Accumulation Variations of selected heavy metals in *Barbus xanathopterus* in Karoon and Dez Rivers of Khuzestan, Iran

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Abstract
The concentrations of Cd, Pb, Ni and Hg were determined in gill, liver and muscle of *Barbus xanathopterus* in down streams of Karoon and Dez Rivers during summer 2009. Heavy metal concentrations varied significantly, depending on the types of the tissue in fish species. The levels of heavy metals such as Cd, Pb, Ni and Hg in the *Barbus xanathopterus* of Karoon River were higher than in *Barbus xanathopterus* of Dez River (P<0.05). In Karoon and Dez Rivers, the concentrations of Cd, Pb and Ni were in the sequence gill>liver>muscle. The concentration of Hg in Karoon River was in the sequence liver>gill>muscle but, in Dez River, it was in the sequence liver>muscle>gill. Among heavy metals (Cd, Pb, Ni and Hg), the accumulation of Pb was more than other heavy metals in fish (P<0.05). In both rivers, the accumulation of heavy metals in muscle of fish was higher than the world health organization (WHO) standard.

Keywords: Heavy metal, *Barbus xanathopterus*, Karoon, Dez, Khuzestan, Iran

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Introduction
The river systems may be excessively contaminated with heavy metals released from domestic, industrial, mining and agricultural effluents (Leland et al., 1978; Langston, 1990). The contamination of fresh waters with a wide range of pollutants has become a matter of great concern over the last few decades (Canli et al., 1998). Contamination of rivers with heavy metals may have devastating effects on the ecological balance of the aquatic environment, and the diversity of aquatic organisms becomes limited with the extent of contamination (Suziki et al., 1988). In fresh water systems, fish is one of the aquatic products that is consumed by humans and also provides a good indicator of trace element pollution (Rashed, 2001). The determination of trace metal concentration in natural water systems has received increasing attention for monitoring environmental pollution. Due to the fact that some metals are not biodegradable, their way in food chain through a number of path ways and they may accumulate in different organs of human beings or animals (Birge et al., 2000). Fish are the major part of the human diet and it is not surprising that numerous studies have been carried out on metal accumulation in different species (Lewis et al., 2002). Heavy metals like cadmium, lead, nickel and mercury are some of the major components of the industrial waters, which along with other products from industrial operations are discharged into the aquatic environment. These substances are toxic to aquatic life (Frostner and Wittman, 1983; Ibok et al., 1991). Cadmium, lead, nickel and mercury are among the most harmful metallic pollutants. Bioaccumulation of these metals is known to adversely affect liver, muscle, kidney, gill and other tissues of fish, disturb metabolism and hamper development and growth of fish (Korai et al., 2008; Dallinger et al., 1987). The gills are the first organs to be exposed to resuspended sediment particles, so they can be significant sites of interaction with metal ions (Pawert et al., 1998). Gills are the uptake site of waterborne ions, where metal concentrations increase especially at the beginning of exposure, before the metal enters other parts of the organism (Kamaruzzaman et al., 2010). In contrast, the muscle tissues are not considered an active site for metal accumulation but it plays an important role in human’s feeding (Romeo et al., 1999). Fish which generally accumulate contaminants from aquatic environments have been largely used in food safety studies. The commercial and edible species have been investigated in order to check for those hazardous to human health. Metals can be taken up by fish from water, food, sediments and suspended particulate material (Agusa et al., 2005). Karoon-Dez River basin, (10° 48’- 30° 52’ E, 20° 30’- 05° 34’ N), is located in the southern part of Iran. Karoon River is one of the largest rivers in Iran and has an area of 60500 Km² and an average annual discharge of 18700 million m³, also Dez River is one of the constant rivers in Iran and has an area of 21100 Km² and an average annual discharge of 7396 million m³. They play an important role in water and fish supply. Barbus
xanthopterus has high market value and is one of the main fish products in Karoon and Dez Rivers. Considering the complexity of heavy metal bioaccumulation of fishes, it was important to study the heavy metal accumulation in different commercial fishes such as B. xanthopterus in Karoon and Dez Rivers for the food safety. The main objective of this study was to determine the contents of heavy metals in different tissues of B. xanthopterus which were collected from downstreams of Karoon and Dez Rivers. This could help us understand the enrichment behavior of heavy metals in downstreams of Karoon and Dez Rivers and emphasize the need to discard the most polluted tissues of the fish.

