Vidyodaya J. of Sci, (2006) Vol. 13, pp 115-126

Heavy metal levels in food fish, Etroplus suratensis inhabiting Bolgoda Lake, Sri Lanka

***P. Senarathne*[1], *K. A. S. Pathiratne*[1*] *and A. Pathiratne*[2]**
1. Department of Chemistry, University of Kelaniya, Kelaniya, Sri Lanka
2. Department of Zoology, University of Kelaniya, Kelaniya, Sri Lanka

* *Corresponding author (**E- mail : kaspathi@kln.ac.lk**)*

Received on : 05/29/07
Accepted on : 06/21/07

Abstract

Bolgoda Lake is used by the local community for traditional fishery. Nevertheless it is becoming polluted with chemicals from various sources. The present study was carried out to assess the total levels of five heavy metals viz lead, cadmium, chromium, copper and zinc in the muscle, gill and liver tissues of Etroplus suratensis, a food fish inhabiting Bolgoda Lake. Fish samples were collected from four main locations of the Lake viz. Weras Ganga, North Lake, Bolgoda Ganga and South Lake and metal levels were determined by atomic absorption spectrometry. Total metal levels in muscle tissue (in $\mu g\ g^{-1}$ wet weight) of the fish collected from Bolgoda Lake showed a wide range: lead, 0.02- 20.1, cadmium, 0.01 - 0.3, chromium, 0.01-0.2, copper, 0.1 – 10.1, and zinc, 2.5-10.4 (n=60). Of the different metal levels detected in the edible muscle of the fish, the level of Pb in 100% of the fish sampled from Weras Ganga, 87% of the fish from Bolgoda Ganga and 13% of the fish from North Lake and the level of Cd in 13 % of the fish from Weras Ganga and Bolgoda Ganga exceeded the maximum levels in food for human consumption specified by European Union. Comparison of the metal levels in the muscle, gills and liver tissues indicated that the highest accumulation of metals occurred in the liver tissue of the fish. The results indicate that heavy consumption of E. suratensis from Bolgoda Lake especially from Weras Ganga and Bolgoda Ganga may pose a health risk to the consumers.

Key words: Heavy metals, Bolgoda Lake, Etroplus

1. Introduction

Heavy metals from man-made pollution sources which are released into aquatic ecosystems pose a serious threat to biota. Heavy metals may accumulate to toxic concentrations under certain environmental conditions and cause ecological damage. Some heavy metals such as lead and cadmium are non-essential metals that are toxic to animals and human even at very low concentrations. Chromium, copper and zinc are essential trace metals. However, high concentrations of some essential trace metals can become toxic at concentrations exceeded the limits, which are required (Wright and Welbourn, 2002).

Fish are considered as indicator organisms in aquatic ecosystems for estimation of the heavy metal pollution and risk potential of human consumption (Peakall & Burger 2003; Marcovecchio 2004; Demirak et al 2005; Dalman et al 2006). Heavy metals are taken up through different organs of the fish and concentrating at different levels in different organs at the fish body. Hence it is important to determine the concentrations of heavy metal levels in commercial fish in order to evaluate the possible risk of fish consumption. The limited number of studies that has been undertaken to assess heavy metal levels in food fish species collected from several inland water bodies in Sri Lanka (Wijesinghe et al. 1999; Allinson et al. 2002; Anil & Pathiratne 2002; Silva & Shimizu 2004; Witharana et al. 2005; Indrajith et al. 2006; Manage et al. 2006) indicate that some heavy metals are concentrating at different levels in the sampled fish.

Bolgoda Lake, a water body in the Western province of Sri Lanka is used by the local community for various purposes including fishery and irrigation of agricultural areas. Nevertheless it receives urban and industrial wastes from multiple sources (Silva, 1996). Fish captured from Bolgoda Lake are used by the local people in the area as a protein source. Few reports are available on metal levels in some food fishes collected from selected sampling sites of Bolgoda Lake. Wijesinghe et al. (1999) have reported high concentrations of several heavy metals in tilapia, Oreochromis mossambicus from Weras Ganga, extreme north end of Bolgoda North Lake. Bioaccumulation of lead, cadmium and copper in tilapia, Oreochromis mossambicus in Bolgoda Lagoon area was assessed by Anil & Pathiratne (2002). Recently bioaccumulation pattern of lead, cadmium, chromium, zinc, nickel and copper of selected edible fish, Ambassis commersoni, Netuma thalassinus, and Mugil cephalus from Horethuduwa and Panadura estuary was studied by Manage and Wijethilaka (2006). Etroplus suratensis (Iri Koraliya) is an edible fish widely distributed in Bolgoda Lake. The present study evaluates the levels of five heavy metals viz. lead, cadmium, chromium, copper and zinc in muscle, gills and liver tissues of E. suratensis collected from Bolgoda Lake, covering Weras Ganga, North Lake, Bolgoda Ganga and South Lake.

