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ORIGINAL PAPERS

DISTRIBUTION OF HEAVY METALS IN THE MEAT, GILLS AND LIVER OF COMMON BREAM (*ABRAMIS BRAMA* L.) CAUGHT FROM ŻNIŃSKIE DUŻE LAKE (POLAND)

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Abstract

The aim of this work was to compare the concentrations of Pb, Cd and Co in the meat, gills and liver of common bream (*Abramis brama* L.) caught from Żnińskie Duże Lake. Analyses were carried out on 20 individual fish caught in autumn, in the natural environment. Measurements of the mass of the fish body (BW) and body length (Lc) were taken on the each specimen. Muscle samples were taken from the large side muscle of the fish body above the lateral line. Pb, Cd and Co concentrations were determined on an atomic absorption spectrometer Solaar 939 QZ. Significance of differences in the average content of metals was calculated by one-way analysis of variance (ANOVA). Analyses of the variance indicated that the mean value of Pb was highest in gills (0.880 mg kg^{-1} dry weight) and lowest in the liver (0.068 mg kg^{-1} dry weight). The same results were observed for Co, and those values were 2.391 and 0.357 mg kg^{-1} dry weight, respectively. The highest mean concentration of Cd was determined in the liver (0.143 mg kg^{-1} dry weight). There were statistically significant differences in the content of all analyzed metals between the muscle, gills and liver (at $p \leq 0.05$). Analyses indicated that the mean concentrations of Pb, Cd and Co were detected in the following order: Co > Pb > Cd (in the meat and gills) and Co > Cd > Pb (in the liver). Co and Pb accumulated in the large amounts in the gills (gills > meat > liver). Cd concentrations detected decreased in the following order: liver > meat > gills. Results of the principal component analysis (PCA) substantiated very good separation of the three analyzed tissues in respect of their metal content, which confirms the results obtained from the analysis of variance.

Keywords: fish, tissues, lead, cadmium and cobalt.

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INTRODUCTION

Fish meat is a good source of protein, low saturated fat and omega fatty acids, which is why it should be an important part of human diet, as it can reduce the risk of coronary diseases, hypertension and cancer (Rahman et al. 2012). The American Heart Association suggest eating fish at least twice a week in order to reach the daily intake of omega 3 fatty acids recommended for healthy adults (Al-Busaidi et al. 2011).

Fish may accumulate toxic trace metals in their tissues through direct absorption or via the food chain and pass them to human organism by consumption. Accumulation of metals in fish depends on concentrations of the metals in water and food, on physiochemical factors and the duration of exposure. Heavy metal pollution in fish has become an important worldwide concern due to the health risk associated with fish consumption (Al-Busaidi et al. 2011). Analysis of predatory and benthic foraging fish are very important, because these fish species may accumulate large amounts of heavy metals.

The distribution of metals varies between fish species, depending on age, season of the catch, development status and other physiological factors (Dural et al. 2006, Mendil et al. 2010, Fallah et al. 2011). It is very difficult to compare metal concentrations even between the same tissues of two different species, because of different feeding habits and growing rates (Yilmaz et al. 2010). As many research reports indicate, different fish species from the same reservoir may contain different metal levels in their tissue (Uysal et al. 2008). It has also been experimentally proven that heavy metal accumulation in fish tissue or organs depends on their physiological role. In fish, metals accumulate mainly in the kidneys, liver and intestinal epithelium. Fish muscles contain low levels of metals, but are often examined for metal content because of their use for human consumption (Pourang 1995, Roméo et al. 1999, Dural et al. 2006, Uysal et al. 2008, Lidwin-Kaźmierkiewicz 2009, Fallah et al. 2011). The gills and liver are target organs for assessing metal accumulation. The liver has a tendency to accumulate metals to a higher degree than muscle tissue.

The aim of this work was to compare concentrations of Pb, Cd and Co in the meat, gills and liver of deep-water fish such as common bream (*Abramis brama* L.) caught from Żnińskie Duże Lake.

MATERIAL AND METHODS

Żnińskie Duże Lake is one of the largest lakes in the Kuyavia-Pomerania Province, in the district of Żnin, within the Gniezno Lake District (Figure 1).

