
Cr	$0.07 \pm 0.003 (0.02 - 0.28)^1$	$0.20\pm0.02(0.09-0.33)^2$	$0.39 \pm 0.05 (0.05 - 0.86)^3$					
Hg	$0.10\pm0.005(0.03-0.33)^1$	$0.18 \pm 0.02 (0.10 - 0.42)^2$	$0.51 \pm 0.03 (0.31 - 0.79)^3$					
Zn	$0.50\pm0.04(0.03-3.02)^1$	$2.13\pm0.26(1.12-5.12)^2$	$4.55\pm0.43(1.24-8.71)^3$					

Table 3. The level of selected metals in different tissues o Etroplus suratensis and Ambassis commersoni from Negombo estuary

Table 3 Continued on page 72

 Table 3. Continued from page 71

Metals		Metals levels in mg kg^1 (we	t weight)
	Muscle	Gills	Liver
Ambassis co	ommersoni (n=95)		
Pb	$0.02 \pm 0.001 (0.004 - 0.06)^{1**}$	$0.04 \pm 0.003 (0.01 - 0.07)^2$	$0.07 \pm 0.006 (0.02 - 0.10)^3$
Cu	$0.08 \pm 0.005 (0.01 - 0.25)^{1**}$	$0.19 \pm 0.02(0.07 - 0.32)^2$	$0.38\pm0.04(0.12-0.74)^3$
Cd	$0.013 \pm 0.005 (0.001 - 0.030)^1$	$0.027 \pm 0.002 (0.014 - 0.045)^2$	$0.034 \pm 0.003 (0.018 - 0.065)^{2*}$
Mn	0.10±0.01(0.01-0.32) ^{1**}	$0.11 \pm 0.01 (0.05 - 0.19)^1$	$0.35 \pm 0.01 (0.30 - 0.48)^2$
Ni	$0.03 \pm 0.003 (0.01 - 0.21)^{1**}$	$0.09 \pm 0.005 (0.03 - 0.14)^2$	$0.11 \pm 0.01 (0.07 - 0.21)^2$
Cr	$0.05 \pm 0.005 (0.01 - 0.24)^{1**}$	$0.22 \pm 0.02 (0.01 - 0.47)^2$	$0.32\pm0.06(0.10-0.74)^2$
Hg	$0.10\pm 0.005(0.04-0.26)^1$	$0.24 \pm 0.03 (0.05 - 0.62)^2$	$0.53 \pm 0.05(0.14 - 0.85)^3$
Zn	$0.83 \pm 0.05 (0.07 - 2.56)^{1**}$	$2.95 \pm 0.27 (0.90 - 6.53)^2$	3.85±0.53(1.23-7.84) ²

Results are presented as mean \pm standard error of the mean, ranges (lowest value-highest value). For each row means not followed by the same superscript are significantly different from each other (ANOV A, Tukey's test \oplus .05). Log transformed data {log (x+1)} were used for the statistical analysis.

* 6% of the samples of *E. suratensis* and 12% of the *A. commersoni* samples exceeded the maximum permissible limit for Cd $(0.05 \text{ mg kg}^{-1})$.

** Significantly different from *E. suratensis* (Students' t test, $\alpha = 0.05$).

	Body weight a	nd metal level	Body length and metal level				
Metal	correlation	P value	correlation	P value			
	coefficient		coefficient				
Etroplus suratensis							
Pb	0.500	0.001	0.472	0.001			
Cu	0.729	0.001	0.742	0.001			
Cr	0.184	0.05	0.233	0.05			
Cd	0.773	0.001	0.745	0.001			
Mn	0.568	0.001	0.609	0.001			
Ni	0.767	0.001	0.794	0.001			
Zn	0.726	0.001	0.716	0.001			
Hg	0.850	0.001	0.859	0.001			
Ambassis commersoni							
Pb	0.770	0.001	0.759	0.001			
Cu	0.843	0.001	0.825	0.001			
Cr	0.704	0.01	0.775	0.001			
Cd	0.224	0.05	0.237	0.05			
Mn	0.814	0.001	0.745	0.001			
Ni	0.614	0.001	0.667	0.001			
Zn	0.830	0.001	0.828	0.001			
Hg	0.940	0.001	0.982	0.001			

