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BIOACCUMULATION OF COPPER, ZINC, MANGAN, IRON AND MAGNESIUM IN SOME ECONOMICALLY IMPORTANT FISH FROM THE WESTERN SHORES OF ANTALYA

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ABSTRACT

Bioaccumulation of copper, zinc, manganese, iron and magnesium which generaly result from agricultural activities was determined in muscle, skin and gills of some economically important sea fish (*Diplodus sargus*, *Siganus rivulatus*, *Lithognathus mormyrus*, *Liza aurata*, *Chelon labrasus*) from the western shores of Antalya, intensive agricultural regions. The minimum and maximum levels of investigated elements (*mg kg^{-1} wet weight*) in tissues of the fish varied from 0.54 to1.69 for copper; from 4.14 to 407.23 for zinc from 0.15 to 9.17 for mangan; from 3.45 to104.49 for iron; and from 204.33 to 784.30 for magnesium. While the lowest levels of elements were determined in the muscle, the highest levels (except copper) were encountered in the gills. Copper and zinc levels in muscle of the species were remarkably lower than the maximum permissible levels informed by World Health Organization (WHO), Food and Agriculture Organisation **(**FAO) and Turkish Legislation although zinc contents in the skin of some species (*D. sargus*, *S. rivulatus*, *L. mormyrus* and *L. aurata*) were higher than that levels.

Keywords: Heavy metal, Sea fish, Accumulation, ICP-OES.

ANTALYA'NIN BATI SAHİLLERİNDEN YAKALANAN EKONOMİK ÖNEMİ OLAN BAZI DENİZ BALIKLARININ BAKIR, ÇİNKO, MANGAN, DEMİR VE MAGNEZYUM BİYOAKÜMÜLASYONU

ÖZ

Yoğun zirai faaliyetlerin yapıldığı Antalya'nın batı sahillerinden yakalanan ekonomik önemi olan bazı deniz balıklarının (*Diplodus sargus*, *Siganus rivulatus*, *Lithognathus mormyrus*, *Liza aurata*, *Chelon labrasus*) kas, deri ve solungaçlarındaki bakır, çinko, mangan, demir ve magnezyum (Genellikle zirai aktivitelerden kaynaklanırlar) biyoakümülasyon seviyeleri araştırılmıştır. Balıkların dokularında incelenen elementlerin minimum ve maksimum seviyeleri (mg kg^{-1} yaş ağırlık) bakır için 0.54-1.69; çinko için 4.14-407.23; mangan için 0.15-9.17; demir için 3.45-104.49 ve magnezyum için 204.33- 784.30 olarak tespit edilmiştir. İncelenen elementlerin en düşük biyoakümülasyon seviyeleri kas dokularda, en yüksek seviyeler (Bakır hariç) ise solungaçlarda tespit edilmiştir. İncelenen türlerin kas dokularındaki bakır ve çinko seviyeleri (Dünya Sağlık Örgütü (WHO), Gıda ve Tarım Örgütü (FAO) ve Türk Gıda Kodeksi'nde belirtilen kabul edilebilir maksimum seviyelerden daha düşük, bazı türlerin (*D. sargus*, *S. rivulatus*, *L. mormyrus* and *L. aurata*) derilerinde bulunan çinko seviyeleri ise kabul edilebilir maksimum seviyelerden daha yüksek bulunmuştur.

Anahtar Kelimeler: Ağır metal, Deniz balıkları, Akümülasyon, ICP-OES.

