## Bioaccumulation of heavy metals (Zn, Cu, As, Cd) in external and internal organs of black pomfret (*Parastromateus niger*) in the northwest of Persian Gulf

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## Abstract

The major objective of this study was to investigate heavy metal content of black pomfret (Parastromateus niger). The concentrations of Zn, Cu, As and Cd were determined in external (gill and shell) and internal organs (muscle and liver) of black pomfret in northwest of Persian Gulf in summer 2013. A total of 60 specimens were collected from fishing bottom trawlers. Concentrations of these heavy metals were measured by using atomic absorption spectrometer. Results showed that heavy metals concentrations varied significantly, depending on the types of the tissue in fish. Bioaccumulation of heavy metals Zn and Cu were in the sequence liver> shell> gill> muscle but for Cd and As were in the sequence of gill> shell> liver> muscle. In total organs and among heavy metals the accumulation of Zn and As were the highest and the lowest (P< 0.05). The results indicated that the gill of fishes were highly contaminated by heavy metals and exceeded of WHO and NHMRC guidelines for As and Cd, respectively.

**Keywords:** *Parastromateus niger*, Bioaccumulation, Heavy metals, Persian Gulf.

### Introduction

The environment is continuously loaded with heavy metals released by urban communities and indus-

**Correspondence** L Roomiani, Department of Fisheries, Ahvaz Branch, Islamic Azad University, Ahvaz, Iran (e-mail: L.roomiani@yahoo.com) tries. Environmental pollution containing hazards of metals contamination represents a major problem both in developed and developing countries. Heavy metals in aquatic environment is hazardous because once the metals enter the aquatic environmental they cannot be destroyed rather they change from one from to another persisting in the aquatic environment (Bandowe, Bigalke, Boamah, Nyarko, Saalia & Wilcke 2014).

Heavy metal is introduced to marine environment through natural and anthropological activities. This element may sink toward the bottom sediment or accumulate directly in marine ecosystem. The accumulation process is affected by different environmental and biological factors such as salinity, temperature, season, size, sex and species. Heavy metals enter aquatic organisms through skin, gill and food (El- Moselhy, Othman, El- Azem & El-Metwally 2014).

The accumulation of toxic risks and strict periodic surveillance of these contaminants is therefore advisable. Toxic metals pollution of the sea water is less visible and direct than other types of marine pollution but its effects on marine ecosystems and humans are intense and very extensive (Velusamy, Kumar, Ram & Chinnadurai 2014).

Fish are notorious for their ability to concentrate heavy metals in their muscles and they also play important role in human nutrition. Therefore, it is important to carefully screen the bioaccumulation factor and exposure concentration of heavy metal to ensure that unnecessary elevated level of some toxic metals is not being transferred to human through fish consumption i.e., health risk as food to human. The presence of metal in aquatic environment may lead to accumulation of metals in aquatic organisms through different mechanism: via the direct uptake from water through gill or skin (bioconcentration), via the uptake of suspended particles (ingestion) and via the consumption of contaminated food (bio-magnification) (Burger, Gochfeld, Jeitner, Pittfield & Donio 2014).

Recent attention has been paid in developed and developing countries on the subject of anomalous distribution of metals in the water, sediments and fishes which are fundamentally important for understanding the behavior of the metals and also the swapping between the sediments and the water column (Yang, Zhang & Wang 2014).

The aim of the present study was to assess the heavy metals (As, Cu, Cd, Zn) in one of the most commercial fish, black pomfret (*Parastromateus niger*) and to determine whether these metals are within permissible limits for human consumption.

## **Materials and Methods**

Sampling site was selected along the Arvand River, northwest coasts of the Persian Gulf (Khuzestan Province, Choebdeh) (Fig. 1). The Arvand River, located in the border between Iraq and Iran, is the biggest river in the Persian Gulf.

The fish specimens were collected from fishing bottom trawlers and then were placed in plastic bags and transported to the laboratory in ice box. 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 international guidelines (UNEP 1991). Fork length (mm) and weight (g) were recorded for fishes.

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, shell 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 (USEPA 1996). Before acid digestion, a porcelain mortar was employed to grind and homogenize the dry tissue samples. Aliquots of approximately 1 g dried gill, liver, shell 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).

Heavy metals analysis: Cd, Cu, Zn and As were measured by graphite furnace atomic absorption spectrophotometry (Perkin-Elmer, 4100 ZL). 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 (Leung, Leung, Wang, Ma, Liang, Ho, Cheung, Tohidi & Yung 2014).

