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HEAVY METAL CONTENT IN THE BRANCHIAE OF SOME TISZA-RIVER FISH

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Abstract

The authors studied the Zn, Cu, Pb, Cd and Hg content in the branchiae of 132 fish from 18 species with atomic absorption and microscopic histology method. Random caught fish were divided into five groups according to thier alimentation habits. In the result of examinations the Zn, Cu, Pb, Cd and Hg concentration in the branchiae exhibited a decreasing order from Zn to Hg.

It was established by the authors that the examined metals, harmful for the environment, were not contained in the branchiae in a concentration warning of an acute danger, likewise the metal concentration of the Tisza river is relatively low: it does not reach neither the Hungarian, nor the international limit values. Knowing the metal content in the branchiae and on the basis of data in literature deductions could be made on the present quality of fish, which, as it could be stated, corresponds to the reguirements of alimentation hygiene from the point of view of the amount of metal ion concentration.

Introduction

The heavy metal content detectable in the body of fresh water fish mostly comes from two sources: it is taken directly from water and from food. Depending on the metal concentration of the water environment these metals — together with other harmful materials — accumulate in the body of the fish. (BADSHA—GOLDSPINK 1982, FÖRSTNER—PROSI 1979, HANNERZ 1968, MAY—MCKINNEY 1981). In surface waters free from toxic heavy metals the heavy metal concentration in the fish basically depends on the geochemical conditions prevailing in the river. A metal content larger than a given value exerts harmful effect on the physiological activity of plant and animal organizations which take part in the nutrition chain. In our country water biology methods studying long term effects (CSÉPAI—MRS. B. SÁRKÁNY) (Fig. 1) as well as parallel chemical analytical measurements are less known in spite of the fact that their importance is at least as great as that of methods revealing acute effects, since with these an early recognition and thus prevention becomes possible.

The metal content in the sediment of the Tisza river bed was first studied in the middle region of the river (WAIJANDT—MRS. SZABÓ 1980) and in a whole longitudinal section (LÁSZLÓ—BERTA 1981) in 1979. As regards the metal content of water there was published only one publication on its Hg concentration (MRS. M. SZABÓ—WAIJANDT 1983). We have no data in the literature on the heavy metal accumulation of Tisza water fish.

The aim of the present work was to obtain basic data by microscopic histologi-

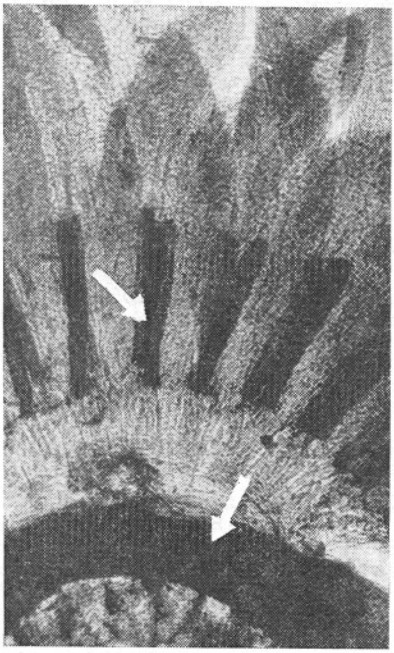


Figure 1. Details of the branchiae of *Xiphophorus helleri* toxidized by lead in an aquarium (CSÉPAI—Mrs. SÁRKÁNY 1983). In corresponding reaction media the darkening of the cartilage membrane around the arch and radii of the branchiae indicates the place of accumulation of lead. (Method: HAIDER's (1964); 60*)

cal and chemical analytical methods of the branchiae of some fish caught from the river Tisza near Szolnok reacting relatively sensitively to environmental harms (MRS. KOVÁCS—GAYER 1975, 1977, MATELEV—OSZETROV 1968, SCHULZ 1970). As a further aim we had a trial of the applicability of the two methods in our area.

Materials and Methods

Branchiae of 132 fish randomly caught from Tisza River near Szolnok, belonging to 18 species, of different age and sex were stored at -20 °C before the start of the investigations.