Table 4. Pearson correlation coefficients for the relationships between metal

 levels and body size* of *Etroplus suratensis* and *Ambassis commersoni*

**E. suratensis:* body weight 47.0-140.6 g, body length 10.3-17.6 cm, n=114-118, *A. commersoni* body weight 21.3-91.7 g, body length 5.2 - 11.2 cm, n=91-94

Discussion

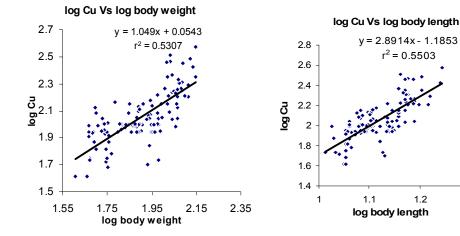
Water quality of Negombo estuary is influenced by several inland water sheds (Dadugam oya and Hamilton canal) and the sea. The fresh water canals and the tidal floods may transport toxic pollutants originating in different parts of the catchments, land use or in the estuary itself. The estuary is susceptible to chemical pollution due to ongoing development activities of Negombo and Katunayake areas (CEA 1994). Estuary water is not utilized for human consumption because of its salty nature. However, monitoring the status of metal pollution in Negombo estuary would be important to human health because of the fishery of the estuary. Metal level of water and sediment directly relate to the aquatic life of the estuary. Metals may enter the resident fish in different ways and absorbed metals could accumulate in various organs of the fish body. The present study provides baseline data with respect to the eight metals viz. Pb, Cu, Cd, Cr, Mn, Ni, Hg and Zn in water and sediments of the Negombo estuary as well as for the two species of food fish available in the estuary.

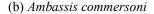
In the present study it was revealed that spatial variations exist in different regions of the estuary, with respect to the dissolved total metals in water: Pb, Cu, Cd, Mn, Ni, and Zn. In general, the levels of Pb, Cu, and Ni in water collected from some sampling sites located in the north region were higher compared to those of the other sites. North region of the estuary is being polluted due to various anthropogenic activities such as solid waste dumping, waste from industries, slaughter houses, shrimp farms, and hatcheries and boat yards. High levels of Pb in water samples collected from the north region in comparison to those in the other sampling sites may be partly due to the discharge of burned and unburned fuel from motor boats to the estuary water. The north region of the estuary is connected to the sea and it receives water from sea as well. Though the levels of Pb, Cu and Ni in water in all or some sampling sites in this region were higher than those in the other sites, the levels of sediments bound Pb and Zn were comparatively low. The status of the sediment of this area is continuously being affected by tidal waves, sedimentation and human activities. Hence the level of metals in the sediments of this area is subjected to change through out the year and this may have partly contributed to low Pb and Zn accumulation in sediment of this area in comparison to the other sites.

The sampling sites 7, 8, 9 and 10 are located in the south region. This region receives water mainly from the Hamilton canal and Dadugam oya and they carry various effluents from Ekala industrial zone (CEA 1994). In comparison to the north region of the estuary, the levels of metals in water of south region (Pb, Cu, Cd, and Mn) were low but the levels of metals in sediments (Pb, Cu, Zn, and Mn) were higher in some sampling sites compared to the metal levels in the sites located in the north region. West region (sampling sites 11 and 12) receives waste from hotels, shrimp farms and fish processing industries. Generally the levels of Pb and Cu in water of this area were low but the levels of sediment bound metals (Pb, Cu, Mn, Ni, Cr and Zn) were comparatively high. East region (Sites 4, 5 and 6) receives effluent from mainly Katunayake industrial processing zone, hotels and a housing scheme. Hence the levels of metals in water (Pb, Cu, Ni and Zn) sampled from this area were relatively high.

Accumulation of metals in different fish species depends on the bioavailable metal concentrations in the abiotic components of their habitats, their feeding habits, ecological needs, metabolism, age and size of the fish (Peakall and Burger 2003; Marcovecchio 2004). Observed wide ranges of specific metal levels detected in the fish collected from the estuary may indicate that these fish had been exposed to a mixture of metals with different bioavailabilities within the estuary through water, sediments and food items.