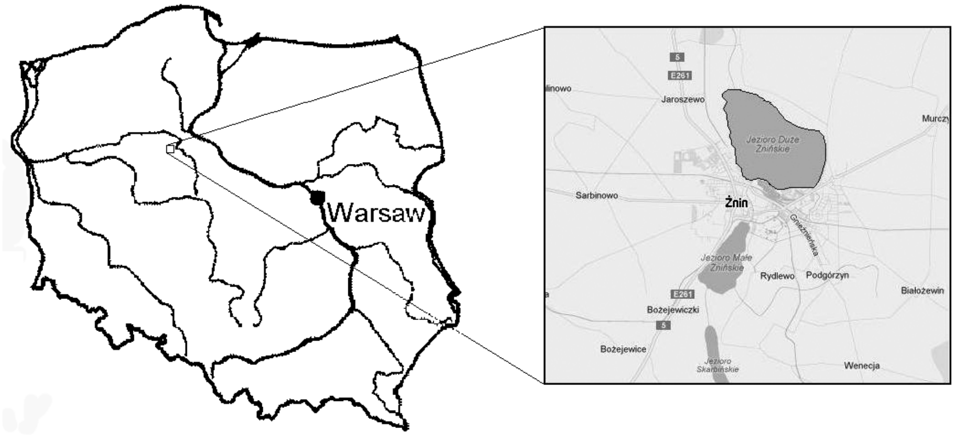

Fig. 1. The location of Żnińskie Duże Lake in Poland

The surface area of lake is 431.6 ha. The average depth of the lake is 6.8 m and the maximum depth is 11.1 m. The lake was a sink for social, domestic and industrial discharges for many years. Żnińskie Duże Lake is ideally suited for windsurfing because of the lack of forests along the shoreline and therefore a windsurfing regattas are organized every year.

The study involved freshwater fish caught in autumn. The experimental fish were obtained in natural conditions from Żnińskie Duże Lake. Analyses were carried out on 20 individuals of 4-year-old common bream (*Abramis brama* L.) females. Measurements of the mass of the fish body – BW (± 0.01 g) and body length – Lc (± 0.1 cm) were taken on each individuals. The individuals selected for analyses were similar in biometric measurements. The body weight ranged from 200.90 to 265.50 g (the mean value 235.60 g) and the body length was from 22.0 to 23.5 cm (the mean value 22.5 cm). Meat, gills and liver were sampled for analyses. The meat samples were taken from the large side muscle of fish body above the lateral line. Due to relativeky small liver samples, material from individuals of a similar body length was combined (about 2-3 pieces).

The samples of fish meat were immediately frozen and kept in a deep freezer before analyzing. All frozen samples were freeze dried in a Finn-Aqua Lyovac GT2 freeze drier (parameters: temp. -40°C, pressure 6 10^{-2} mbar, duration at least 48 h).

The freeze dried samples were mineralized in a microwave mineralizator Ethos Plus, Milestone. For mineralization, 0.1g of the tissue was weighted and then HNO_3 and H_2O_2 were added in the ratio 4:1. During the first 10 minutes, the temperature was increased to 190°C. During the next 7 minutes, the temperature was maintained at 190 ± 5°C. The mineralized samples were transferred quantitatively to 50 ml measuring flasks.

Pb, Cd and Co concentrations were determined on an atomic absorption spectrometer Solaar 939 QZ, ATI Unicam. Analyses were carried out according to PN-EN 14084 – 2004. Tissue concentrations of minerals were reported as mg kg^{-1} dry weight (mg kg^{-1} dw).

The analytical accuracy was controlled by adding standard solutions (Recipe Chemicals + Instrument GmbH, Germany). The results showed that the recovery percentage was in the range from 92 to 105% and this values were included in the final results. All analytical samples and blanks were prepared in triplicate.

Data analyses were performed by using Statistica 8.0 software (StatSoft, USA). Significance of differences in the average content of metals in the meat, gills and liver was calculated by one-way analysis of variance (ANOVA). The Tukey's test was applied, and statistically significant differences were evaluated as significant at $p \leq 0.05$. The normality of the data was tested using the Shapiro-Wilk's test and the homogeneity of variance was verified by means of the Levene's test. The classification and discrimination of different tissues were achieved by principal component analysis (PCA).