Microscopic histological study of the branchiae was carried out with a slightly modified Haider method (HAIDER 1964). It was experimentally stated that the heavy metal accumulation happens even in the hemibranchiae playing anatomically important part with the body of branchiae (Mrs. KOVÁCS-GAYER 1975, 1977) and the Haider method can be applied to detect this by microscopy. Taking into consideration the possibilities rendered by the method, because of their strong toxicity we preferred to detect Pb + Cd taken together and Hg selectively. The method is the following: a part of the hemibranchiae skeleton cleared of cells under a stereomicroscope, after repeated "washing" and separating steps corresponding to the detection of Pb + Cd, another part after preparations for detecting Hg were transformed by a chemical reaction into metal sulfide and then examined by light microscopy. The presence of the metal in question is indicated by the darkening of the longitudinal middle part of the skeleton. The intensity of the shade depends on the amount of the accumulated metal. Thus, it becomes possible to detect negative cases as well as the presence of the examined metal(s).

The bulk of samples dried for the chemical analysis was between 0.3 and 0.5 g, prepared according to the method described by KRISHNAMURTY (1976). Starting the studies comparative examinations were carried out with two fish species from 3—3 parallel samples, from an original sample in a teflonpadded bomb and from a dried sample with a reflux cooler, in order to control the possible Hg loss during drying and clearing. Values obtained by the latter method in the case of *Esox 1.* were a mean 96.2%, in case of *Hypophtalamichthys m.* 93.4% for the value measured with the bomb. These results agree with those measured by PAUS (1972). In view of the above mentioned we disregarded a detailed exposure of the samples in order to detect mercury. The zinc, copper and cadmium concentration of the exposed samples was measured by flame atomization method, lead was measured with an electrothermal atomizator by an AAS—3 atomic absortpion apparatus (Zeiss product, field correction). Hg content was determined by a Spektromom 190A apparatus with the so-called colsdteam method (HATCH-OTT 1968, PUNGOR MRS. GROF 1973).

Results and Evaluation

The intake and bioaccumulation of heavy metals is influenced by several factors. In case of fish water and food are decisive for the metal intake. Water as living space and carrier for a long exposition time carries metals into the organism first of all through the branchiae and through the skin. Food of plant and animal origin can contain metal ions to various extent. Absorption largely depends on the composition of food, the degree of oxidation of the contained metals, the acidity of gastric juices and the metal content of the organism.

The same way of respiration of the fish does not yield a corresponding basis for classification. Different forms of alimentation characterizing the particular species give a reasonable basis for grouping according to alimentation habits. Accordingly the examined fish species were divided into five groups (Table 1).

Zinc

Of the five examined metals zinc showed the greatest concentration, in harmony with the concentration of this metal in water and sediments. For most species the bulk of values was around 100 µg/l (Figure 1a. Names of the fish are given in Table 1). With the exception of three species *(Cyprinus c., Carassius a.g., Esox 1.)* there is no remarkable difference in the results. The Zn content in the branchiae of Balaton *Abramis b.* and *Stizostedion* 1. (SALÁNKI 1981, V. BALOCH—SALÁNKI 1986) shows a good agreement with that found by us in the Tisza species (Table 2).

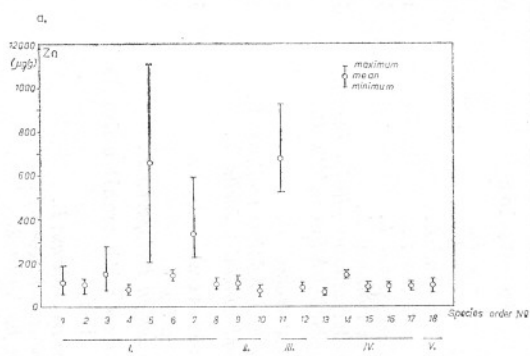


Fig. 1. Metal content in the branchiae of the examined species in the Tisza river (µg metal/g dry branchiae)
a) Zn

Table 1. *Heavy metal content in the branchiae of fish species from the Tisza river and results of microscopic histological investigation* (*μg metal/g dry branchiae;* C_V=*relative scattering*)