2. Materials and Methods

Sampling of fish

Samples of E. suratensis (18.4-24.3 cm in total length, 172.9-529.7g in body weight) were collected from the four main locations of Bolgoda Lake namely Weras Ganga, Bolgoda North Lake, Bolgoda Ganga and Bolgoda South Lake during March - April 2003. Freshly dead fish were randomly selected from the fish caught from the sampling sites using gill nets and cast nets. They were immediately transported to the laboratory in polyethylene bags under chilled condition and kept at -20^{0}C until analysis.

Processing of fish tissues for metal analysis

Fish samples were allowed for thawing and the scales and the skin of the fish were removed using a plastic knife to avoid metal contamination. The muscles, gills and liver of fish were taken into pre acid washed crucibles and they were homogenized separately. The weight of the each homogenized sample was recorded. Then the samples were kept in a drying oven at 100 °C for more than 48 hours until a constant weight was observed. The dried sample was then ground into a fine powder using a porcelain motor and pestle. Powdered fish tissues were digested using a dry ashing procedure (Jorhem, 1993). A quantity of 1-2 g of dried sample was accurately weighed into pre-acid washed crucibles and kept in a muffle furnace at an initial temperature less than 200 °C. Then the temperature was increased up to 450 °C at the rate of about 50 °C per hour and kept at the same temperature overnight. After cooling, 1-3 ml of deionized water was added to ash and the resultant solution was evaporated in a hot plate. The crucibles were returned to the muffle furnace and raised the temperature up to 450 °C with steps of 50 °C at a time. The ashing was preceded at 450 °C for more than three hours. This procedure was repeated until the sample was completely combusted. Then the samples were treated with 5 ml of 6 M hydrochloric acid and evaporated to dryness on a hot plate. The residue was dissolved using a volume of 0.1 M nitric acid in the range of 10-30 ml as appropriate. The crucible was swirled to contact all ash with nitric acid. The crucible was covered with a watch glass and kept for 1-2 hours. Then the solution was transferred into plastic tubes for the analysis.

Metal Analysis

Concentrations of lead, cadmium, chromium, copper and zinc in the fish tissues were analysed by graphite furnace atomic absorption spectrometry and/or flame atomic absorption spectrometry. The GBC mode 932 plus atomic absorption spectrometer equipped with a graphite furnace 3000 system and PAL 3000 auto sampler was used for determinations. BDH analytical grade reagents were used to establish calibration curves and blank measurements and recovery tests were carried out to examine the reliability of the test methods. External calibration method or the standard addition method was used to determine the metal concentrations in the samples. The percentage recovery of metals ranged between 87-99%.

Statistical analysis

Data are presented as mean SEM of 15 fish for each location. Sampling location, metal type, and tissue specific differences were statistically tested by one way analysis of variance (ANOVA). Mean values were compared by Tukey's test. Log transformed data [log (x+1)] were employed for all the statistical analysis and $p<0.05$ was considered as statistically significant (Zar, 1996).

3. Results

Metal levels in muscle tissue

The levels of the five metals in the muscle tissue of E. suratensis collected from four locations are presented in Table 1. Of the five metals measured, zinc was the most abundant metal in the muscle tissue of the fish collected from all locations. The levels of copper in muscle tissue of the fish from North Lake and South Lake were significantly higher than those of lead, cadmium and chromium. The fish collected from Weras Ganga and Bolgoda Ganga had significantly higher levels of lead in the muscle tissue in comparison to the cadmium, chromium and copper levels in the muscle.

