Animal welfare, etológia és tartástechnológia

Animal welfare, ethology and housing systems

Volume 9 Issue 3

Különszám/Special Issue

Gödöllő

2013

LEAD AND COPPER CONTENT OF THE ROACH, WHITE BREAM AND PERCH ORGANS

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The aim of this study was to describe the place where elements like lead and copper has the highest level. Only females of three species of fish from Nida River were taken. After collected organs – (bones, gonads and heart muscle) elements by atomic absorption spectrophotometry (AAS) was determined in it. The highest level of lead and copper were detected in heart muscle of roach. But in case of the lowest level of lead and copper were detected in white bream gonads.

Keywords: white bream, roach, perch, AAS, lead, copper, fish organs

Introduction

These elements can get to an organism with food, by taking air or by a direct contact with skin. They have mainly an influence on the functionality of the organism they get to. The most harmful metals for living organisms and occurring in environment we are toxic elements such as cadmium, mercury, lead, or caesium. However, biogenic elements which are essential for the functionality of the organism such as zinc or copper, can become as toxic as xenobiotic elements if they are absorbed in too big amount.

Water organisms such as fish are particularly sensitive to water pollution. Since years scientists throughout the world have been doing some researches into the water pollution by different toxins and by some elements, too.

Copper is an essential element to live and it is presented in all organism tissues. . Copper is essential to the process of hemoglobin synthesis, as a coenzyme it falls in the composition of certain enzymes involved in redox processes. It is a component, inter alia, of ceruloplasmin (EC 1.16.3.1) – plasma protein which regulates metabolism and transport of iron, cytochrome c oxidase (EC 1.9.3.1) taking part in transport of electrons, tyrosinase (EC 1.14.18.1) involved in the process of melanin production, lysine oxidase (EC 1.4.3.13) which takes part, inter alia, in collagen fibers synthesis, dopamine beta – monooxygenase (EC 1.14.17.1) which takes part in the synthesis of peptide hormones, superoxide dismutase (EC 1.11.1.6) which takes part in the protection of aerobic organisms against cytotoxic activity of superoxide, and peptidyl α-amidating monooxygenase (PAM) (EC 1.14.17.3) taking part in the catalysis of incorporating an oxygen atom to the hydroxyl substrate (Mercer, 2001).

It is considered that the tissues of certain animals have a particular tendency to collect copper. The highest level of copper was found in invertebrates - in the blood of annelids, crustaceans and molluscs, where it is a component of hemocyanins (Kabata – Pendias, Pendias, 1999). In vertebrate animals, high copper content was found, inter alia, in the liver of birds (Eun – Young, et al., 1996), human milk (Dorea, 2000), muscles of dolphin (Wood, Van Vleet, 1996), kidneys of rat (Cui, Okayasu, 2008) and gills of fish (Romeo, et al., 1999).

However, lead is an element which gets to living organisms similarly as another xenobiotic element – cadmium by the digestive and respiratory track. Independently of the way of absorption lead gets to peripheral blood, where it is tied by erythrocyte membranes and it is transported together with

blood to different organs and tissues (Hoffman et al., 1985). The highest lead concentration in animals body occurs mainly in the liver, kidneys, brain and bones. Other organs like muscles, gonads, feathers or adipose tissue can also absorb easily lead, what was shown for example of small Australian birds – Zebra Finches by Dauwe et al. (2002). High concentration of this element was found also in human milk teeth, where the highest lead content was in the incisors (Karahalil, et al., 2007).

This metal has a strong influence on activity of many enzymes as acetylcholinesterase (EC 3.1.1.7) (Reddy, et al., 2007), which takes part in nervous impulses transmitting or glutamine synthetase (EC 1.4.1.14), which takes part in transforming glutamate into glutamine (Sierra, Tiffany-Castiglioni, 1991). It also impacts on blood and haematological system harmfully, it hinders the process of hemoglobin synthesis and it shortens the life of erythrocytes (Orłowski 2008). Lead is a strong cytoplasmic poison, it combines with groups of sulfhydryl enzymes and cellular proteins inactivating them. It disorders redox processes, nucleic acids synthesis (RNA and DNA), transformation of high-energy compounds as well as metabolism of some elements (Czerwionka-Szaflarska, Nowak, 1996).

In human's point of view it is important how undesirable substances get to his body. The content of elements in various products eating by man was studied by many scientists throughout the world. The content of such metals for example as Pb, Ni, Cd or Cu was found among others things in honey and bees products (Formicki et.al. 2013), in milk (Abdou, K.A. and Korashy, E. 2001), or in cheese (Ekbal M.A. Ibrahim, 2004), as well as in fish muscles (Yilmaz, et al., 2007, Zyśk, 2013). Fish are an integral component of human's diet so it is important to get to know the content of these elements which have important roles in life in these animal's tissues. All the elements that got to the body wander through it with blood and get beside dorsal muscles also to the myocardium, gonads or bones. Three species of fish: roach (*Rutilis rutilus* L.), white bream (*Blicca bjoerkna* L.) and perch (*Perca fluviatilis* L.) were chosen to the study because they represent various ecological types and they are often caught by anglers in many countries. Roach was chosen as a pelagic species living in the water depths and it eats mainly plant food. White bream is a species living at the bottom of water body and it eats both plant food and animal food, whereas perch is a predatory species penetrating different parts of water.