(a) Etroplus suratensis





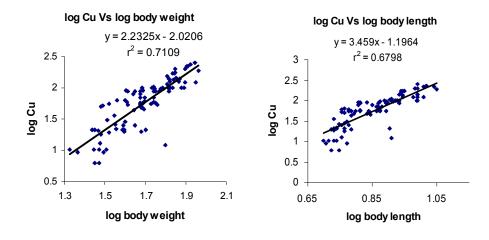
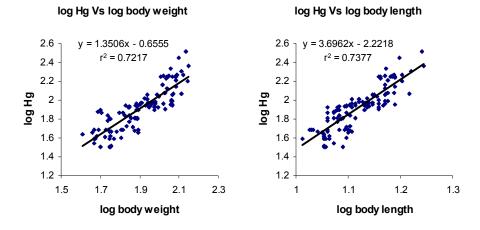


Figure 2. Relationship between copper level in muscle tissue and log body weight of Etroplus suratensis and Ambassis commersoni from Negombo estuary

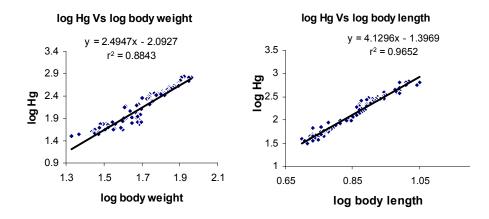
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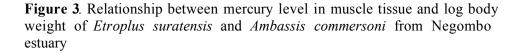
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(a) Etroplus suratensis



(b) Ambassis commersoni





In the present study, results of the measured metals of the two species followed the order, muscle < gil \leq liver showing different metal accumulation capacities of these tissues of *E. suratensis* and *A. commersoni* from Negombo estuary. The results of this study show that the muscle tissue of these fish is not an active organ in accumulating heavy metals. Metal levels in liver reflect

the high metal storage capacity of the organ. Liver stores heavy metals by producing metallothioneins which appears as a metal detoxification mechanism within the body (Roesijadi and Robinson 1994; Peakall and Burger 2003). Metallothioneins are a family of low molecular weight cystein rich proteins that occur in the animals. Their synthesis can be induced by a wide variety of metal ions including cadmium, copper, and zinc. In the present study gill tissues of the fish collected from several occasions also had high metal contents comparable to liver tissues. In general the concentrations of metals in the gills may reflect the concentration of metals in water. Metal concentrations in the gills could also be due to complexing of the elements with mucus remaining between the gill lamella, which is hard to remove completely from the gills before preparation of the tissue for analysis (Demirak et al. 2005). The accumulation of metals in gills and liver of food fish do not directly affect human health because these are not edible parts. Nevertheless the predatory animals such as birds who consume the whole fish from the estuary are at risk of excess metal contamination.

Pb, Cd, and Hg are biologically non essential- metals which are accumulated in human tissues and harmful to human health. The level of Pb, Cd and Hg in the edible muscle of E. suratensis collected from Negombo estuary ranged from 0.01 - 0.08, 0.002 - 0.048 and 0.03 - 0.33 mg kg⁻¹ wet weight respectively. Corresponding values for edible muscle of *commersoni* were 0.004 - 0.06, 0.001 - 0.030 and 0.04 - 0.26 mg kg⁻¹ wet weight. The maximum allowable levels of Pb, Cd and Hg in the fish for human consumption specified by the European Union are 0.2, 0.05 and 1 mg kg wet weight respectively (EU 2002). Of the different metal levels detected inE. suratensis and A. commersoni the levels of the three metals in the edible muscle of all the fish collected from Negombo estuary did not exceed these allowable limits in fish for human consumption specified by European Union. However, the liver tissue of 6% of E. suratensis and 12% of A. commersoni collected from the estuary exceeded the limit for Cd. The maximum levels of Pb, Cd, and Hg in food fish specified by the EU are lower than the other international standards for food. According to the median international standards for food the tolerable levels for Pb, and Cd in food are 2 and 0.3 mg kg⁻¹ wet weight respectively (Philips 1993). The metal levels in the fish from Negombo estuary were much below these international standards for Pb and Cd.