Materials and methods
The concentration of heavy metals such as cadmium, lead, nickel and mercury were measured in the muscle, gills and liver of B. xanthopterus, collected by gillnet from downstreams of Karoon (Shekariyeh 3 village) and Dez (Ali abad village) Rivers in Khuzestan province during summer 2009. The number of samples was 48 fish in each river. After capture, fish were placed in plastic bags and transported to the laboratory in freezer bags with ice. The length and weight were measured which ranged from minimum and maximum value as 248.5 and 377.5mm for length and 173 and 561.3 g for weight of B. Xanthopterus in Karoon and Dez Rivers. After biometry, fish were immediately frozen at -20°C. All samples were cut into pieces and labeled, and then all sampling procedures were carried out according to internationally recognized guidelines (UNEP, 1991). Fish samples for heavy metals were put onto a dissection tray and thawed at room temperature. They were dissected using stainless steel scalpels and Teflon forceps using a laminar flow bench. In parallel gill, liver and a part of the muscle (dorsal muscle without skin) were removed and transferred in polypropylene vials. Subsequently, samples were put into an oven to dry at 90°C and reached constant weights in the oven. Before acid digestion, a porcelain mortar was employed to grind and homogenize the dry tissue samples. Aliquots of approximately 1g dried gill, liver and muscle were digested in Teflon beakers for 12 h at room temperature, and then for 4h at 100°C with 5 ml ultrapure nitric acid (65%, Merck).

Sample analysis
Heavy metals analysis: Cd, Ni and Pb were measured by graphite furnace atomic absorption spectrophotometry (Perkin-Elmer, 4100 ZL). Hg concentrations were determined with a Perkin–Elmer MHS-FIAS coupled to a Perkin–Elmer 4100 ZL spectrophotometer. Results are expressed as mgkg⁻¹ dry weight. The analytical procedure was checked using reference material [MESS-1, the National Center of Canada and CRM 277, the Community Bureau of Reference, Brussels, Belgium, and details were in (Robisch and Clark, 1993; Meador et al., 1994)]. For each matrix, analyses of three blank samples were performed along with the samples. Quality control was assured by the analysis of reagent blank and procedural blanks. Data statistics were performed using SPSS 17 software.
Figure 1: Heavy metal concentrations in muscle, gill and liver of *Barbus xanthopterus* in Karoon River

Figure 2: Heavy metal concentrations in muscle, gill and liver of *Barbus xanthopterus* in Dez River

Figure 3: Heavy metal concentrations in gill of *Barbus xanthopterus*
Table 1: The concentrations of heavy metals (mg kg\(^{-1}\) dry weight) in various tissues of *Barbus xanthopterus* in Karoon and Dez Rivers, Khuzestan, Iran, summer 2009

<table>
<thead>
<tr>
<th>Tissues</th>
<th>River</th>
<th>Cd</th>
<th>Pb</th>
<th>Ni</th>
<th>Hg</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Karoon</td>
<td>2.173±0.1089(^a)</td>
<td>2.918±0.480(^a)</td>
<td>1.4388±0.0256(^a)</td>
<td>1.3422±0.1003(^a)</td>
</tr>
<tr>
<td>Gill</td>
<td>Dez</td>
<td>1.947±0.0833(^b)</td>
<td>1.406±0.1110(^b)</td>
<td>0.8550±0.0342(^b)</td>
<td>0.6331±0.0213(^b)</td>
</tr>
<tr>
<td></td>
<td>Karoon</td>
<td>1.9197±0.0491(^a)</td>
<td>2.789±0.474(^a)</td>
<td>1.3078±0.0704(^a)</td>
<td>1.4269±0.0811(^a)</td>
</tr>
<tr>
<td>Liver</td>
<td>Dez</td>
<td>1.046±0.0726(^b)</td>
<td>1.266±0.1110(^b)</td>
<td>0.75188±0.01281(^b)</td>
<td>0.9588±0.0256(^b)</td>
</tr>
<tr>
<td></td>
<td>Karoon</td>
<td>1.6781±0.0555(^a)</td>
<td>2.370±0.429(^a)</td>
<td>1.1597±0.0833(^a)</td>
<td>1.2894±0.0640(^a)</td>
</tr>
<tr>
<td>Muscle</td>
<td>Dez</td>
<td>0.7994±0.0640(^b)</td>
<td>0.9550±0.1025(^b)</td>
<td>0.59844±0.00640(^b)</td>
<td>0.7741±0.1473(^b)</td>
</tr>
</tbody>
</table>