No. of spec.	Fish species	n/ piece	Total body weight (g)	Zn Mean	Zn C_V	Cu Mean	Cu C_V	Pb Mean	Pb C_V	Cd Mean	Cd C_V	Pd+ Cd tissue bio-log. invest.	Hg Mean	Hg C_V	Hg. tissue biolog. invest.
I.	Mixed alimentation	63													
1.	*Abramis brama L.*	5	350—1500	104	0.49	3.5	0.35	3.0	0.28	0.85	0.68	$+_5$	0.42	0.62	$+_2$
2.	*Leuciscus idus L.*	4	400—1200	97	0.32	5.7	0.31	3.3	0.35	0.57	0.33	$+_4$	0.28	0.41	–
3.	*Blicca bjoerkna L.*	6	130—300	149	0.49	7.2	0.41	3.8	0.30	2.67	0.52	$+_6$	0.62	0.73	$+_3$
4.	*Barbus barbus L.*	12	500—2300	74	0.19	3.4	0.24	4.4	0.58	0.66	0.34	$+_{12}$	0.30	0.31	–
5.	*Cyprinus carpio L.*	12	800—5000	654	0.47	6.6	0.51	3.5	0.34	1.04	0.57	$+_{12}$	0.29	0.49	$+_2$
6.	*Pelecus cultratus L.*	3	200—350	138	0.14	9.1	0.27	2.9	0.40	1.49	0.42	$+_3$	1.29	0.37	$+_3$
7.	*Carassius auratus gibelio* Bloch	12	150—750	324	0.33	4.1	0.33	4.3	0.32	1.25	0.85	$+_{12}$	0.44	0.38	$+_4$
8.	*Hypophtalmichthys nobilis R.*	9	550—15 800	97	0.19	5.1	0.56	5.3	0.43	0.53	0.54	$+^9$	0.35	0.46	$+_1$
II.	Plantivorous	19													
9.	*Hypophtalmichthys molitrix V.*	11	700—11 000	98	0.23	5.6	0.27	2.6	0.34	0.48	0.73	$+_{11}$	0.21	0.34	–
10.	*Ctenopharingodon idella V.*	8	1000—10 000	68	0.28	3.2	0.27	2.8	0.56	0.48	0.92	$+^8$	0.14	0.91	
III.	Carnivorous	19													
11.	*Esox lucius L.*	10	500—2400	670	0.24	2.8	0.20	3.6	0.74	0.51	0.47	$+_{10}$	0.52	0.23	$+_6$
12.	*Stizostedion lucioperca L.*	9	800—6000	80	0.20	2.4	0.23	4.2	0.49	0.35	0.46	$+^9$	0.55	0.44	$+_4$
IV.	Feeding with small animals and fish	28													
13.	*Acipenser ruthenus L.*	7	380—810	62	0.18	3.8	0.23	6.7	0.41	0.79	0.41	$+_7$	0.43	0.24	$+_2$
14.	*Ictalurus nebulesus L.*	3	70—120	141	0.13	8.3	0.07	3.1	0.41	3.80	0.46	$+_3$	4.26	1.51	$+_2$
15.	*Perca fluviatilis L.*	3	130—250	87	0.25	3.2	0.22	5.9	0.28	0.83	0.36	$+_3$	0.64	0.37	$+_2$
16.	*Silurus glanis L.*	10	800—18 000	76	0.19	2.3	0.45	4.6	0.40	0.28	0.27	$+_{10}$	0.26	0.20	–
17.	*Aspius aspius L.*	5	550—1850	95	0.16	5.3	0.36	3.0	0.23	1.04	0.25	$+_5$	0.64	0.40	$+_2$
V.	Feeding with small animals	3													
18.	*Tinca tinca L.*	3	100—500	94	0.33	4.0	0.11	6.7	0.61	0.69	0.24	$+_3$	0.86	0.69	$+_2$

* = Designation according to (MSZ 1977)
** + positive number of cases
– negative result

Table 2. *Comparison of results of studies of the Tisza river with data in literature (µg metal/g dry branchiae)*

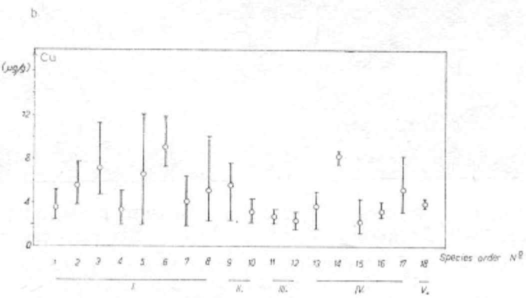
Place of Origin	*Abramis brama* L.					*Esox lucius* L.		
	Zn	Cu	Pb	Cd	Hg	Zn	Pb	Cd
Ellesmere (BADSHA-GOLDSPINK 1977)	367	—	5.2	7.8	—	—	—	—
Compstall (BADSHA-GOLDSPINK 1977)	147	—	17.5	11.2	—	245	4.0	1.8
Balaton (SALÁNKI et al. 1981)	155	8.0	13	1.4	0.42	—	—	—
Tisza	104	3.5	3.0	0.85	0.42	670	3.6	0.51