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1. INTRODUCTION

A wide range of contaminants are continuously introduced into the aquatic environment mostly associated with industrial, agricultural and domestic wastes run-off (Lima et al., 2008). Among these contaminants, heavy metals constitute one of the main dangerous groups, because they are toxic, persistent and not easily biodegradable. Metals may exert beneficial or harmful effects on life, depending on their concentration. Some heavy metals like mercury, cadmium and lead have no known role in aquatic organisms while some others such as copper, zinc, iron and manganese are essential for fish metabolism. For the normal metabolism of fish, the essential metals must be taken up from water, food or sediment (Canli and Atli, 2003). These essential metals always function in combination with organic molecules, usually proteins. But the essential metals being above the threshold bioavailable level are also toxic to aquatic organisms and humans (Kucuksezgin et al., 2006). Fish and their some tissues can also be considered as the most significant indicators in water systems for the estimation of metal pollution level (Henry et al., 2004). For instance, it was informed that zinc concentration in gill of *Liza saliens* can be used as environmental indicators of metal stress (Fernandes et al., 2007). For these reasons, it is important to determine the concentrations of heavy metals in fish in order to evaluate the possible risks relating both aquatic environment and human health. Therefore, the commercial and edible species have been widely investigated in order to check for those hazards (Begum et al., 2005).

Heavy metals released to aquatic environments result from both natural sources and anthropogenic activities. It was reported that fertilizers containing heavy metals cause pollution of the water body and fish, and that washing down the agricultural wastes by heavy rainfall increases heavy metal concentration of water. It was also found that some heavy metal concentrations such as copper and zinc increase in fish tissues in agricultural areas (Rashed, 2001; Dural et al., 2007). There are not heavily industrial activities in the western regions of Antalya province. But, because of its proper climate, it has been done dense agricultural activities in the region and has been used a lot of fertilizers and pesticides. This region is also an important touristic area. Due to heavy agricultural activities in the region, the use of chemical fertilizers and chemical pesticides may lead to water and fish contamination with heavy metals. Although some researchers have presented the elemental contents in fish from various regions of Mediterranean (Canli and Atli, 2003; Türkmen et al., 2008), there is not any study on heavy metal content of investigated fish from west Mediterranean shores of Antalya province. The investigated species (*Diplodus sargus*, *Siganus rivulatus*, *Lithognathus mormyrus*, *Liza aurata*, *Chelon labrasus*) are also very common in Mediterranean coastal regions of Turkey, and have commercial importance. For this reason, our objective from this study was to determine the some heavy metal bioaccumulation levels of edible portions (muscle and skin) and gills of the five fish species from the western Mediterranean shores of Antalya province (Turkey).

2. MATERIALS AND METHODS

2.1 Study Area and Fish Samples

The investigated fish species White seabream (Sparidae, *Diplodus sargus*), Marbled spinefoot (Siganidae, *Siganus rivulatus*), Striped seabream (Sparidae, *Lithognathus mormyrus*), Golden grey mullet (Mugilidae, *Liza aurata*), Thicklip grey mullet (Mugilidae, *Chelon labrasus*) were captured from the western Mediterranean shore of Antalya province (In Turkey). The fish were first wrapped into polyethylene plastic, put into an isolated container, and brought to the Biology Laboratory of Dumlupınar University. After biometric measurements, the fish were immediately frozen and stored at $-80^{o}C$ until dissection. The lengths and weights of the representative fish were presented in Table 1.

2.2 Sample Preparation

To minimize contamination, all the materials used in the experiments were previously washed in ultra pure water, and a stainless steel knife was used to cut the tissues. Before analysis the fish were thawed and approximate a 0.5 g sample was taken from each tissue (Muscle, gill and skin). Microwave method was applied for the digestion procedure of samples (Moeller et al., 2001). Three thawed 0.5 g homogenates from each tissue were placed in a Teflon digestion vessel with 7 ml of concentrated nitric acid (HNO_3) and 1 ml of Hydrogen peroxide (H_2O_2). The samples in the vessels were digested using an optimized microwave method. Then, they were let to cool at room temperature. The residues were dissolved and diluted to 50 ml. The chemicals used for the sample dissolution were of analytical grade. Ultra pure water was used throughout the study (Tuzen, 2003).