Data are presented as means ± S.E. of Karoon and Dez Rivers: \(^a\), \(^b\): P<0.05, significantly different in each tissue between two rivers.
Table 2: Comparison of heavy metals (mg kg\(^{-1}\)) in some studies with present study

<table>
<thead>
<tr>
<th>Species</th>
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</tr>
<tr>
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<td>-</td>
<td>Alhas et al. (2009)</td>
</tr>
<tr>
<td></td>
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<td>-</td>
<td>0.68</td>
<td>0.08</td>
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<td>-</td>
<td>-</td>
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<tr>
<td></td>
<td>Liver</td>
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<td>-</td>
<td>-</td>
<td>-</td>
<td>Alhas et al. (2009)</td>
</tr>
<tr>
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<td>-</td>
<td>0.66</td>
<td>0.04</td>
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<td>Tor grypus</td>
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<td>0.35</td>
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<td>Muscle</td>
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<td>2.98</td>
<td>0.17</td>
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<td>Labeo rohita</td>
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<td>-</td>
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<tr>
<td></td>
<td>Liver</td>
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<td>0.529</td>
<td>1.263</td>
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<tr>
<td></td>
<td>Muscle</td>
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<td>0.427</td>
<td>0.393</td>
<td>0.207</td>
<td>Malik et al. (2010)</td>
</tr>
<tr>
<td>Ctenopharyngodon</td>
<td>Gill</td>
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<td>0.393</td>
<td>1.63</td>
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<td>-</td>
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<tr>
<td>idella</td>
<td>Liver</td>
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<td>Erdogru (2007)</td>
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</tr>
<tr>
<td>Silurus glanis</td>
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<td>-</td>
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<td></td>
<td>Liver</td>
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<td>Erdogru (2007)</td>
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<tr>
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<td>-</td>
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</tr>
<tr>
<td>Elops lacerta</td>
<td>Gill</td>
<td>-</td>
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</tr>
<tr>
<td></td>
<td>Liver</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Ekpo &amp; Ibok (1999)</td>
</tr>
<tr>
<td></td>
<td>Muscle</td>
<td>-</td>
<td>0.01</td>
<td>0.02</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Psettias sebae</td>
<td>Gill</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
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</tr>
<tr>
<td></td>
<td>Liver</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Ekpo &amp; Ibok (1999)</td>
</tr>
<tr>
<td></td>
<td>Muscle</td>
<td>-</td>
<td>0.03</td>
<td>0.01</td>
<td>-</td>
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</tr>
<tr>
<td>Liza parida</td>
<td>Gill</td>
<td>-</td>
<td>-</td>
<td>-</td>
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</tr>
<tr>
<td></td>
<td>Liver</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>Saha et al. (2006)</td>
</tr>
<tr>
<td></td>
<td>Muscle</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.54</td>
<td>-</td>
</tr>
<tr>
<td>Barbus xanthurerus</td>
<td>Gill</td>
<td>2.1734</td>
<td>2.918</td>
<td>1.4388</td>
<td>1.3422</td>
<td>Present study (2009)</td>
</tr>
<tr>
<td>(Karoon)</td>
<td>Liver</td>
<td>1.9197</td>
<td>2.789</td>
<td>1.3078</td>
<td>1.4269</td>
<td>Present study (2009)</td>
</tr>
<tr>
<td></td>
<td>Muscle</td>
<td>1.6781</td>
<td>2.370</td>
<td>1.1597</td>
<td>1.2894</td>
<td>Present study (2009)</td>
</tr>
<tr>
<td>Barbus xanthurerus</td>
<td>Gill</td>
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<td>1.4063</td>
<td>0.8550</td>
<td>0.6331</td>
<td>Present study (2009)</td>
</tr>
<tr>
<td>(Dez)</td>
<td>Liver</td>
<td>1.0406</td>
<td>1.2663</td>
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<tr>
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<td>Muscle</td>
<td>0.7994</td>
<td>0.9550</td>
<td>0.59844</td>
<td>0.7741</td>
<td>Present study (2009)</td>
</tr>
</tbody>
</table>
Table 3: Comparison of heavy metals (mg kg\(^{-1}\) dry weight) in muscle of *Barbus xanthopterus* with WHO standard in Karoon and Dez Rivers, Khuzestan, Iran