Location related comparison showed that the levels of lead in the muscle tissue was significantly higher in the fish collected from Weras Ganga compared to the levels in the fish collected from the other three locations (Table 1). Muscle tissue of the fish collected from Weras Ganga and Bolgoda Ganga had the highest cadmium and chromium content compared to the fish from North Lake and South Lake. The levels of copper in the muscles of fish from North Lake were significantly higher compared to the copper level in the fish collected from the other locations. Zinc levels in the muscle tissue of the fish were higher in the fish collected from Weras Ganga, North Lake and Bolgoda Ganga compared to the fish from South Lake. Location-specific comparison showed that the levels of the five metals in the muscle tissue of E. suratensis collected from Bolgoda Lake followed the order: Lead, Weras Ganga > Bolgoda Ganga >North Lake > South Lake, Cadmium and Chromium, Weras Ganga ≈ Bolgoda Ganga > North Lake ≈ South Lake, Copper, North Lake >Weras Ganga ≈ South Lake > Bolgoda Ganga and Zinc, Weras Ganga ≈North Lake ≈Bolgoda Ganga > South Lake. The measured heavy metals in the fish collected from South Lake were comparatively lower than the levels in the fish from other locations in the Lake.

Results shown in the Table 1 also indicate that the lead levels in the muscle of all E. suratensis sampled from Weras Ganga and 13% of the fish from North lake and 87% of the fish from Bolgoda Ganga exceeded the EU maximum permissible level for lead (0.2 $\mu g\ g^{-1}$) in food fish for human consumption. The cadmium level in the muscle of 13% of the fish collected from Weras Ganga exceeded the maximum permissible levels (cadmium 0.05 $\mu g\ g^{-1}$).

Metal levels in gill and liver tissues

The levels of the five metals in the gill and liver tissues of E. suratensis collected from four locations are presented in Tables 2 and 3 respectively. Of the five metals measured, zinc was the most abundant metal also in the gills and liver of the fish collected from all locations. Location-specific comparison showed that the levels of the five metals in the gills of E. suratensis collected from Bolgoda Lake followed the order: Lead, Weras Ganga > Bolgoda Ganga ≈ North Lake ≈ South Lake, Cadmium, Weras Ganga > Bolgoda Ganga ≈ North Lake > South Lake, Chromium, Weras Ganga ≈ Bolgoda Ganga > North Lake > South Lake, Copper, North Lake >Weras Ganga ≈ South Lake ≈ Bolgoda Ganga and Zinc, Weras Ganga ≈ North Lake ≈ South Lake >Bolgoda Ganga. The levels of the five metals in the liver of E. suratensis collected from Bolgoda Lake followed the order: Lead, Weras Ganga ≈ Bolgoda Ganga >North Lake ≈ South Lake, Cadmium, Weras Ganga ≈ Bolgoda Ganga > North Lake ≈ South Lake, Chromium, Bolgoda Ganga > Weras Ganga ≈ North Lake ≈ South Lake, Copper, North Lake >Weras Ganga ≈ Bolgoda Ganga ≈ South Lake and Zinc, Bolgoda Ganga ≈ South Lake > Weras Ganga ≈ North Lake .The measured heavy metals in the gills and liver of the fish collected from South Lake (except zinc levels in liver) were comparatively lower than the levels in the fish from other locations in the Lake.

Comparison of metal levels in three tissues

The levels of five metals in the muscle, gill and liver tissues of E. suratensis are compared in Table 4. The levels of metals in liver tissues were significantly higher than the respective metal levels in muscle tissues. The metal levels in liver tissues compared to the muscle tissues were several folds higher than the levels in the muscle issue: lead 5-76 folds, cadmium 9-40 folds, chromium 5-10 folds, copper 10-72 folds, and zinc 4-19 folds. The levels of zinc, in muscle tissue of the fish from all locations were significantly lower than the respective metal levels in the gill tissues. The levels of other four metals in the muscle tissues of the fish with a few exceptions were significantly not different from the respective metal levels in the gill tissues. Level of copper in the muscle of the fish from Bolgoda Ganga and North Lake and level of lead in the muscle of fish from South Lake were significantly lower than the respective metal levels in the gill tissues.

4. Discussion

Accumulation of metals in different fish species depends on the bioavailable metal concentrations in their habitats, their feeding habits, ecological needs, metabolism, age and size of the fish (Peakall and Burger 2003; Marcovecchio 2004). E suratensis is an omnivorous fish naturally inhabiting in the Bolgoda Lake. It mainly feeds on macrophytes (Ref). Plants can bioaccumulate high levels of metals. Hence organisms that feed on plant material, generally may expose to higher levels of metals through food in addition to the metal uptake from the surrounding water.