RESULTS AND DISCUSSION

As numerous studies confirm, environmental conditions such as the season of the catch of fish, temperature and the chemical composition of water (salinity, pH, hardness and temperature) affect the physiological status of fish and the ability to accumulate metals in their organs (Canli, Atli 2003, Dural et al. 2006, Pourang 1995). Ecological needs, sex, size and seasonal changes were also found to influence metal accumulation in fish tissue (Farkas et al. 2003, Łuczyńska, Brucka-Jastrzębska 2005, Yilmaz et al. 2010). Metal tissue concentrations are influenced by environmental contamination, too. Fish collected from more contaminated or cooler lakes had lower indicators of physical condition than individuals from cleaner water bodies (Eastwood, Couture 2002). Farkas et al. (2003) observed significant and positive correlations between the level of heavy metals accumulated in the organs of fish and the pollutant load of the water. Fish meat, gill and liver tissues were chosen for analyses. The gills and liver are target organs for assessing metal accumulation. The content of metals in gills reflects the concentrations of metals in waters, where the fish species lives. Whereas the concentrations metals in liver represent storage of metals (Roméo et al. 1999).

Lead is a toxic element, which does not play any metabolic role. It is a non-essential element, which may cause neurotoxicity, nephrotoxicity and many others adverse health effects (Rahman et al. 2012). An important biochemical effect of Pb on the hematopoietic human system is its inhibition of the biosynthesis of heme. Pb binds strongly to the sulfhydryl functional group in δ-aminolevulinic acid dehydrase (ALAD) which is a key enzyme in the formation of heme. Inhibition of ALAD occurs at blood Pb concentration of 10-20 μg 100 cm^{-3} (AL-BUSAIDI et al. 2011) or 20-40 μg 100 cm^{-3} (SKOCZYŃSKA 2008). Pb can affect humans when ingested or inhaled in high doses. In fish, it can cause deficits or decreases in survival, growth rates, development and metabolism (YILMAZ et al. 2010). The maximum permitted lead level is from 0.3 mg kg^{-1} wet weight – ww (*Commission* EU... 2015) to 2.0 mg kg^{-1} ww (WHO 1996). The analyses of variance indicated that the mean value of Pb was the highest in the gills (0.897 mg kg^{-1} dw) and the lowest in the liver (0.067 mg kg^{-1} dw) – Table 1.

Table 1

Pb, Cd and Co content in the meat, gills and liver of common bream (*Abramis brama* L.) caught form the Żnińskie Duże Lake

Organ/tissue	*n*	Metal content (mg kg^{-1} dw) (mean ± SD)		
		Pb	Cd	Co
Meat	20	0.300 ± 0.065[a]	0.060 ± 0.024[a]	0.764 ± 0.076[a]
Gills	20	0.880 ± 0.048[b]	0.028 ± 0.009[b]	2.507 ± 0.271[b]
Liver	8	0.068 ± 0.019[c]	0.143 ± 0.026[c]	0.357 ± 0.029[c]

Mean values designated various letters in the same column are statistically significantly different ($p < 0.05$, the Tukey's test)

Statistically significant differences were observed in the content of all analyzed metals between the muscles, gills and liver (at $p \leq 0.05$). Similar results were observed by ERDOĞRUL, ERBILIR (2007). They detected that the mean content of Pb in the meat and gills ranged from 0.06 to 0.14 (mg kg^{-1} ww), and from 0.06 to 0.50 (mg kg^{-1} ww), respectively. The mean concentration of Pb in the meat of common bream (*Abramis brama* L.) caught from lakes in the Olsztyn Lake District ranged from 0.027 to 0.182 mg kg^{-1} ww (ŁUCZYŃSKA, BRUCKA-JASTRZĘBSKA 2005). Those values were very similar to the ones in this paper. Concentrations of Pb in the meat of common bream collected from the two basin of Lake Balaton (the western and eastern one) were 1.53 and 0.85 mg kg^{-1} dw, respectively in October and 0.49 and 0.43 mg kg^{-1} in May (FARKAS et al. 2002). The average Pb concentration in the meat of common bleak (*Alburnus alburnus*) and common bream collected from the Šalek lakes (Slovenia) ranged from 0.01 to 0.06 mg kg^{-1} and from 0.01 to 0.10 mg kg^{-1} ww, respectively (PETKOVŠEK et al. 2012). As this study showed, the Pb concentration was similar to those of unpolluted areas. The Pb content in the fish meat analyzed by LIDWIN-KAŹMIERKIEWICZ et al. (2009) ranged from 0.01 to 0.02 mg kg^{-1}. These values were low-similar or lower than those reported for fish by other authors. In the meat of perch (*Perca fluviatilis* L.) from the Pomeranian Bay and Szczecin Lagoon, Pb concentrations ranged within 0.007-0.033 mg kg^{-1} ww and 0.013 mg kg^{-1} ww, respectively (SZEFER et al. 2003).