Table 1. Lengths (total and fork) and weights of investigated fishes (means ± SE)

Species	Total Length	Fork Length	Weight
Diplodus sargus	19.56±1.27	17.56±1.09	117.00±23.26
Siganus rivulatus	21.90±0.80	20.43±0.84	150.66±26.67
Lithognathus mormyrus	23.30±1.04	21.66±0.92	157.66±2.02
Liza aurata	18.80±0.03	17.43±0.08	56.16±2.45
Chelon labrasus	28.00±1.51	25.20±1.41	211.00±32.54

2.3 Chemical Analysis

The levels of elements (copper, zinc, manganese, iron and magnesium) were determined by a Perkin-Elmer DV 4300 inductively coupled plasma-optical emission spectrometry (ICP-OES). In the ICP–OES analysis, the following wavelength lines were used; copper 324.8 nm, zinc 213.9 nm, manganese 232 nm, iron 248.3 nm and magnesium 285.2 nm. The quality of the analytical process was also controlled by the analysis of NIST-CE278 certified standard reference materials of mussel tissue. The replicate analysis of these reference materials showed good accuracy.

2.4. Statistical Analyses

Each reported result was the mean value of three analyses. The results were offered as means ± SE. The statistical differences of the mean metal levels among tissues and species were analyzed using multiple comparison tests (SPSS package program). One-way ANOVA was utilized to compare the data by tissue. Results were considered significant at $p<0.05$.

3. RESULTS AND DISCUSSION

The accumulation levels of copper, zinc, manganese, iron and magnesium in muscle, skin and gills of investigated species were showed in figure 1, 2, 3, 4 and 5. The investigated fish species contained strikingly different metal concentrations in their tissues. As indicated in earlier studies, this may be related to the differences in ecological needs, swimming behaviors and the metabolic activities among different species (Canli and Atli, 2003).

In the present study, metal accumulation levels in tissues of same species were also significantly different ($p<0.05$). In general, the concentrations of all elements except copper were remarkably higher in the gills (Figure 1, 2, 3, 4, 5). In an experimental study, it was informed that the relative accumulation of copper in tissue of *Oreochromis niloticus* was of the order gills>liver>muscle (Birungi et al., 2007). It was reported that gills are the prime target for the toxic action of waterborne metals, and they accumulate heavy metal in higher concentrations because of its relatively higher potential for metal accumulation than muscle (Altindag and Yigit, 2005; Dural et al., 2007). It was also informed that these differences in metal concentrations of the tissues might be as a result of their capacity to induce metal-binding proteins such as metallothioneins (Usero et al., 2004; Urena et al., 2007). It was seen that our results supported those of the researchers. But copper accumulation levels in muscle and gills of the investigated species were contrary to the results reported by those researchers.

In this study, copper concentrations (*mg kg^{-1} wet weight*) in tissues of the species varied from 0.56 to 1.69 for muscle, from 0.55 to 1.03 for skin and from 0.75 to 1.49 for gill (Figure 1). There was no any difference in the concentration of copper between muscle and gills of the investigated species ($p>0.05$). It was informed that copper binding metallothioneins, which are known to be biomarker of metal exposures, in tissues cannot increase unless basal levels of copper in water are increased considerably (Atli and Canli, 2008). It was investigated the relationship between copper concentration and fish (*Poecilia reticulata*) size and indicated that body concentrations of copper were regulated and maintained at a certain concentration (Widianarko et al., 2000). It was also reported that the bioaccumulation of trace metals into aquatic organisms occurs when metal uptake rates exceed the depuration or detoxification rates (Baykan et al., 2007). Therefore, it may be said that copper is not biomagnified in fish tissues unless it's concentration in habitats increases to certain levels. On the other hand, it was informed that the maximum acceptable level of copper in edible portions of fish for human consumption was 20-30 mg kg^{-1} according to Turkish Standards, FAO and WHO (Table 2).