	Perca fluviatilis L.		
	Zn	Pb	Cd
Ellesmere (BADSHA—GOLDSPINK 1977)	158	19.5	20.8
Tatton (BADSHA—GOLDSPINK 1977)	125	15.1	1.6
Tisza	87	5.9	0.83

	Stizostedion luciuperca L.				
	Zn	Cu	Pd	Cd	Hg
Balaton (SALÁNKI et al. 1981)	95	4.9	5.4	0.80	0.31
Tisza	80	2.4	4.2	0.35	0.55

Copper

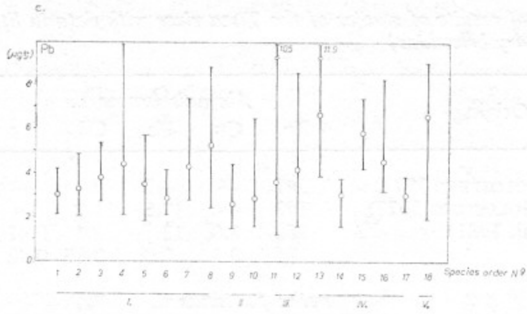
The results suggested that the Cu content in the branchiae of the particular species of fish is almost independent of the alimentation habit itself. Even within the groups the values relatively widely scatter (Fig. 1b). The highest values were found with some kinds of fish of mixed alimentation *(Blicca b., Cyprinus c., Pelecus c.)*, but we can mention the greater concentration experienced with the plantivorous *Hypophtalmichthys m.* and *Aspius a.* regarded as feeding with small animals and fish. We also mention that in the case of *Abramis b,* and *Stizostedion 1.* the concentrations were smaller (Table 2) than in the Balaton kinds (SALÁNKI 1981, V. BALOGH—SALÁNKI 1986).



b) Cu

Lead

We found lead concentration in a relatively wide range within the single alimentation group as well as in the case of some species (Fig. 1c). The highest values could be measured in *Barbus b., Hypophtalmichthys n., Esoc 1.* and *Acipenser r.*



c) Pb

The lowest mean values were found in the two plantivorous species, however, the sample number is small here. It is well known that lead is toxic for living organisms. According to Kőrös cadmium, similar to mercury, is bound to sulphur containing ligands e.g. cystein side chains of proteins, thus it blocks the work of a number of enzymes (Kőrös 1980).

We dealt with the histological study of Pb and Cd taken together because these two metals cannot be separated from each other with the mentioned method. Values for lead concentration are as a rule one order of magnitude greater than for cadmium. Therefore on the basis of microscopic picture we could undertake only a qualitative evaluation. Accordingly we got a positive result in the examined species and individuals whereby, except *Ictalurus n.* the quantitaive effect of lead dominated over cadmium (Figure 2).

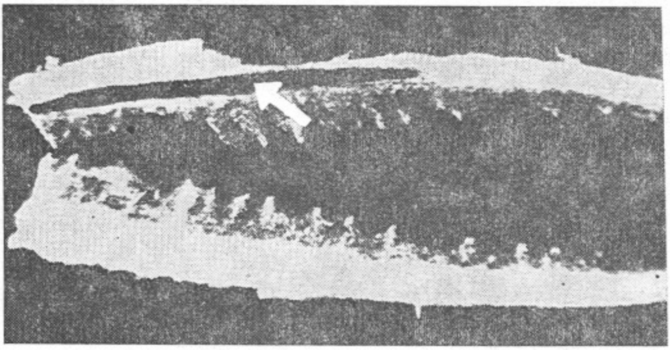
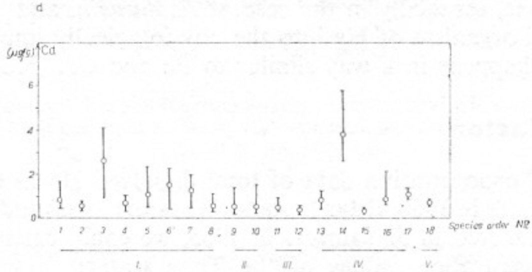