It has been demonstrated that fish meat is not a physiologically specific site for Pb concentration (AL-YOUSUF et al. 2000). Pb accumulates in the largest amounts in bones. As PERKOWSKA, PROTASOWICKI (2000) showed, Pb accumulated in different ratio in the liver and kidneys. In all the analyzed fish species, the content of this metal was higher in the gills than in the meat. This proves that the respiratory system is the main route of Pb acquisition by fish. YILMAZ et al. (2010) reported that the highest Pb concentration was determined in the liver of *Solea lascaris* (2.98 µg g^{-1} ww) and the lowest Pb concentration was always found in the meat of fish (0.14-0.39 µg g^{-1} ww). The skin accumulated more Pb than the meat. The reason for high metal concentrations in the skin could be the metal complexing with the mucus, which is impossible to remove completely from the tissue before analysis. JARIĆ et al. (2011) observed that heavy metal accumulation in the muscles, gills, liver and intestines differed statistically significantly in the case of all the assessed metals. The highest concentrations of most of the analyzed metals and trace elements was recorded in the liver, while the lowest ones occurred in the meat. Analyses carried out by PETKOVŠEK et al. (2012) indicated that the accumulation of Pb was more pronounced in the gills than in the meat or liver. The authors suggested that an elevated content in the gills reflected the high content of this element in plankton and was correlated with feeding habits. The results showed that Pb levels in the meat of common bream were lower than the maximum allowable concentrations.

Cadmium is a serious contaminant, a very toxic element and potentially more lethal than any other metal (RAHMAN et al. 2012). The maximum admissible value for fish is 0.05 mg kg^{-1} ww (YILMAZ et al. 2010). According to the European Commission, the permissible limit for Cd in fish for human consumption is 0.05-0.1 µg g^{-1} (AL-BUSAIDI et al. 2011, *Commission* EEC... 2001, *Commission* EU 2014). Cd is a toxic element, does not play any metabolic function and can be harmful for humans, even at a low concentration, when ingested over a long period of time. Cd is an element capable of producing chronic toxicity even it is present at a concentration of about 1 mg kg^{-1} (RAHMAN et al. 2012). Analyses of variance indicated that the mean value of Cd was the highest in the liver (0.143 mg kg^{-1} dw) and the lowest in the gills (0.028 mg kg^{-1} dw) – Table 1. BRUCKA-JASTRZĘBSKA, PROTASOWICKI (2006) carried out analyses on levels of selected metals in tissues and organs of 5-month-old carp (*Cyprinus carpio* L.). They determined that the mean content of Cd in the meat, gills and liver was 0.004, 0.039 and 0.053 µg g^{-1} ww, respectively. The Cd concentration in fish samples from the Bangshi River ranged from 0.09 to 0.87 mg kg^{-1} and 0.16 to 0.68 mg kg^{-1} in pre-monsoon and post-monsoon season, respectively (RAHMAN et al. 2012). The Cd concentration in the muscle, gills and liver of bream collected from the Western Basin of Lake Balaton was 0.94, 2.25 and 1.94 (mg kg^{-1} dw) in October, and 0.56, 1.37 and 1.51 (mg kg^{-1} dw) in May. In the organs collected from the Eastern Basin of Lake Balaton, those values were 0.40, 0.84 and 1.64 (mg kg^{-1} dw) in October and 0.62, 1.12 and 1.79 (mg kg^{-1} dw) in May (FARKAS et al. 2002). JARIĆ et al.

(2011) determined that Cd reached the highest concentration in the liver (2.826 μg g^{-1} dw) and the lowest in the meat (0.085 μg g^{-1} dw). The liver is the target organ, which is involved in detoxification and accumulation. The same results were observed by Canli, Atli (2003). Al-Yousuf et al. (2000) determined that the content of Cd in the liver was higher than in the skin and the meat. Yilmaz et al. (2010) observed that Cd levels in the skin, which might also be the route of uptake in all the samples, were higher than those in muscles. A higher Cd concentration was found in the liver of red gurnard (*Trigla cuculus*), while the lowest Cd levels were always found in the muscle tissue of the analyzed fish. As demonstrated analytically, the level of Cd in the meat of common bream was less than tha maximum allowable concentrations, but it seems advisable to repeat analyses on the differences in accumulation of this metal by the fish meat and gills.

