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SOME METALS IN THE ORGANS OF FISH IN LAKE GARDNO

JAN TROJANOWSKI[1], PIOTR TROJANOWSKI[2], CZESŁAW JAŃCZAK[1], JÓZEF ANTONOWICZ[1]

[1]Department of Environmental Chemistry, Pomeranian Pedagogical University, Arciszewskiego 22, 76-200 Słupsk, Poland
[2]Mental Clinic "Dziekanka", Gniezno, Poland

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WYBRANE METALE W NARZĄDACH RYB JEZIORA GARDNO

Dominującymi gatunkami ryb w jeziorze Gardno są leszcz, okoń i płoć. W mięśniach, wątrobie i skrzelach tych ryb badano zawartość Cu, Cd, Pb, Mn, Zn i Fe. Badane narządy w największej ilości zawierały żelazo (5,35–57,38 μg g^{-1} mokrej masy) i cynk (3,98–23,93 μg g^{-1}) niezależnie od gatunku ryby. Najmniejszą zawartością charakteryzował się kadm (0,002–0,168 μg g^{-1}). Spośród badanych tkanek wątroba wykazywała największe zdolności kumulacyjne analizowanych metali. Ich zawartość w wątrobie była kilka lub kilkanaście razy większa niż w mięśniach. Koncentracja manganu, miedzi, cynku i ołowiu w mięśniach badanych leszczy zmniejszała się wraz ze wzrostem ich masy. Natomiast odwrotną zależność obserwowano w przypadku żelaza i kadmu. Przyrostowi masy badanych ryb towarzyszył wzrost stężenia tych metali. Stwierdzono, że zawartość miedzi, żelaza i kadmu w skrzelach w sposób istotny wpływała na ich kumulację w mięśniach i wątrobie.

Summary

The total concentrations of Cu, Cd, Pb, Mn, Zn and Fe in the tissues (muscles, gills, liver) of fish from Lake Gardno were studied. Iron (5.35–57.38 μg g^{-1} wet wt.) and zinc (3.98–23.93 μg g^{-1}) were present in highest concentrations in all tissues, irrespective of fish species. Cadmium was present in the lowest concentrations (0.002–0.168 μg g^{-1}). Among studied tissues, the liver had the greatest capacity to accumulate metals. Their content in the liver was several to a dozen or sometimes even more times higher than in the muscles. The content of manganese, copper, zinc and lead in the muscles of the studied bream decreased with weight, whereas the content of iron and cadmium increased with weight. The content of copper, iron and cadmium in the gills considerably influenced the level of accumulation of those metals in muscles and liver.

INTRODUCTION

Heavy metals are easily involved in the matter circulation and are characterized by a high bioaccumulation coefficient [12, 19, 24, 29]. Water pollution with heavy metals is reflected in their level in fish [10, 11]. The sources of those microelements for fish are water and food. Aquatic organisms take heavy metals directly from water or indirectly through trophic chain. In general, metals in the dissolved form as well as adsorbed on suspended particles are taken from the water by alimentary canal.

Chemical form in which heavy metals are present in water determines their quantity absorbed by fish directly from water. The fish during feeding take up elements built into the tissues of consumed organisms and adsorbed on their surface [19, 25]. The contribution of elements coming from water differs and depends on the species and age of fish, and on the form of organism. In fish, the largest amounts of toxic elements get into their organism through gills or through alimentary canal [2]. The results of research showing the differences between fish species from different trophic levels of the same water region indirectly confirm the effect of food on the level of heavy metals in fish [4, 18, 20, 21]. Heavy metals taken from the environment are bound by metalproteins and used in metabolic processes or are accumulated in the liver.

Measurements of heavy metal concentrations in different components of an ecosystem show the possibility of the pollution of Lake Gardno with those metals [27]. The anthropogenic origin of different metals in abiotic components of the ecosystem was determined using geo-chemical coefficients [24].

The aims of our research were:

1. the determination of heavy metal bioaccumulation in muscle tissues and in some organs participating in absorption, voidance and accumulation of microelements;
2. the comparison of lead, cadmium, copper, zinc, manganese, and iron concentrations in the tissues of chosen fish species of different size;
3. the determination of the origin of heavy metals in the lake ecosystem;
4. the comparison of heavy metal contents in fish from the studied lake with the same species from the southern part of the Baltic Sea and other inland reservoirs.

STUDIED AREA

Lake Gardno is situated in the central part of the Słowiński Sea-coast. This lake is rather shallow, and therefore it belongs to a polimictic type of lake (the average depth – 1.6 m, max. – 2.6 m). The lake's surface is 24.681 km^2. Its bottom is rather flat, covered with a lot of sludges and sediments. Small bottom surfaces have hard sand floor, mainly in its north-east part and near Kamienna Island.

The land 0.8–2 km wide separates the lake from the Baltic coast. This land forms a spit with different sand eminent covered with pine trees. The marsh and peat lands surround the lake from the south-west and from the east. The River Łupawa, as a main tributary falling into the lake in its east part, supplies the water. In its north-eastern part, Lake Gardno is connected with Lake Łebsko by a channel which is about 8 km long. The difference of water levels between the lake and the sea facilitates the reciprocal exchange of water through a mouth of the River Łupawa. No large rivers such as the Bagiennica, the Grabownica and the Broda flow into the lake.