Concentrations of selected five metals detected in muscle, gills and liver tissues of E. suratensis from Bolgoda Lake showed different capacities for metal accumulation. In general the metal accumulation was found to be very high in liver in comparison to the muscle tissues of the fish. Metal levels in liver reflect the high metal storage capacity of the organ. Several studies showed that heavy metal accumulate mainly in metabolic organs such as liver that stores metals by producing metallothioneins which appears as a metal detoxification mechanism within the body (Ref). 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. Hence metallothioneins have been proposed as biomarkers to indicate the presence of high levels of metals in the environment (Peakall and Burger 2003). The results of this study show that the muscle of E. suratensis is not an active organ in accumulating heavy metals. In the present study gill tissues of the fish collected from several occasions also had high metal contents comparable to liver tissues where as in the other occasions the metal contents in gills were not significantly different from the levels in the muscle tissue. In general the concentrations of metals in the gills reflect the concentration of metals in water (Rao and Padmiga 2000). 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 (ref) 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 are at risk of excess metal contamination.

Lead and cadmium are non essential elements which are accumulated in human tissues and harmful to human health. The level of lead and cadmium in the edible muscle of E. suratensis collected from Bolgoda Lake ranged from 0.02- 20.1 and 0.01 - 0.3 g g^{-1} wet weight respectively. The maximum allowable levels of lead and cadmium in the fish for human consumption specified by the European Union are 0.2 and 0.05 g g^{-1} wet weight respectively (EU 2002). Of the different metal levels detected in the edible muscle of the fish in the present study the level of Pb in 100% of the fish from Weras Ganga, 87% of the fish from Bolgoda Ganga and 13% of the fish from North Lake and the level of Cd in 13 % of the fish from Weras Ganga and Bolgoda Ganga exceeded the maximum levels in fish for human consumption specified by European Union. The maximum levels of lead and cadmium specified by the EU are lower than the other international standards. According to the median international standards, the tolerable levels for lead and cadmium in food are 2 and 0.3 g g^{-1} wet weight respectively (Philips 1997). Lead levels in the muscle tissue of 67 % of the fish collected from Weras Ganga and 27 % of the fish from Bolgoda Ganga exceeded this international standard for lead. Cadmium levels in the 13% of the fish from Bolgoda Ganga exceeded the international standard for cadmium in food. Hence heavy consumption of E. suratensis especially from Weras Ganga and Bolgoda Ganga may pose a health risk for fish consumers.

Although biota can accumulate chromium, tissue concentrations are often low as it is carefully regulated by physiological mechanisms (Wright and Welbourn, 2002). The level of chromium in the edible muscle of E. suratensis collected from Bolgoda Lake ranged from 0.01-0.2 g g^{-1} wet weight. Chromium levels in the muscle were lower than the median international standards (1 g g^{-1} wet weight) of chromium in food (Philips 1997). However chromium levels in the liver of the fish collected from Bolgoda Ganga exceeded this limit.

Copper and Zinc are essential elements and are regulated by physiological mechanisms in most organisms. They accumulate in porphyrins and enzymes. 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 levels of copper and zinc in the edible muscle of E. suratensis collected from Bolgoda Lake ranged from 0.1 – 10.1 and 2.5-10.4 g g^{-1} wet weight respectively. The median international standard levels of copper and zinc in the food for human consumption are 20 and 45 g g^{-1} wet weight respectively (Philips 1997). The levels of copper and zinc in the edible muscle of E. suratensis collected from all locations of Bolgoda Lake were lower that these median international standard levels. Hence copper and zinc levels have posed no threat for consumption of the fish muscle from the Lake. However liver tissues of the fish collected from North Lake, Bolgoda Ganga and South Lake exceeded these standards.

Importance of Biomonitors Concentrations of heavy metals, lead, cadmium, chromium, copper and zinc in E. suratensis from four main locations of the Bolgoda Lake reflect the metal content in the fish taken from the Lake. Hence it may be used as a quality index for evaluation of bioavalable metal pollution in the Lake. It may be argued that E. suratensis could move from one location of the Lake to another and fish may be exposed to a mixture of metals with different bioavailabilities within the Lake through water and food items. This may be one of the reasons for the observed wide range of specific metal levels detected in the fish samples collected from each main location. Nonetheless location-specific comparison of the five metals in E. suratensis revealed that the metals in the fish collected from Weras Ganga and Bolgoda Ganga are comparatively higher than the respective levels in the fish from South Lake. Hence, there is more heavy metal adding sources especially in Weras