Cu, Mn, Ni, Cr and Zn are essential metals for the biota and are regulated by physiological mechanisms in most organisms. However, occurrence of excessive levels of them is regarded as potential hazards that can endanger both animal and human health (Wright and Welbourn 2002). The ranges of the levels of Cu, Mn, Ni, Cr and Zn in the edible muscle of *E. suratensis* collected from Negombo estuary were 0.02 - 0.37, 0.05 - 0.51, 0.01 - 0.31, 0.02 - 0.28 and 0.03 - 3.02 mg kg⁻¹ wet weight respectively. The corresponding values in the muscle of *A. commersoni* were 0.01 - 0.25, 0.01 - 0.32, 0.01 - 0.21, 0.01 - 0.24, and 0.07 - 2.56 mg kg⁻¹ wet weight respectively. The median international standard levels of Cu, Cr and Zn in the food for

human consumption are 20, 1 and 45 mg kg¹ wet weight respectively (Philips 1993). The levels of these essential metals in the edible muscle of both species of fish collected from Negombo estuary were much lower than the median international standard levels. Hence, these metal levels have posed no threat for consumption of the fish muscle from the estuary.

A. commersoni is a carnivorous fish whereas *E. suratensis* is an omnivorous fish which feeds on macrophytes, detritus, diatoms and animal matter (Pethiyagoda 1991). In the present study it was revealed that levels of Cd, Cr, Mn, and Ni in muscle tissues of *E. suratensis* is higher than those of *A. commersoni*. The level of Zn in the edible muscle of *A. commersoni* was higher than that in *E. suratensis*. Degree of bioaccumulation of metals in the muscle of these fish may be related to the food and feeding habits of these fish.

Accumulations of Pb, Cu, Cd, Cr, Hg, Mn, Ni and Zn in the muscle tissue of these two fish species were positively correlated with the body weight and body length. Further, slope of log-log regression of the relationship between Cu level in muscle tissue of *E. suratensis* and total body weight was not significantly different from 1 indicating that the metal level increase in direct proportion to an increase in body weight showing isometry (Weathery and Gill, 1987). Moreover, the slope of the log-log regression of the relationship between Cu level of this fish and total length was not significantly different from 3 indicating the isometric relationship. Other metals inE. suratensis and all the eight metals in A. commersoni also showed increase in metal concentration with increase in body length or body weight. However, the slopes of the regression lines between the other metals and body weight or total length were significantly different from 1 (for body weight) or 3 (for total length) showing positive allometric relationships (Weathery and Gill 1987). The results suggest that these metals can accumulate in the muscle tissue of *E*. and A. commersoni at a higher rate compared to the rate of suratensis excretion of these metals as the fish grows.

In conclusion, accumulation of metals in the selected two food fish species (E. suratensis and A. commersoni) from Negombo estuary was organ specific and species specific. Generally metal accumulation in tissues of these two species followed the increasing order, muscle < gill≰ liver. Based on the levels of toxic non-essential heavy metalsviz. Hg, Pb, and Cd in body muscle of both fish species, the fish muscle was found to be safe for human consumption. The levels of essential metals testedviz. Cu, Cr, Mn, Ni and Zn have posed no threat for consumption of the muscle of these fish from the estuary. However, heavy consumption of bigger sizeE. suratensis and A. commersoni from the Negombo estuary may pose a health risk to the consumers as there were strong positive correlations between body weight or body length and metal levels in edible muscle tissue of both species of fish. Hence, the necessity of regular monitoring of heavy metal pollution in Negombo estuary is emphasized as there are multiple sources of heavy metal contamination in the vicinity of the estuary. The current study may provide baseline data for water and sediments of Negombo estuary and for fish available from the estuary. More detail study on seasonal trends and spatial

variations in heavy metal pollution in the Negombo estuary would be useful to locate point and non-point sources of heavy metal pollution and to take necessary steps to manage and remediate the ecological damage.

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