FISH IN LAKE GARDNO

The lake's ichtiofauna is very rich and diversified. Lake Gardno is a beam-type lake. In its brackish waters there are species of fresh-water fish that tolerate law salinity levels as well as sea fish that tolerate the level of salinity lower than that in the open sea. The most abundant species of fish are: bream (*Abramis brama*), roach (*Rutilus rutilus*), eel (*Anguilla anguilla*), zander (*Stizostedion lucioperca*) and perch (*Perca fluviatilis*) [26]. The following sea fish are also present: stunted flatfish (*Pleuronectus fleus*), Baltic lavaret (*Coregonus lavaretus*) and garfish (*Belone belone*). The presence of sea lamprey (*Petromyzon marinus*), Baltic barnacle (*Balanus baltica*) and the moluscs: Baltic cockle (*Cardium glaucum*), Baltic molusc (*Macona baltica*), and blue mussel (*Mytilus edulis*) emphasizes the partially marine character of the biocoenosis of this in-principle fresh-water lake [17].

MATERIAL AND METHODS

Fish to be studied were acquired during the autumn and spring catch of fishes in 2001. Bream, roach and perch have been chosen because of their large population size. The fish samples contained 196 bream, 60 roach and 42 perch. Among compared bream there were 94 males and 102 females, among reach there were 38 males and 22 females, whereas among perch there were 12 males and 30 females. Bream (*Abramis brama*) were segregated; the groups of similar weight and length were chosen and finally divided into males and females. Among the rest of fish species / roach (*Rutilus rutilus*) and perch (*Perca fluviatilis*)/ the specimens of similar weight were also chosen. Table 2 shows the average values for all specimens in a given group. Muscles were obtained from the central over-axis part of a great side muscle. Branchial (gill rakers, gill arch and gill filaments together) and the liver were obtained as a whole.

Samples to be examined were taken from different fish species. The samples weighed from 2 g to 5 g of material dried in the temperature of 105°C. The contents of heavy metals in muscles, gills and the liver were examined. The samples were wet-mineralized in the mixture of nitric(V) acid and chloric(VII) acid. Acids were evaporated after the mineralization. So obtained dried residue was dissolved in 10 cmł of 0.2 M HCl solutions. The concentration of cadmium, zinc, manganese, lead, copper and iron were determined in this solution. Determinations were made using AAS flame absorption atomic spectrometry method. Work parameters of the apparatus were the most favorable for a given element. The wave length, slot width and lamp current intensity were set up according to producer's recommendations. The content of heavy metals in the sample was calculated using calibration curve. The results of the analyses are given in μg g^{-1} on a wet-weight basis.

Enrichment factors with those metals were calculated for tissues basing on the values of trace element concentrations in the examined tissues. The aim was to determine the origin of the metals in the ecosystem. Enrichment factor is defined as a quotient of a given metal concentration /me/ in studied tissues of fish $(C_{me})_s$ and in the earth's crust $(C_{me})_{ec}$ [24]:

$$EF = \frac{(C_{me}/C_{Fe})_s}{(C_{me}/C_{Fe})_{ec}}$$

where: C_{Fe} – iron concentration.

The obtained results were compared by the analysis of variance (ANOVA). The statistically significant results of the ANOVA were further analyzed with the Tukey Test. Significance of the correlation between two variables was assessed with Person's linear correlation coefficient.

RESULTS AND DISCUSSION

The present research concerned three fish species: perch, bream, and roach. Table 1 shows average concentrations of copper, cadmium, lead, manganese, zinc and iron in bream's muscles, gills and liver in relation to the weight of the fish. The concentration of examined metals in bream changed according to its weight and analyzed tissue. Muscle tissue of small specimens contained much more heavy metals than the same tissue in big fish of the same species. The opposite phenomena were observed only in the case of cadmium and iron.

Table 1. Average content of heavy metals (in μg g^{-1} wet weight) in muscles, gills and liver of bream (*Abramis brama*) from Lake Gardno in relation to their weight

