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STUDY OF THE BIOACCUMULATION OF HEAVY METALS IN ASTACUS LEPTODACTYLUS IN SOME AREAS OF THE KARDZHALI DAM

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ABSTRACT

It is examined the contents of lead, cadmium and nickel in the water of Kardzhali dam, as well as the bioaccumulation of the heavy metals in two organs – liver and muscles of *Astacus leptodactylus*. In tissue samples is reported increasing of cadmium in samples of liver. The bioaccumulation coefficient was calculated on the basis of the average content of lead, cadmium and nickel in the organs of *Astacus leptodactylus*. We recorded that the studied crayfish are macroconcentrators for cadmium.

The analyzes have shown that lake crayfish to be defined as a biomarker in toxicity testing in contaminated waters.

Key words: heavy metals, *Astacus leptodactylus*, crayfish, bioaccumulation.

Introduction

Depending on the water sites' location by the source of contamination, different levels of lead, cadmium and nickel accumulation have been found in the tissues of crayfish (7).

The concentration of heavy metals in the organs of the lake crawfish *Astacus leptodactylus* Eschscholtz, 1823 and other crayfish has been researched by 1, 3, 4, 6, 10, 11, 13 etc., but there are no data on the transfer of lead, cadmium and nickel in *Astacus leptodactylus* along the "water–crayfish" in the food chain.

The research was made in different zones of the Kardzhali dam which is anthropogenically influenced by heavy metals. The tributaries of Arda river, flowing into the Kardzhali dam, are near the Madan ore region, well-known for its lead/zinc lode and metasomatic ore enrichments that were one of the greatest deposits of those metals in Europe, intensely worked over the last 50 years (5). Besides this fact, the Lead/Zinc Plant is close to the city, which until recently functioned as a factory for the processing of polymetallic ore, Gorubso-Kardzhali Holding, whose main activity is the extraction and processing of gold-rich ores, and Bentonit Holding.

The goals of the study are to make an analysis of the water of the Kardzhali dam for the presence of lead, cadmium and nickel.

To determine the content of lead, cadmium and nickel in samples from the visceral organs of *Astacus leptodactylus* and to compare them to the maximum allowed concentrations according to accepted standards.

To determine the degree of bioaccumulation by the bioaccumulation coefficient (BC) via a formula that represents the ratio of the content of heavy metals in the organism and their content in the water.

Materials and methods

Water samples, taken from 5 points of the Kardzhali dam, were examined. The samples were analyzed after acid mineralization in closed containers under pressure when treated in a microwave oven.

The determination of trace content of metal ions was done with an ICP-MS (Agilent 7700). While still collecting the samples, measures were taken for canning by putting super pure HNO_3 (Suprapur® Merck, Darmstadt, Germany) into the collection containers in a 0,1 $mol.l^{-1}$ concentration.

To control the quality, each series of samples for analysis had a portion of certified material surface water SPS-SW Level 1 (Standards as, Oslo, Norway), added to it.

The crawfish for the present research were caught in the Kardzhali dam at the five points of the water extraction regions. Two parallel joint samples from the tissue of the lake crawfish *(Astacus leptodactylus)* were examined: liver and muscles.

The results are submitted in mg/kg (ppm) – average values from five pieces of replicas of instrumental measurement (N=5) and their respective relative standard deviation in percent (RSD%). The values under the limit of determination are given as < of the respective limit of determination (LOQ).

Results and discussion

The results of the analysis of the water in the five examined points of the Kardzhali dam are given in Table 1. They were compared to the Regulation on standards of quality of the environment for priority substances and some other contaminants (*SG*, issue 97 of 11.12.2015).

The research of the Kardzhali dam shows the concentrations of lead do not go over the received norms in four points, except the point in the area of the village of Glavatartsi (8.0 µg/l). For levels of maximum concentrations of 7.2 µg/l, an increase of 1.11 µg/l above the norms was reported.

The heavy metals cadmium and nickel do not show values over the maximum allowed concentrations (Table 1).