Ganga and Bolgoda Ganga compared to South Lake. A study on spatial variations in the heavy metal levels in water and sediments in Bolgoda Lake also revealed that overall pollution of the metals in water and sediments is comparatively low in the South Lake (Senarathne et al. 2006). In a more recent study on accumulation pattern of selected heavy metals in some edible fish species from Panadura estuary and Horethuduwa detected high bioaccumulation factors for cadmium, chromium and copper in plankton from Panadura estuary and zinc in a food fish, Netuma thalassinus.(Manage and Wijethilaka 2006). Observed high levels of copper in the muscle, gills and liver tissues of E. suratensis sampled from North Lake may indicate that more copper adding sources are present along the vicinity of the North Lake. CEA report

In conclusion, analysis of metals in the tissues of E. suratensis indicates the levels of lead in muscle (edible part) of the fish collected from Weras Ganga, Bolgoda Ganga and North Lake and the level of Cd in some of the fish from Weras Ganga and Bolgoda Ganga exceeded the food safety limits specified by the international authorities. Comparison of the metal levels in the muscle, gills and liver tissues indicated that the highest accumulation of metals occurred in the liver tissue of the fish. Location-wise comparison of the five metals in the fish revealed that the metals in the fish collected from Weras Ganga and Bolgoda Ganga are comparatively higher than the respective levels in the fish from South Lake except in few occasions. The results showed that heavy consumption of E. suratensis from Bolgoda Lake especially from Weras Ganga and Bolgoda Ganga may pose a health risk to the consumers due to accumulation of high levels of lead and cadmium. The results emphasize the importance of regular monitoring of heavy metals in the food fish species in Bolgoda Lake as the Lake receives urban and industrial wastes from multiple sources located along the vicinity of the Lake.

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Table 1. Levels of metals in edible muscle tissues of *Etroplus suratensis* collected from different locations of Bolgoda Lake.

	Metal level (μg g^{-1} wet weight)				
Location	Lead	Cadmium	Chromium	Copper	Zinc
Weras Ganga	**4.4 ± 1.5**^{a2} 0.5 - 20 [100%]	**0.1 ± 0.01**^{a3} 0.02 - 0.11 [13%]	0.11 ± 0.01^{a3} 0.1 - 0.2	0.6 ± 0.1^{b3} 0.2 - 1.6	6.5 ± 0.3^{a1} 4.1 - 8.5
North Lake	**0.3 ± 0.04**^{c3} 0.1 - 0.8 [13%]	0.02 ± 0.01^{b3} 0.01 - 0.04	0.03 ± 0.01^{b3} 0.01 – 0.1	4.2 ± 0.5^{a2} 0.2 - 10.1	5.7 ± 0.4^{a1} 3.2 - 10.0
Bolgoda Ganga	**1.0 ± 0.2**^{b2} 0.2 - 3.0 [87%]	**0.1 ± 0.02**^{a3} 0.02 - 0.31 [13%]	0.10 ± 01^{a4} 0.03 - 0.1	0.3 ± 0.05^{c3} 0.03 - 0.8	6.7 ± 0.5^{a1} 3.7-10.4
South Lake	0.05 ± 0.01^{d3} 0.02 - 0.1	0.01 ± 0.00^{b3} 0.01- 0.02	0.02 ± 0.00^{b3} 0.01 - 0.04	1.0 ± 0.2^{b2} 0.3 - 3.1	3.6 ± 1.5^{b1} 2.5 - 4.6

*Metal levels are presented as mean ± SEM and ranges of minimum and maximum levels, n=15. In a column, data with no common superscript letters are significantly different. In a row, data with no common superscript numeral are significantly different (ANOVA, Tukey's test, $p<0.05$). Numbers in parentheses indicate percentage of samples exceeded the EU maximum allowable limits.

Table 2: Metal levels* in gills of *Etroplus suratensis* collected from four main locations of Bolgoda Lake.

Location	Metal level (µg g^{-1} wet weight)				
	Lead	Cadmium	Chromium	Copper	Zinc
Weras Ganga	8.2 ± 2.6^{a2} 1.2-20.7	0.9 ± 0.5^{a3} 0.03-3.3	0.2 ± 0.07^{a3} 0.1-0.5	1.8 ± 0.6^{b3} 1.0-4.5	23.1 ± 1.5^{a1} 17.0-27.4
North Lake	1.6 ± 0.1^{b3} 0.2-2.8	0.1 ± 0.03^{b3} 0.04-0.4	0.1 ± 0.04^{b3} 0.01-0.3	5.7 ± 1.6^{a2} 0.3-12.6	30.1 ± 3.4^{a1} 17.2-38.1
Bolgoda Ganga	2.2 ± 0.5^{b2} 0.7-4.9	0.2 ± 0.03^{b3} 0.1-0.3	0.3 ± 0.08^{a3} 0.1-0.8	1.7 ± 0.2^{b2} 0.8-2.4	14.1 ± 1.4^{b1} 9.1-19.0
South Lake	1.8 ± 1.0^{b2} 0.1-6.5	0.02 ± 0.01^{c3} 0.01-0.05	0.03 ± 0.01^{c3} 0.01-0.04	2.0 ± 0.5^{b2} 0.4-3.9	34.4 ± 4.0^{a1} 24.3-38.5