Materials and methods

The study involved 10 females of three species of white bream (*Blicca bjoerkna*), roach (*Rutilus rutilus*) and perch (*Perca fluviatilis*). The fish were caught in Nida River in the south of Poland. From all caught alive fish were dissected samples (1g) of fresh weight. The samples were dissected from particular organs such as heart muscle (all), fragment of gonads, and bones (ribs). Taken organs were dried at 105° C in order to obtain dry mass of organs. Next the material was mineralized in concentrated nitric acid (HNO3) at the temperature of 90° C until complete dissolution of tissues using VELP Scientifica DK 20 mineralizator. Next the samples were thinned with spectrally pure water to cubic capacity of 10 ml. The samples obtained this way were analysed for the content of lead and copper, which were assayed with atomic absorption spectrophotometry (AAS) in a Cole – Palmer, BUCK 200A apparatus. The results were subjected to statistical analysis using the STATISTICA software involving ANOVA tests and post hoc Tuckey analysis.

Results distribution was checked by Shapiro – Wilk's test. Homogeneity of variance was checked by Levene's test. Statistical significance was defined at $P < 0,05$.

The content of studied elements in water and bottom sediment from Nida River was compared on the basis of the information received from Voivodship Inspectorate for Environmental Protection in WIOŚ Kielce.

Tab.1. Copper and lead content in water and bottom sediment of Nida River according to WIOŚ Kielce

Water	**Cu**	0,0013 mg Cu•L^{-1}
Bottom sediment		4 mg•kg^{-1}
Water	**Pb**	0,0015 mg Pb•L^{-1}
Bottom sediment		8 mg•kg^{-1}

Results

The average copper content in gonads of roach coming from Nida River was 18.645±14.244 µg•g^{-1} s.m but in gonads of white bream was the lowest and it was 7.700±3.372 µg•g^{-1} s.m. The highest copper content in gonads of the examined fish species was in perches and it was 24.899±8.395 µg•g-1 s.m.

There was statistical significance in copper content in gonads between white bream and roach (p=0.001).

The average lead content in gonads of roach was 29.300±36.830 µg•g^{-1} s.m. and was the highest among examined fish species. The average lead concentration in gonads of white bream was 4.043±4.340 µg•g^{-1} s.m and it was the lowest among examined fish species. However, the average lead content in gonads of perches was 16.980±3.931 µg•g^{-1} s.m.

There was statistical significance in lead content in gonads between white bream and roach (p=0.001).

Tab.1 The average lead and copper content in gonads of white breams, perches and roaches expressed in *µg•g^{-1} s.m.±* standard deviation (SD)

	River	**Roach**	**White bream**	**Perch**
Copper	Nida	18.645 ±14.244[a]	7.700 ± 3.372[a]	24.899 ± 8.395
Lead	Nida	29.300 ±36.830[a]	4.043 ± 4.340[a]	16.980 ± 3.931

a- statistical significance ,p < 0.05, between individual fish species

The average copper content in the heart muscle of roach was the highest - 105.423±115.751 µg•g^{-1} s.m, whereas white breams accumulated in their heart only 34.101±24.572 µg•g^{-1} s.m., what gave the lowest average concentration of this element among examined fish species. The concentration of copper in the myocardium of perches was 104.915±9.765 µg•g^{-1} s.m.

There was statistical significance between the roach and white bream (p=0.018).

The average lead content in the heart muscle of roach was 105.484±80.284 μg•g^{-1} s.m. and similarly like in case of copper was the highest in this species among examined fishes. However, the average concentration of lead in the myocardium of white bream was 58.281±108.769 μg•g^{-1} s.m., and in the myocardium of perch was 41.191±30.502 μg•g^{-1} s.m.

There was not statistical significance in lead content in the heart muscle between species (p=0.807).

Tab.2 The average lead and copper content in the heart muscle of white breams, perches and roaches expressed in *μg•g^{-1} s.m.±* standard deviation (SD)

	River	**Roach**	**White bream**	**Perch**
Copper	Nida	105.432±115.751[a]	34.101±24.572[a]	104.915±9.765
Lead	Nida	105.484±80.284	58.281±108.769	41.191±30.502

[a]- statistical significance p < 0.05, between individual fish species

The average copper content in bones of roach was 23.448±19.633 μg•g^{-1} s.m., in white bream only 9.547±6.754 μg•g^{-1} s.m., while the highest average copper content was in bones of perch - 45.287±25.117 μg•g^{-1} s.m.

There was statistical significance in copper content in bones between white bream and roach (p=0.020), and between white bream and perch (p=0.026).