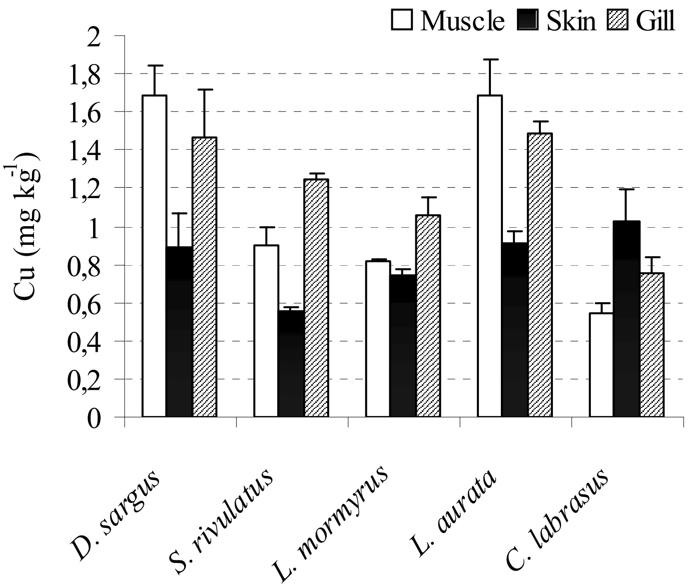

Figure 1. Copper levels (mg kg^{-1} wet weight) in muscle, skin and gills of the species

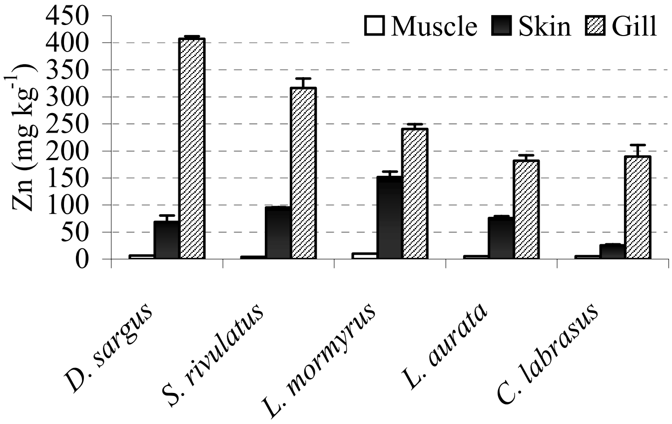

Figure 2. Zinc levels (mg kg^{-1} wet weight) in muscle, skin and gills of the species

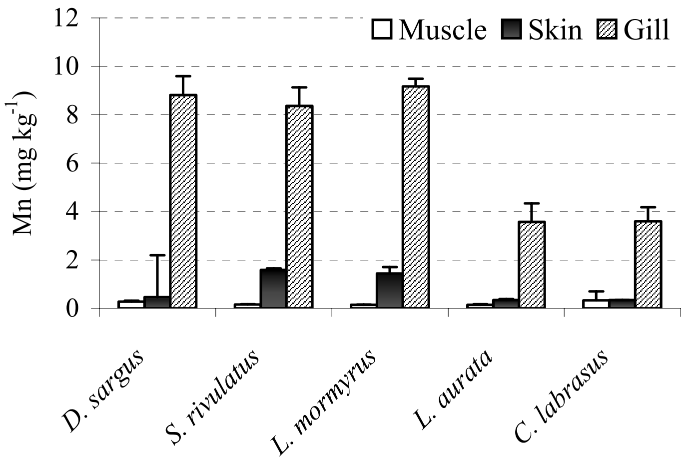

Figure 3. Manganese levels (mg kg^{-1} wet weight) in muscle, skin and gills of the species