The whole data were subjected to a statistical analysis and correlation matrices were produced to examine the inter-relationships between the investigated trace metal concentrations of the samples. Data statistics were performed using SPSS 20 software. Paired samples T Test were used to compare differences between samples. A P-value less than 0.05 were considered statistically significant (Hakimimofrad & Hakimimofrad 2014).

## Results

Their mean weight of fish specimen was  $545\pm 30.00$  g and mean length  $30.5\pm 4.20$  cm FL. The concentration of heavy metals expressed as mg/100g dry weight for Zn and Cu and ppb for Cd and As in liver, shell, gill and muscle samples is shown in Fig. 2. The results confirm the differences of heavy metal accumulation in the tissues. It is apparent that all samples are contaminated with different levels of heavy metals and metal concentrations in livers of examined species were generally higher than those in muscles.

Overall the sequence of metal accumulation in external and internal organs of black pomfret was Zn> Cu> Cd> As. There were significant differences in metal concentrations between external and internal organs (P< 0.05).

The distribution patterns of Zn in organs of black pomfret follows the order: liver> shell> gill> muscle. The highest and the lowest concentration of Zn  $(2.26\pm0.09 \text{ mg}/100\text{ g} \text{ and } 0.84\pm0.02 \text{ mg}/100\text{ g})$  were observed in liver and muscle. There were no signifi-

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Standards	Zn	Cu	Cd	As	Reference
WHO1	100	30	0.2	0.02	WHO 1996
FDA2	-	-	2.0	-	Chen & Chen 2001
UK (MAFF)3	50	20	0.2	1.00	MAFF 1995
NHMRC4	150	10	0.05	1.00	Chen & Chen 2001
FAO5	30	30	0.5	7.88	FAO 1983

Table 1 The tolerable values of some heavy metals in the fish (mg/kg)

1- World Health Organization, 2- U.S. Food and Drug Administration, 3- Ministry of Agriculture, Fisheries & Food (UK), 4- National Health & Medical Research Council (Australia), 5- Food and Agriculture Organization.

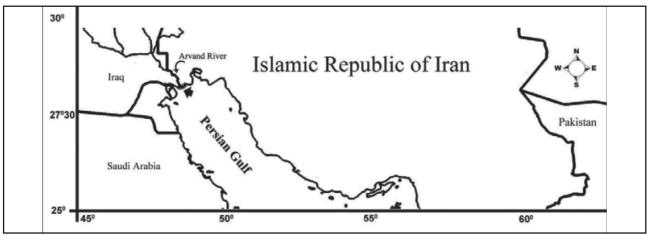


Figure 1 Map of study area.

cant differences in accumulation of Zn in liver, gill and shell (P> 0.05) while observed for muscle (P< 0.05). A positive interaction was observed between Zn and accumulation it in liver (R=0.4).

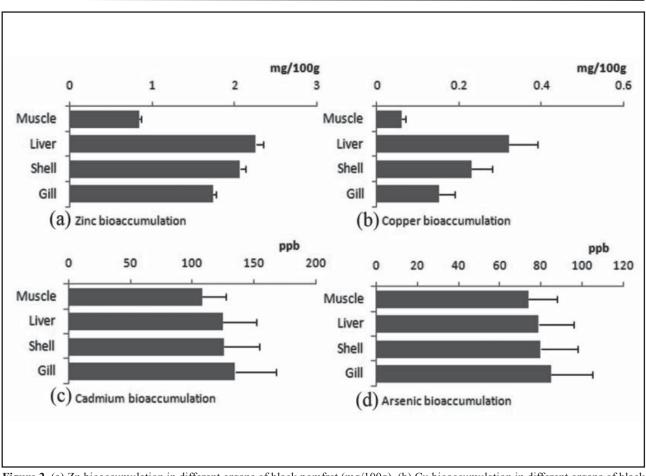
The highest concentration of Cu  $(0.32\pm0.07 \text{ mg/100g})$  was observed in liver and the lowest of muscle  $(0.06\pm0.01 \text{ mg/100g})$ . The sequence of Cu in organs of black pomfret was liver> shell> gill> muscle. There was significant difference in organs of fish for Cu (P< 0.05). A positive correlation was observed between of bioaccumulation Cu and liver (R= 0.2).

Results showed that no correlation between of metals in external and internal of fish for Cd and As. The highest and the lowest of bioaccumulation of Cd and As were in gill ( $135\pm33.44$  and  $85\pm20.03$ ) and muscle ( $108\pm19.33$  and  $74\pm13.73$  ppb), respectively.

The level of the taken into consideration metal were shown significant differences between the tissues throughout in black pomfret. In this study, the concentration of heavy metals in gill were higher than those in other tissues for As and Cd, but for Zn and Cu which was high in liver (P < 0.05). The concentration of heavy metals in muscle were lower than in other tissues (P < 0.05).