The highest average lead content was found in bones of roach - 70.976±40.992 μg•g^{-1} s.m., the lowest lead concentration was found in bones of white bream - 36.544±10.746 μg•g^{-1} s.m., and the average lead content was in bones of perches - 59.816 ±10.646 μg•g^{-1} s.m.

There was statistical significance in lead content in bones between white bream and roach coming from the Nida (p=0.001).

Tab.3 The average lead and copper content in bones of white breams, perches and roaches expressed in *μg•g^{-1} s.m.±* standard deviation (SD)

	River	**Roach**	**White bream**	**Perch**
Copper	Nida	23.448±19.633[a]	9.547±6.754[a,b]	45.287±25.117[b]
Lead	Nida	70.976±40.992	36.544±10.746	59.816±10.646

[a] - statistical significance, p < 0.05, between individual fish species

[b] - statistical significance, p < 0.05, between individual fish species

Conclusion

Fish are a very specific group of vertebrate animals because they spend all their life in water. They are continually under the influence of various physical and chemical factors which occurs in water. Fish food are different water organisms, plants and animals which are under the influence of the same factors. Metals absorption to the body of fish takes place in two ways, the first one is to absorb them directly from the environment and the second to take them together with food (Sapota, 2008).

Taking into account the concentration of both studied elements in water and the bottom of water body of Nida River it was possible to expect the presence of these metals in examined fish tissues as well.

It was found that organs of all examined fish species – white bream, perch and roach contain as well copper as lead. However, the concentration of these elements in organs of particular fish species was different. Analyzing the results it is important to note high standard errors that were caused by a large result of single scattering group. This can prove about a big interspecific variation in the way of food intake or about presence of single individuals coming from Nida River.

The highest copper level among studied organs was found in the heart muscle of all examined fish species. The concentration of this element in the heart of roaches and perches was definitely higher than in the heart of white breams. Clearly higher copper content was also found in the bones of perches. Similar results of high copper accumulation in the bones were obtained by Moissenko i Kurdyavtseva (2001), studying organs of lavaret (Coregonus lavaretus) and brown trout (*Salmo trutta morpha fario*). High level of this metal in the heart muscle may be associated with high metabolic activity of this organ.

The lowest copper content was in gonads, wherein the lowest its level was found in white bream. However, copper concentration in animals gonads can have seasonal changes which depend on the biology of the species. This fact was found in common frog (*Rana temporaria*), between the period of winter torpor and the mating season of this species - that is the time spent in the water (Stawarz, 1998). According to the author copper content in ovaries of frogs rises in the mating season. Examined fishes were caught in the spring period where it is spawning season of these species. It can be inferred that copper accumulation in gonads of studied fishes was also higher in this period. Apart from that a big amount of substances as well as examined elements is washed out from fields and meadows directly to rivers where fish live during the spring thaw.

The highest lead content was in the heart muscle and bone tissue in all examined fish species. Average the highest lead level was in the heart of roach, whereas the lowest its concentration was in gonads of white bream. At the same time gonads were the place where the lowest was lead accumulation in all studied fish species.

The lead content in the bone tissue of fish was high. This can be attributed to the fact that this metal is continuously installing in the bone structure and its accumulation occurs throughout the life of the organism (Orłowski, 2008). High lead concentration in bones of 6 fish species from the Mediterranean was found by Khalifa et al. (2010), studying some elements content In their organs. Study of lead content, inter alia, in bones of amphibians was also conducted by Stawarz (1998). He found that lead accumulation in bones of common frog (*Rana temporaria*) was very high during the mating season and the period of winter torpor, that is the time spent in the water.

Lead is a metal that has a harmful influence on gonads, it impairs, inter alia, the secretion of testosterone in testicles, thus it causes disorders of spermatogenesis and steroidogenesis (Martynowicz, et al., 2005).

No great concentration of this metal in gonads of examined fish can provide about the processes occurring in the body, which protect gonads against the absorption of harmful substances such as lead to them. Kosior‘s et al. (2002) research confirms the low lead content in gonads of female fish living in the Southern Baltic Sea Lagoons.

The heart muscle of all examined by me fish species contained lead in a very large amount. Most likely it appears that the big amounts of lead in the heart muscle are caused by high metabolic activity of this organ. Lead is transported through the animals’ body with blood (Hoffman et al., 1985). The whole blood contained in the body together with all compounds contained therein,

passes through the heart, and some of these compounds like, for instance lead can be accumulated in cardiac tissue.

The conducted research also demonstrated that the species the least susceptible to accumulation of heavy metals in its organs is white bream, although, it takes the most amount of food from the bottom of water body. Perch and roach contained much more examined elements in their organs than white bream. Such high contents of examined elements that were marked in organs of roach can be explained by UV radiation, which penetrates easier in the high part of water, so in the place of roach living in the reservoirs. Formicki et al. (2006) wrote about such cases studying amphibian larvae. It is possible that physical and chemical factors or species-individual properties have also an influence on it but they were not taken into account. However, high concentration of examined chemical elements in perches can be ca mainly used by predation of this species.

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