Figure 2. Left: Hemibranchia without respiratory epithelium, prepared from Tisza *Cyprinus* c. branchia, shown in plane "C". Darkening of a part of the cartilageous radius shows the accumulation of Pb+Cd together. Right: Detail of a hemibranchia of a control *Cyprinus c.* bred in an aquarium; negative for heavy metals.
(Method: Haider's (1964); 50*)

Cadmium

Similar to zinc cadmium concentrations changed in a narrow range with the exception of two species (Fig. 1d). Branchiae of *Blicca b.* and *Ictalurus n.* exhibited remarkably greater values. In the formation of nearly the same concentrations there may play a role the relatively higher cadmium content of the sediment, what is due to



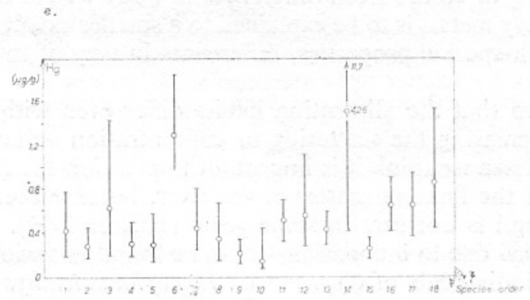
d) Cd

geochemical reasons (László—Berta 1981, Waijandt—Mrs. Szabó 1980). The Cd concentration of the water, however, is low.

The cadmium content in the branchiae of Tisza *Abramis b.* was lower than the values found with the Balaton and Zala river species, similarly this content remained below that of the Balaton values (Salánki 1981). Results of microscopic studies related to this metal are treated under the heading of lead. The mechanism of binding and accumulating of cadmium, toxic for living organisms, is similar to that of lead. It can build in the place of zinc without substituting the biological function of the zinc-protein complex.

Mercury

Of the examined metals the concentration of mecrury changed in the widest range (Fig. 1e). Highest values were found in the branchiae of *Ictalurus n., Blicca b.,*



e) Hg

Pelecus c. and *Tinca t.* Lowest concentration values were found with species belonging to the plantivorous (Table 1).

For *Abramis b.* caught in Balaton and the Zala river the concenration values were similar, (V. Balogh—Salánki 1986), while the mercury content in the branchiae of Tisza *Stizostedion 1.* was somewhat over that measured in Balaton *Stizostedion 1.* (Salánki 1981) (Table 2).

No mercury positive reaction was found in the histological studies below the value 0.29 μg/g, while over this a mercury positive reaction was characteristic with some exceptions, especially in the case of *Ictalurus n.* and *Tinca t.* (Table 1). The building in the organism of Hg into the physiologically important mostly Zn-protein complexes happens in a way similar to Pb and Cd. (KŐRÖS 1980).

Concentration factor

Making use of concentration data of total dissolved Hg in the water of Tisza measured in 51 samples in 1980. (MRS SZABÓ—WAIANDT 1983) and of total dissolved Zn, Cu and Cd measured in 52 samples in 1985, we could calculate concentration factors. Mean concentration values of the Tisza water:
Hg: 0.12 μg/l, Cd: 0.37 μg/l, Cu: 4.4 μg/l, Zn: 27 μg/l. The value for the concentration factor of Zn is $2.3 \cdot 10^3$—$2.4 \cdot 10^4$; of Cu: $5.2 \cdot 10^2$—$2.1 \cdot 10^3$; of Cd $7.6 \cdot 10^2$—$1.0 \cdot 10^4$; of Hg: $1.2 \cdot 10^3$—$3.6 \cdot 10^4$. Considering the values of the particular alimentation groups we got the following set of concentration factors:

$$Hg > Zn > Cd > Cu$$

In spite of the fact that we have only one organ in view, the range between extreme values is astonishingly wide. At the same time the mean value for the single metals (Hg: $5.6 \cdot 10^3$; Zn: $3.8 \cdot 10^3$; Cd: $2.2 \cdot 10^3$; Cu: $1.0 \cdot 10^3$) was not markedly different.

Conclusions

There is a number of data in the literature showing that heavy metals accumulate to a different extent in different parts of the fish. (BADSHA—GOLDSPINK 1982, METHIS—CUMMINGS 1973, SALÁNKI 1981).

A comparative analysis of our results suggests that the accumulation in the branchiae tissues of the Tisza river fish is not an unequivocal process happening directly via breathing or comes from differences in body weight and measures. The accumulation of heavy metals is to be explained to a smaller extent with breathing, to a greater extent with specific properties, differences in way of living and alimentation.