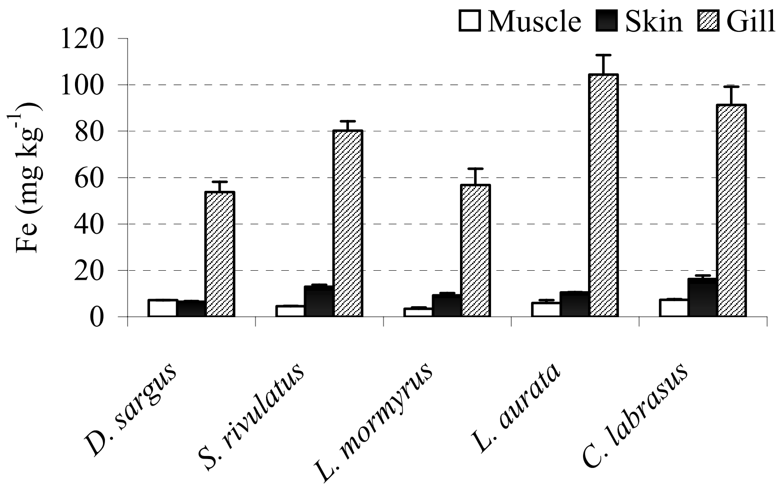

Figure 4. Iron levels (mg kg^{-1} wet weight) in muscle, skin and gills of the species

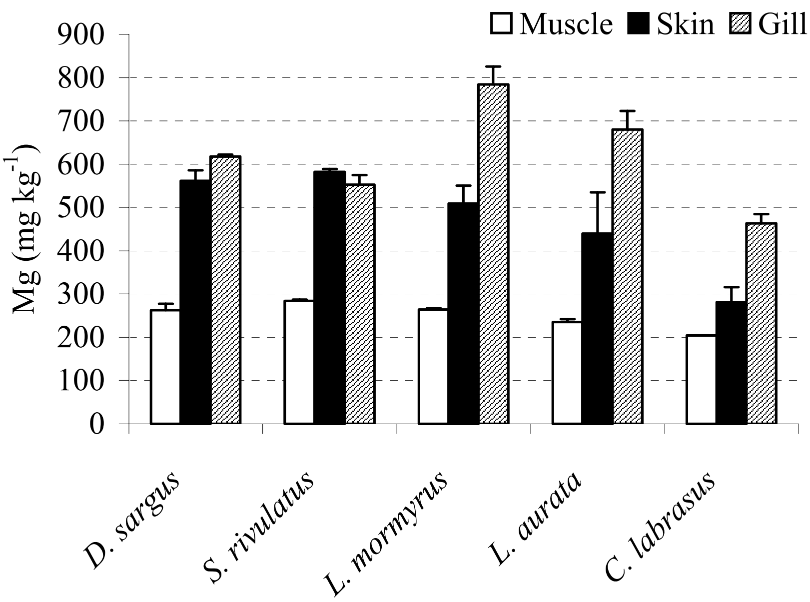

Figure 5. Magnesium levels (mg kg^{-1} wet weight) in muscle, skin and gills of the species

Table 2. Maximum permitted concentrations (mg kg^{-1} wet weight) of some heavy metals proposed by WHO, FAO and Turkish Legislation (Ikem and Egiebor, 2005; Demirak et al., 2006; Dural et al., 2007).

Metals	World Health Organization (WHO)	Food and Agriculture Organisation (FAO)	Turkish Legislation
Copper	30	30	20
Zinc	50	50	50
Cd	0.1	0.1	0.1
Pb	0.5	0.5	1

Copper concentrations in all tissues of the investigated species were also remarkable lower than that limit. For instance, copper concentration of the muscle of *Liza aurata*, which have the highest copper level among tissues of investigated species, was approximately 10 fold lower than that proposed acceptable maximum level by WHO, FAO and Turkish Standards (Table 2).