#### Discussion

Fishes have been successfully used as accurate indicator organisms for environmental monitoring programmes because they possess numerous advantages (Gamito, Pasquaud, Courrat, Drouineau, Fonseca, Goncalves, Wouters Costa, Lepage, Costa & Cabral 2012; Sheaves, Johnston & Connolly 2012; Alvarez, Franco, Perez-Dominguez & Elliott 2013) which include: (1) they are typically present in all aquatic systems; (2) there is extensive life-history and environmental response information available for most species; (3) fish communities usually include a range of species that represent a variety of trophic levels and include foods of both aquatic and terrestrial origin; (4) they are comparatively stable and therefore provide a long-term record of environmental stress; (5) they contain many life forms and functional guilds and thus are likely to cover all



**Figure 2** (a) Zn bioaccumulation in different organs of black pomfret (mg/100g), (b) Cu bioaccumulation in different organs of black pomfret (mg/100g), (c) Cd bioaccumulation in different organs of black pomfret (ppb), (d) As bioaccumulation in different organs of black pomfret (ppb).

components of aquatic ecosystems affected by anthropogenic disturbance; and (6) they are both sedentary and mobile and thus reflect stressors within one area as well as providing scientists to give a broader assessment of effects. Therefore, the elucidation of heavy metal levels in the fish species investigated in this study provided an indication of the current environmental conditions of the northwest of Persian Gulf.

This investigation showed higher accumulation of Zn and Cu in tissue of black pomfret as compared to As and Cd. On the other hand, the external organs in accumulation of Cd and As have effective than internal organs in comparison to Zn and Cu. It is generally related to physiological activities of organs, for instance Zn and Cu are involved in enzyme activities in liver consequently higher quantities of these elements are accumulated in liver. It is well documented that levels of heavy metals vary in fish depending on factors such as habitat, migration, age, ecological needs, size, length fish, metabolism and feeding activities (Subotic, Spasic, Visnjic-Jeftic, Hegedis, Krpo-Cetkovic, Mickovic, Skoric & Lenhatdt 2013). In study of Askary Sary, Velayatzadeh & Beheshti (2012) results showed that the distribution patterns of heavy metals (Zn, Cd, Cu, Fe, Hg, Mn) in tissue of Liza abu in northern of Persian Gulf follows the order: gill> liver> muscle. Heavy metal concentrations were higher in gill and liver comparing to muscle tissue.

Also the results showed that the heavy metal concentrations were higher in the gill and liver, when compared with muscle. In different reports (Karadede & Ünlü 2000; Karadede, Oymak & Unlu 2004; Alhas, Oymak & Akin 2009; Askary Sary & Moammadi 2011; Copta, Arena, Ledda, Fallico, Sciacca & Ferrante 2013) it was showed that the concentrations of heavy metals in gill, shell and liver were higher than muscle. The metal accumulation depended on the tissues probably as a consequence of metabolic needs, physiochemical properties, and detoxification processes specific for each element. Fish generally accumulate contaminants from aquatic environments, have been largely used in food safety studies. Muscles are often examined for metal content due to their use for animal and human consumption. They were also implicated in long term metal storage in fish organisms.

Gills and livers were chosen as target organs for assessing metal accumulation. Metal concentration in the gills could be due to the element complexion with the mucus, which is impossible to remove completely from between the lamellae, before tissue is prepared for analysis. Thus high concentration of various metals can be observed there. The absorption of metals on to the gill surface, as the first target for pollutants in water, could also be an important influence in the total metal levels of the gill. Studies have shown that muscle is not an active tissue in accumulating heavy metals. This may reflect the low levels of metal-lothionein, low molecular weight binding proteins, in the muscle. Metals that enter the body via food are carried by the blood bound to 256 proteins, where they move first move into the liver and gradually into the muscle tissues (Authman, Abbas & Abbas 2013).

The accumulation of metals in the liver could be due to the greater tendency of the elements to react with the oxygen carboxylate, amino group, nitrogen and/or sulphur of the mercapto group in the metal-lothionein protein, whose concentration is highest in the liver. The liver has the ability to accumulate large quantities of pollutants from the external environment and also plays an important role in storage, redistribution, detoxification and transformation of pollutants. The liver accumulates high concentration of metal regardless of the uptake route. It is one of the major sites of metal metabolism and detoxication in fish, and it is considered as a good monitor of water pollution (Souza, Duarte, Pimentel, Rocha, Morozesk, Bonomo, Azevedo, Pereira, Monferran, Milanez, Matsumoto, Wunderlin & Fernandes 2013).