Table 1: Content of lead, cadmium and nickel in the water of the Kardzhali dam in µg/L

Chemical element	Sample	Village of Zvanika	Village of Dazhdovnitsa	The wall of the Kardzhali dam	Village of Enchets	Village of Glavatartsi
Lead	**µg/l**	0.75	0.34	0.81	1.21	8.0
MAC*7,2µg/l	*RSD %*	*1.8*	*2.7*	*0.6*	*0.9*	*1.1*
Cadmium	**µg/l**	0.03	0.02	0.03	0.03	0.04
MAC=0,5µg/l	*RSD %*	*13.6*	*11.9*	*9.2*	*12.2*	*4.9*
Nickel	**µg/l**	0.48	0.46	0.51	0.54	0.82
MAC =20 µg/l	*RSD %*	*2.9*	*4.2*	*7.8*	*4.5*	*3.2*

MAC – maximum allowed concentrations*

In Table 2, the results are displayed of researching the quantity of the heavy metals lead, cadmium and nickel in the liver and muscles of *Astacus leptodactylus*.

Table 2: Content of lead, cadmium and nickel in the muscles and the liver of Astacus leptodactylus from the Kardzhali dam in mg/kg

Chemical element (mg/kg)	**Muscles**		**Liver**		MAC (mg/kg)
	sample 1	sample 2	sample 1	sample 2	
Lead	0.065	0.25	0.06	0.08	0.50
Cadmium	< 0.05	< 0.05	0.65	0.58	0.5
Nickel	0.16	0.08	0.26	0.31	-

According to Regulation #31 (SG, issue 88 2004), maximum contaminant levels in foods are as follows: lead – 0.50 mg/kg, cadmium – 0.5 mg/kg, for nickel it is not indicated.

Bioaccumulation of lead, cadmium and nickel in the liver and muscles of the lake crawfish in compare of the maximum contaminant levels shows increased values of cadmium in the two samples from the liver. At levels of MAC of 0.5 mg/kg, an increase of 0.58 mg/kg and 0.65 mg/kg is reported (Table 2).

The quantities of lead, cadmium and nickel in the two analyzed organs shows a higher sorption effect with the liver. This can be explained with the parenchyma structure of the organ and its higher metabolic activity (9).

A similar bioaccumulation of cadmium is observed in the works of (7), according to which most heavy metals of the analyzed ones have been found in the gills and the liver of *Astaus leptodactylus*, and their lowest values are in the muscles. Such accumulation in the organism would cause different pathogenic states. It has been ascertained that heavy metals mostly afflict the hepatocytes (14), and in the case of subchronical poisoning with cadmium, the organs afflicted are basically the kidneys and the liver (2, 12).

In our study, the muscles do not show a high bioaccumulation affinity towards lead, cadmium and nickel, which indicates their lower sorption activity.

To ascertain the degree of bioaccumulation of liver and muscles towards lead, cadmium and nickel on the grounds of the results achieved, we used the bioaccumulation coefficient.

The bioaccumulation coefficient (BC) was calculated on the basis of the average content of lead, cadmium and nickel in the liver and the muscles of *Astacus leptodactylus*. Depending on its value, the hydrobionts are defined as macroconcentrators (BC>2), microconcentrators (BC= 1 to 2), and deconcentrators (BC<2) (as per 8).

The results for the bioaccumulation coefficient show that in the five examined zones of the Kardzhali dam the lake crawfish is with a proven bioaccumulation capacity chiefly towards cadmium and can be determined as a macroconcentrator of cadmium (Table 3). That fact also explains the high content of that metal in the liver as an organ participating in the metabolic processes of the organism.

Table 3: Bioaccumulation coefficient of lead, cadmium and nickel

Chemical element	Village of Zvanika	Village of Dazhdovnitsa	The wall of the Kardzhali dam	Village of Enchets	Village of Glavatartsi
Lead	0.1516	0.3345	0.1404	0.0940	0.0142
Cadmium	11.0833	16.625	11.0833	11.0833	8.3125
Nickel	0.4218	0.4402	0.3970	0.375	0.2469

Conclusion

Based on the present investigations, the following conclusions may be arrived at:

1. By the heavy metals analyzed in the water, a slight increase of lead is reported in one of the examined areas.
2. The distribution of the heavy metals lead, cadmium and nickel in the organ samples shows tendencies of the collection of cadmium mainly in the liver.
3. The three metals are distributed selectively in the organs, the liver having a tendency of higher sorption activity towards cadmium than the muscles.

4. The lake crawfish is classified as a macroconcentrator in respect of cadmium, for which BC is higher than 2.

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