The differences in metal concentrations of the tissues were more obvious for zinc. Mean zinc concentrations (*mg* kg^{-1} *wet weight*) in tissues of the investigated species ranged from 4.15 to 9.97 for muscle, from 25.47 to 151.66 for skin, and from 182.30 to 407.09 for gills. In all species, zinc concentrations of muscle, skin and gills were significantly different from each others ($P<0.05$). Although the highest zinc concentrations were in the gills, the lowest zinc concentrations were in the muscle. Many researches indicated that muscle tissues are not an active site for metal accumulation, gills are also an active heavy metal accumulator organ (Fernandes et al., 2007; Yilmaz and Yilmaz, 2007). The main route of zinc entry into the fish was via water. When zinc in water rises to a level where the amount entering the organism through the gills exceeds the requirement for this metal, the surplus has to be excreted by liver (Birungi et al., 2007). On the other hand, a positive relationship was found between zinc levels in the gills and size of *Mugil cephalus* (Canli and Atli, 2003). It was reported that the highest bioaccumulation factor (BAFs) for zinc was in the gills of *Liza saliens*, and that zinc-gill accumulation in this species can be used as environmental indicators of metal stress (Fernandes et al., 2007). In this study, the findings concerning the zinc accumulation levels of gills conform to the findings of the previous researchers. Zinc concentrations in skin of the investigated species in this study are also between muscle and gills. Dermal route is also usually a minimal contributor of exposure, due to the often effective barrier provided by the external epithelium (Fernandes et al., 2007). Therefore, while the skin is directly contacting with water like gills of fish, it does not accumulate heavy metals as much as that of gills. Zinc is an essential element in human nutrition. Too little zinc can cause some health problems; however, too much zinc is harmful to human health (Duman et al., 2006). The maximum permissible level of zinc in fish proposed by WHO, FAO and Turkish Standards is 50 *mg* kg^{-1} *wet weight*. Zinc concentrations in muscle of all fish in this study were considerably lower than that limit. But zinc concentrations in skin of the studied fish except *Chelon labrasus* were higher than 50 *mg* kg^{-1} *wet weight*.

Manganese, iron and magnesium are important components of many metabolic processes and essential trace elements for humans (Birungi et al., 2007). The concentrations of manganese and iron in gills of all fish were remarkably higher than that of in muscle and skin ($P<0.05$). Mean Manganese concentration of edible portions (Muscle and skin) varied from 0.15 to 1.59 *mg* kg^{-1} *wet weight*. It was reported that the concentration of Manganese in muscle of fish was very low because Manganese does not accumulate in muscle of fish (Karadede et al., 2004). It was also reported that Iron deficiency is the most widespread nutritional disorder, and that fish has the potential to contribute to food-based strategies to reduce the risk of iron deficiency (Roos et al., 2007). The iron contents in muscle and skin of all species in this study were not significantly different from each other ($P>0.05$). Iron contents (*mg* kg^{-1} *wet weight*) in edible portions of the studied fish were ranged from 3.45 to 7.30 in muscle, and from 6.40 to 16.21 in skin. Magnesium contents of the species were found to be in the range of 204.33–283.73 *mg* kg^{-1} for muscle, 280.80-582.33 *mg* kg^{-1} for skin and 469.30-784.30 *mg* kg^{-1} for gills. There is no record on maximum permissible Manganese, iron and magnesium concentrations in fish tissues in Turkish standards.

4. CONCLUSION

Knowledge of heavy metal concentrations in fish is important both with respect to nature management and human health. In general, the investigated metal concentrations of the tissues in the present study were similar or lower than those of fish caught from many other aquatic habitats in Turkey (Karadede and Unlu, 2000; Topcuoglu et al., 2002; Yilmaz, 2003; Mendil and Uluozlu, 2007; Uluozlu et al., 2007; Türkmen et al., 2008). The mean concentrations of heavy metals analyzed in the muscle of all investigated fish were lower than the maximum permitted concentrations proposed by WHO, FAO and Turkish Legislation. But a potential danger may occur in the future depending on the dense agricultural and other anthropogenic activities in this region.

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