In this study the metals Zn and As were the highest and lowest in tissues of black pomfret. Distribution patterns of metal concentrations in the gill, liver, shell and muscle of black pomfret follows the sequence: Zn> Cu> Cd> As. The results of Meng, Li & Wu (2014) indicated significant differences in metals concentrations were found between the tissues of fish. Cd was mainly distributed in the gill of the fish. Heavy metals (Pb, Cd, Ni, Cr, Cu, Zn, Mn, and As) concentration was investigated in the industrial effluents, water, sediment, and fish samples collected around the Dhaka Export Processing Zone, Savar, Bangladesh, to evaluate the level of contamination. The Pearson correlation analysis showed significant correlations (P < 0.01 and P <(0.05) between most of the metals in the samples of effluents, water, sediments, and fish muscles (Rahman, Saha, Molla & Al-Reza 2014). According to Leung et al. (2014) the highest concentration of heavy metals is related to Zn (15.7-29.5), therefore Cu (0.79-2.26), As (0.03-1.53) and Cd (0.02-0.06) mg/kg. Concentrations of the heavy metals copper (Cu), cadmium (Cd), zinc (Zn), lead (Pb) and nickel (Ni) were determined in the liver, gills and muscles of tilapia fish from the Langat River and Engineering Lake, Bangi, Selangor, Malaysia. There were differences in the concentrations of the studied heavy metals between different organs and between sites. In the liver samples sequences of metals were Cu> Zn> Ni> Pb> Cd and in the gills and muscle Zn > Ni > Cu > Pb > Cd (Taweel, Shuhaimi-Othman & Ahmad 2013).

According to the Priority List of Hazardous Substances established by the Agency for Toxic Substances and Disease Registry (ATSDR 2013), the descending order of heavy metals threatening to human health were As> Pb> Cd> Ni> Zn> Cr> Cu> Mn. Arsenic levels in analyzed fish samples were found to be higher than legal limit and have significant differences (P<0.05) in fish samples with WHO legal limit.

Zinc and copper levels in analyzed fish samples were found to be lower than legal limits. The maximum cadmium level permitted in fish is 0.2 mg/kg for WHO (1996), 0.5 mg/kg for FAO (1983), 0.2 for MAFF (1995), 0.05 mg/kg for NHMRC (1990) and 2.0 mg/kg for FDA (1996) (Table 1). Generally, in this research cadmium levels in fish from marine water were found to be higher than legal limits and have significant differences (P<0.05) in fish samples with NHMRC legal limits.

It is recommended that heavy metal concentrations in different fish species must be determined on a regular basis in the future so as to reduce human health risks from acute and chronic food intoxication.

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# مقایسه تجمع زیستی فلزات سنگین (روی، مس، آرسنیک، کادمیوم) در اندامهای خارجی و داخلی حلوا سیاه در شمال غرب خلیجفارس

لاله رومیانی "»، منصوره قائنی '، امیر اقبال خواجه رحیمی ۲ ۱ گروه شیلات واحد اهواز، دانشگاه آزاد اسلامی، اهواز، ایران ۲ گروه شیلات واحد تهران شمال، دانشگاه آزاد اسلامی، تهران، ایران

#### چکیدہ

هدف از این مطالعه بررسی فلزات سنگین ماهی حلوا سیاه (Parastromateus niger) بود. غلظت روی، مس، آرسنیک و کادمیوم در اندامهای خارجی (آبشش و پوست) و داخلی (عضله و کبد) ماهی حلوا سیاه در شمال غرب خلیج فارس در تابستان ۱۳۹۲ تعیین شد. ۶۰ نمونه ماهی با استفاده از ابزار صیادی ترال کف جمع آوری شدند. غلظت فلزات با استفاده از دستگاه جذب اتمی اندازه گیری شدند. نتایج نشان داد که بسته به نوع بافت غلظتهای فلزات تفاوت معنیدار نشان دادند. تجمع زیستی فلزات ساتگین روی و مس به ترتیب کبد> پوست> آبشش» عضله اما برای کادمیوم و آرسنیک آبشش> پوست> کبد> عضله بود. در تمام اندامها و در میان فلزات سنگین روی و آرسنیک به ترتیب بیشترین و کمترین تجمع را داشتند (۲۰۰<pp). نتایج ثابت کرد که فلزات آرسنیک و کادمیوم در آبشش ماهیان از حد مجاز سازمان بهداشت جهانی و تحقیقات ملی بهداشت و داروی استرالیا بیشتر بود. **و اژههای کلیدی:** حلوا سیاه، تجمع زیستی، فلزات سنگین، خلیجفارس.

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