Cobalt is an essential nutrient for humans, as it is an integral part of vitamin B_{12}. Cobalt deficiency contributes to anoxia and myocardial muscle injuries. This metal is necessary for a proper function of the thyroid (Yilmaz et al. 2010). The daily recommended range of cobalt in the human diet is 40-50 μg day^{-1} (Kabata-Pendias, Kabata 1999, Heghedűş-Mîndru et al. 2014). Analyses of variance indicated that the mean value of Co was the highest in the gills (2.507 mg kg^{-1} dw) and the lowest in the liver (0.368 mg kg^{-1} dw) – Table 1. Similar relationships (values calculated as wet weight) were observed by Erdoğrul, Erbilir (2007). They detected that the mean content of Co in the meat and gills ranged from 0.005 to 0.01 (mg kg^{-1} ww), and from 0.013 to 0.05 (mg kg^{-1} ww), respectively. Analyses carried out by Yilmaz et al. (2010) demonstrated that the mean content of Co in the meat, skin and liver ranged from 0.06 to 0.39, from 0.32 to 0.47 and from 0.50 to 0.75 μg g^{-1} ww, respectively. Co concentrations in our study were in agreement with data in the literature.

The PCA analysis indicated that PC1 and PC2 scores justified very good separation of the three investigated tissues. The results of these analyses are plotted in Figure 2, where three different clusters for the gills, liver and meat can be seen. Some earlier research concerning fatty acids profile, which was conducted by Kupcewicz et al. (2011) according to the chemometric method, confirmed good separation of three fish species and two geographical areas investigated.

CONCLUSIONS

1. Analyses showed that the mean concentrations of Pb, Cd and Co were detected in the following order: Co > Pb > Cd (in the meat and gills) and Co > Cd > Pb (in the liver). The content of Pb, Cd and Co in the meat of analyzed fish was lower than the maximum levels set by law.

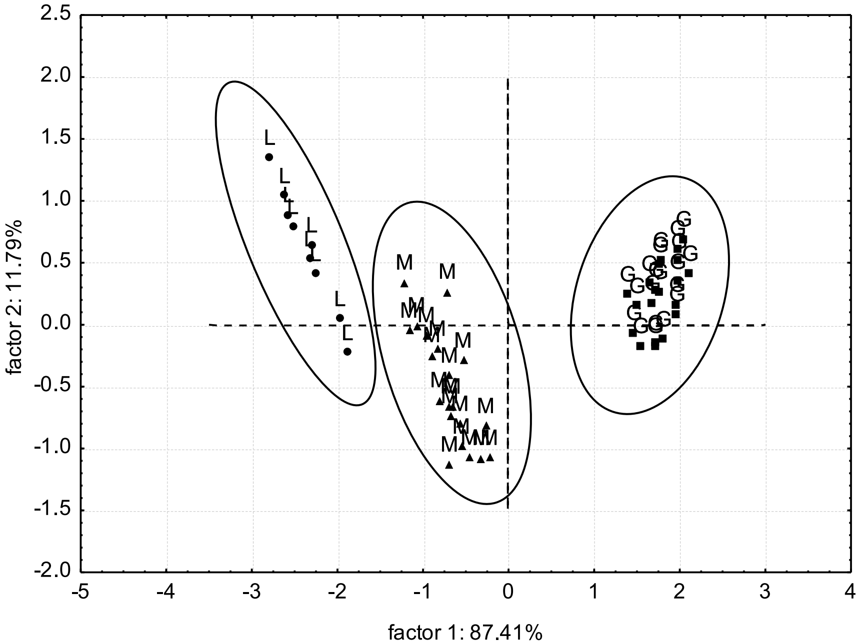

Fig. 2. Score plot for PC1 and PC2 for three tissues: G – gills, L – liver, M – meat

2. Co and Pb accumulated in large amounts in the gills (gills > meat > liver). Cd concentrations decreased in the following order: liver > meat > gills.

3. Large variations were observed in the content of the analyzed elements, depending on the type of tissue. The PCA revealed very good separation of the three investigated tissues owing to statistically significant differences in the metal accumulation in the different organs.

4. Although the meat of analyzed fish is healthy and safe for human consumption, further monitoring should be conducted. Fish tissues are good indicators of the presence of metals in the environment. The scarcity of literature data about the content of heavy metals in the waters of Żnińskie Duże Lake substantiates further expansion and continuation of the research of this lake, with special attention paid to tendencies in metal accumulation in different fish tissues.

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