<table>
<thead>
<tr>
<th>Tissue</th>
<th>River</th>
<th>Cd</th>
<th>Pb</th>
<th>Ni</th>
<th>Hg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Muscle</td>
<td>Karoon</td>
<td>1.6781±0.0555(^{a})</td>
<td>2.370±0.429(^{a})</td>
<td>1.1597±0.0833(^{a})</td>
<td>1.2894±0.0640(^{a})</td>
</tr>
<tr>
<td></td>
<td>Dez</td>
<td>0.7994±0.0640(^{b})</td>
<td>0.9550±0.1025(^{b})</td>
<td>0.5984±0.00640(^{b})</td>
<td>0.7741±0.1473(^{b})</td>
</tr>
<tr>
<td>WHO</td>
<td></td>
<td>0.2(^{c})</td>
<td>0.3(^{c})</td>
<td>0.3(^{c})</td>
<td>0.1(^{c})</td>
</tr>
</tbody>
</table>

a, b, c: P<0.05, significantly different in muscle of *Barbus xanthopterus* with WHO standard.


Results

Heavy metal concentrations in various tissues of *B. xanthopterus* in Karoon and Dez Rivers decreased in the sequence of Pb> Cd> Hg> Ni, but heavy metal concentrations in gill of *B. xanthopterus* in Karoon and Dez Rivers decreased in the sequence of Pb> Cd> Ni> Hg (Fig. 1 and 2).

The comparison of heavy metal levels (Cd, Pb, Ni, Hg) in various tissues of *B. xanthopterus* between Karoon and Dez Rivers showed that heavy metal levels in *B. xanthopterus* of Karoon River were higher than in *B. xanthopterus* of Dez River (P<0.05) (Fig. 3-5). The levels of Cd, Pb, Ni and Hg in various tissues of *B. xanthopterus* in Karoon and Dez Rivers are given in Table 1.