*Metal levels are presented as mean ± SEM and ranges of minimum and maximum levels, n=15. In a column, data with no common superscript letters are significantly different. In a row, data with no common superscript numeral are significantly different (ANOVA, Tukey's test, $p<0.05$).

Table 3: Metal levels* in liver tissue of *Etroplus suratensis* collected from four main locations of Bolgoda Lake.

Location	Metal level (µg g^{-1} wet weight)				
	Lead	Cadmium	Chromium	Copper	Zinc
Weras Ganga	21.4 ± 5.1^{a2} 8.8-42.1	0.9 ± 0.2^{a3} 0.3-1.6	0.6 ± 0.1^{a3} 0.1-0.9	10.3 ± 3.3^{b3} 3.5-19.8	26.0 ± 4.2^{b1} 15.9-40.6
North Lake	1.9 ± 0.6^{b3} 0.3-4.6	0.2 ± 0.05^{b3} 0.1-0.4	0.3 ± 0.1^{b3} 0.01-0.6	50.8 ± 9.6^{a2} 31.4-90.8	29.5 ± 9.5^{b1} 3.4-60.8
Bolgoda Ganga	73.5 ± 56.6^{a2} 2.3-382.6	1.5 ± 0.5^{a3} 0.3-3.6	0.7 ± 0.2^{a3} 0.1-2.2	21.7 ± 5.5^{b2} 2.5-32.4	126.2 ± 58.2^{a1} 9.1-19.0
South Lake	3.8 ± 0.7^{b2} 1.2-11.3	0.4 ± 0.3^{b3} 0.1-1.6	0.2 ± 0.1^{b3} 0.02-0.7	9.9 ± 1.5^{b2} 3.2-12.1	70.1 ± 31.3^{a1} 28.6-208.3

*Metal levels are presented as mean ± SEM and ranges of minimum and maximum levels, n=15. In a column, data with no common superscript letters are significantly different. In a row, data with no common superscript numeral are significantly different (ANOVA, Tukey's test, $p<0.05$).

Table 4. Comparison of metal levels (in g g^{-1} wet weight) in muscle, gill and liver tissues of *Etroplus suratensis* collected from the four main locations of Bolgoda Lake.

Metal	Location	Muscle	Gills	Liver
Lead	Weras Ganga	4.4^b	8.2^b	21.4^a
	North Lake	0.3^b	1.6^a	1.9^a
	Bolgoda Ganga	1.0^b	2.2^b	73.5^a
	South Lake	0.05^b	1.8^a	3.8^a
Cadmium	Weras Ganga	0.1^b	0.9^a	0.9^a
	North Lake	0.02^b	0.1^a	0.2^a
	Bolgoda Ganga	0.1^b	0.2^b	1.5^a
	South Lake	0.01^b	0.02^b	0.4^a
Chromium	Weras Ganga	0.11^b	0.2^b	0.6^a
	North Lake	0.03^b	0.1^b	0.3^a
	Bolgoda Ganga	0.1^b	0.3^b	0.7^a
	South Lake	0.02^b	0.03^b	0.2^a
Copper	Weras Ganga	0.6^b	1.8^b	10.3^a
	North Lake	4.2^b	5.7^b	50.8^a
	Bolgoda Ganga	0.3^c	1.7^b	21.7^a
	South Lake	1.0^b	2.0^b	9.9^a
Zinc	Weras Ganga	6.5^b	23.1^a	26.0^a
	North Lake	5.7^b	30.1^a	29.5^a
	Bolgoda Ganga	6.7^c	14.1^b	126.2^a
	South Lake	3.6^c	34.4^b	70.1^a

Results are presented as mean values of the metals. For each row, means not followed by the same superscript are significantly different from each other (ANOVA, Tukey's test $p<0.05$).