It is well known that the alimention habits differ even within one genus. All these are factors increasing the scattering of concentration values.

In the case of Tisza we think it is important to mention the fairly extreme concentration values of the floating matter in the river, because even a change in the range of 1—2000 mg/l is not rare in some years (BANCSI 1977). Very fine grained floating matter formed due to inundations could be found in various amounts in the branchiae of some individuals of *Cyprinus c.*, *Hypophtalmichthys m.* and *Carassius a.g.* This floating mater of uncertain amount raised the real metal concentration to an unceratin extent. That is why the obtained metal content results for some species can be regarded only as an orientation indicating the metal concentration of the river.

Summarizing it can be stated that the level of metals contaminating the environment in the water of the Tisza river is relatively low. It is markedly lower than the international level (RHEINBERICHT 1978) or what is regarded as dangerous in our country (Technical Data by the National Water Directory 1985).

In the present study we did not want to examine fish from the point of view of nutrition. There are data in the literature that a unit amount of fish contains 3—5 times less metals than the branchiae (BADSHA—GOLDSPINK 1982, MATHIS—CUMMINGS 1973). Accordingly we can suppose that the metal content in fish exhibits lower values than recognized as acceptable (Egészségügyi Közlöny 1975).

Tiszai halak kopoltyújának nehézfém tartalma

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Kivonat

A szerzők 132 db, a Tisza folyó szolnoki térségéből kifogott, 18 féle fajú hal kopoltyújának Zn, Cu, Pb, Cd és Hg tartalmát vizsgálták atomabszorpciós és mikroszkópos szövettani módszer segítségével. A random-szerűen kifogott halakat táplálkozási szokásaik szerint 5 csoportba sorolták. Vizsgálati eredményeikben a halkopoltyúk Zn, Cu, Pb, Cd és Hg koncentrációja a Zn-től a Hg felé csökkenő értéket mutattak.

A szerzők megállapították, hogy a kopoltyúk a vizsgált környezetszennyező fémeket nem tartalmazzák akut veszélyt jelző koncentrációban, mint ahogyan a Tisza vizének fémkoncentrációja is viszonylag alacsony; nem éri el a magyar és nemzetközi határértékeket. A kopoltyúk fémtartalmának ismeretéből irodalmi adatok alapján következtetni lehet arra is, hogy a halhús minősége jelenleg a fém-ionok koncentrációjának mennyisége szempontjából megfelel az élelmezés-egészségügyi követelményeknek.

Содержание тяжелых металлов в жабрах рыб Тисы

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Резюме

Авторы анализировали содержение меди, кадмия, цинка, хрома и ртути в жабрах 8 видов рыб с помощью атомноабсорпционного и микроскопического гистологического метода. Они группировали выловленных безвыборочно рыб в 5 групп по их привычкам питания. В результатах выясняется, что содержание меди, кадмия, цинка, хрома и ртути в жабрах исходя из меди и заключая ртутью показывает снижающиеся стоимости.

Авторы установили, что жабры не содержат таких металлов в концентрации, которая показала бы острую опасность; ситуация аналогична касательно концентрации металлов воды Тисы, она не достигает венгерских и международных предельных величин.

На основании литературных данных и данных о содержании металлов жабер можно с делать вывод, что качество мяса рыб в настояшее время соответствует санитарным требованиям на основе количества концентрации ионов металлов.

Sasržaj teških metala u škrgama riba srednje Tise

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Izvod

Autori su histološkim metodama atomskoabsorpcionim i svetlosnim mikroskopijom ispitivali prisustvo Zn, Cu, Pb, Cd i Hg u škrgama 132 jedinki, pripadnika 18 vrsta riba reke Tise sa područja Solnok. Random-postupkom izlovjene ribe razvrstavane su u pet grupa, na osnovu tipa ishrane.

Utvrdjeno je da koncentracija teških metala u škrgama riba pokazuje tendencniju smanjivanja vrednosti od Zn prema Hg. Takodje autori konstatuju da utvrdjena koncentracija ispitivanih teških metala u škrgama riba ne ukazuje na akutnu opasnost, samim tim što je koncentracija metala i u vodi Tise relativno niska, i nalazi se ispod madjarskih i medjunarodnih normi. Autori smatraju da riblje meso po kvalitetu i prema literaturnim podacima u odnosu na konstatovane količne metala u škrgama riba prema jonskim koncentracijama, udovoljava prehranbenim zdravstvenim zahtevima.