Discussion

In this study, the concentration of heavy metals in gill were higher than those in other tissues, except for Hg which was high in liver (P<0.05). The concentration of heavy metals in muscle were lower than in other tissues, except for Hg in *B. xanthopterus* in Dez River which was the least in gill (P<0.05). In this research, heavy metal concentrations (Cd, Pb, Ni and Hg) in various tissues of *B. xanthopterus* in Karoon River was higher than in *B. xanthopterus* in Dez River (P<0.05) and the concentration of heavy metals, differed significantly in each tissue of *B. xanthopterus* between Karoon and Dez Rivers (P<0.05), (Table 1). Heavy metal concentrations varied significantly depending upon the type of fish tissues and locations. Alhas et al. (2009) reported that in *Barbus xanthopterus* and *Barbus rajanorum mystaceus* in Ataturk Dam Lake, Turkey, heavy metal concentrations in gill and liver were maximum, while these concentrations were the least in muscle. Oymak et al. (2009) has reported the concentrations of Co, Cr, Cu, Fe, Mn, Ni, Pb and Zn in gill, liver, muscle and kidney of *Tor grypus* in Ataturk Dam Lake, Turkey. Malik et al. (2010) determined the concentrations of Pb, Cd, Zn, Ni, Cu, Cr and Hg in gill, liver and muscle of *Labeo rohita* and *Ctenopharyngodon idella* in the Lake of Bhopal, India. In different reports it was showed that the concentrations of heavy metals in liver and gill were higher than...
muscle. The results of this study were similar to the above studies (Table 2). Thus, heavy metals when discharged into the rivers enter the food chain and accumulate in the fish body as determined during this investigation. In this study the obtained results showed that heavy metal concentrations in *B. xanthopterus* in two rivers were more than the WHO (World Health Organization, 1985, 1994) standard. The concentrations of heavy metals have significant differences (P<0.05) in muscle of *B. xanthopterus* with WHO standard in Karoon and Dez Rivers (Table 3). In Sir Dam Lake in Turkey, the mean Hg concentration in the muscle of *Cyprinus carpio* and *Siluris glanis* was reported as: 0.07 µg g⁻¹ and 0.26 µg g⁻¹, respectively (Erdogrul, 2007). The values recorded for Cd and Pb in the muscles of *Elops lacerta* and *Psettias sebae* in Calabar River of Nigeria, were 0.01, 0.02 µg g⁻¹; and 0.03, 0.01 µg g⁻¹ respectively (Ekpo and Ibok, 1999). The mean of Ni and Pb concentrations in the muscles of *Barbus xanthopterus* and *Barbus rajanorum* in Ataturk Dam Lake of Turkey, were reported as: 0.08, 0.68 µg g⁻¹; 0.04, 0.66 µg g⁻¹, respectively (Alhas et al., 2009) and in Sunderban mangrove wetland of northeast India, the mean Hg concentration in the muscle, liver and gill of *Liza parsia* was reported as: 0.12, 0.65 and 0.54 µg g⁻¹ respectively (Saha et al., 2006), a comparison of these concentrations with our data clearly shows higher values in *B. xanthopterus* in Karoon and Dez Rivers (Table 2). Differences in ecological needs, metabolism and feeding patterns of fish and also the season in which studies were carried out. In the river, fish are often at the top of the food chain and have the tendency to concentrate heavy metals from water (Mansour and Sidky, 2002). The results from this research indicated that metal accumulation depended on the tissues probably as a consequence of metabolic needs, physiochemical properties, and detoxification processes specific for each element. The results of this study showed that Pb was the highest accumulating metal compared to other metals (P<0.05), Ahmad et al. (2010) reported that among the five metals (Pb, Cd, Ni, Cu and Cr) studied, Pb concentration was the highest in *Gudusia chapra* of Buriganga River, Bangladesh. So, the result of our study was similar to the above result. The Pb finds its way in rivers through the discharge of industrial waste waters, such as from painting, dyeing, battery manufacturing units and oil refineries etc. Pb also enters the rivers both from terrestrial sources and atmosphere and the atmospheric input of Pb aerosols can be substantial (Mitra et al., 2010). The high levels of Pb in the Karoon and Dez Rivers have toxic effects on fish metabolism and it is important to consider the biological effects of contamination on fish health in Karoon and Dez Rivers.

This study was carried out to provide information on heavy metal concentrations in *B. xanthopterus* in Karoon and Dez Rivers in Khuzestan, Iran. All results were higher than the limits for fish proposed by the WHO (Table 3). High levels of heavy metals were found in the gill and liver of *B. xanthopterus* in Karoon and Dez Rivers while the lowest levels of *B. xanthopterus* in Karoon River were found in the muscle and in the Dez River they were found in...
the gill. The concentrations of heavy metals in _B. xanthopterus_ in Karoon River were higher than in _B. xanthopterus_ in Dez River (P<0.05). Research conducted by this paper found high levels of Pb in _B. xanthopterus_ in Karoon and Dez Rivers (P<0.05), that could be traced to urban and industrial places near the Karoon and Dez Rivers and their wastes that are discharged into these rivers.

**Acknowledgements**

We would like to thank Islamic Azad University, Khuzestan Science and Research Branch that have supported us through the process of developing my thesis.

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