Number of fishes		24	22	25	20	23	18	18	16	15	15
Species of tissue	Metals	to 200 g	200–400 g	400–600 g	600–800 g	800–1000 g	1000–1200 g	1200–1400 g	1400–1600 g	1600–1800 g	1800–2000 g
Muscles	Cu	0.53	0.49	0.38	0.40	0.43	0.41	0.36	0.37	0.29	0.31
Gills	Cu	4.48	4.56	3.99	0.65	2.84	2.98	2.65	2.70	1.19	0.89
Liver	Cu	7.56	8.62	6.90	0.86	6.31	4.98	3,37	3.65	2.10	0.88
Muscles	Cd	0.035	0.033	0.042	0.036	0.041	0.055	0.043	0.062	0.065	0.068
Gills	Cd	0.071	0.075	0.059	0.078	0.117	0.108	0.073	0.091	0.098	0.077
Liver	Cd	0.086	0.072	0.067	0.096	0.151	0.153	0.112	0.118	0.145	0.136
Muscles	Pb	0.41	0.37	0.28	0.24	0.39	0.30	0.26	0.22	0.21	0.24
Gills	Pb	0.89	1.02	0.98	1.10	1.16	1.09	1.29	1.67	1.89	1.79
Liver	Pb	1.01	1.26	1.34	1.56	1.68	1.99	1.88	2.06	2.15	2.59
Muscles	Mn	0.48	0.21	0.26	0.26	0.20	0.19	0.11	0.13	0.18	0.19
Gills	Mn	0.67	0.56	0.72	0.99	0.97	0.86	0.53	0.84	0.63	0.88
Liver	Mn	0.61	0.99	1.33	1.28	1.20	1.87	1.14	0.89	1.22	1.44
Muscles	Zn	6.25	7.65	6.92	5.20	4.18	3.52	3.22	3.97	3.19	3.37
Gills	Zn	10.05	13.13	8.79	14.99	11.83	15.89	14.62	17.40	20.36	16.83
Liver	Zn	16.29	15.38	14.87	23.30	28.38	26.98	27.65	26.91	28.60	27.15
Muscles	Fe	2.19	1.88	3.22	4.21	2.02	4,.0	6.35	3.88	8.82	6.04
Gills	Fe	1.1	28.5	29.9	23.6	23.7	50.2	44.5	58.0	45.2	64.4
Liver	Fe	37.8	27.5	37.1	51.8	57.6	43.1	68.2	58.0	73.3	79.4

The concentration of copper in the bream's muscle tissue varied between 0.58 μg g^{-1} wet wt. in the group of specimens weighing up to 200 g, and 0.25 μg g^{-1} wet wt. in the specimens of the same species weighing 1400–1600 g. The concentration of copper in the bream's gills ranged from 5.11 μg g^{-1} to 0.87 μg g^{-1} and in the liver it ranged from 9.22 μg g^{-1} to 1.73 μg g^{-1} (Tab.2). The average copper content in the muscle tissue of studied bream amounted to 0.39 μg g^{-1}, in gills to 3.09 μg g^{-1} and in the liver to 5.32 μg g^{-1}. Differences between those averages are statistically highly significant. Differences between concentrations of that metal in the muscles of bream, roach and perch were rather small and statistically not significant, whereas differences between concentrations of metals in gills and the liver of the same species were statistically highly significant. The gills of bream contained the highest amounts of copper (3.09 μg g^{-1}), whereas the gills of roach contained the smallest amounts (1.99 μg g^{-1}); the liver of perch contained the most (5.72 μg g^{-1}) and the liver of roach the least copper (4.88 μg g^{-1}).

Table 2. The content of heavy metals (in μg g^{-1} wet weight) in muscles (A), gills (B) and liver (C) of fish from Lake Gardno (X – mean value, S – standard deviation, x_{min}, x_{max} – minimum and maximum values)

Metals	Indicator	Bream			Roach			Perch		
		A	B	C	A	B	C	A	B	C
Cu	X	0.39	3.09	5.32	0.46	1.99	4.88	0.43	2.48	5.72
	S	0.06	0.28	1.04	0.08	0.37	0.95	0.10	0.21	1.54
	x_{min}	0.25	0.87	1.73	0.38	0.72	1.83	0.30	0.64	2.06
	x_{max}	0.58	5.11	9.22	0.63	4.24	904	0.62	4.35	9.59
Cd	X	0.044	0.085	0.119	0.033	0.061	0.108	0.038	0.082	0.103
	S	0.010	0.017	0.033	0.011	0.015	0.025	0.006	0.018	0.032
	x_{min}	0.019	0.046	0.075	0.007	0.039	0.078	0.002	0.051	0.045
	x_{max}	0.070	0.130	0.152	0.060	0.89	0.134	0.069	0.120	0.168
Pb	X	0.29	1.29	1.74	0.45	0.80	1.45	0.24	0.88	1.61
	S	0.07	0.31	0.42	0.05	0.18	0.36	0.09	0.17	0.15
	x_{min}	0.18	0.68	0.78	0.21	0.47	0.96	0.09	0.34	1.06
	x_{max}	0.46	2.06	3.03	0.70	1.18	2.01	0.38	1.37	2.08
Mn	X	0.21	0.75	1.27	0.16	0.55	0.89	0.24	0.66	1.20
	S	0.08	0.10	0.27	0.04	0.10	0.31	0.05	0.11	0.23
	x_{min}	0.04	0.15	0.56	0.07	0.24	0.52	0.09	0.25	0.67
	x_{max}	0.41	1.20	1.20	0.29	0.81	1.18	0.41	0.92	1.72
Zn	X	4.47	14.39	23.55	5.84	11.87	23.93	3.98	10.55	17.31
	S	0.65	2.06	3.62	0.84	1.45	4.36	0.81	1.28	2.28
	x_{min}	2.85	9.92	12.60	3.79	9.58	19.57	2.03	8.26	14.92
	x_{max}	7.88	23.57	30.25	8.02	13.64	26.94	5.16	13.07	20.18
Fe	X	5.35	38.84	57.38	6.31	24.56	45.72	5.51	30.91	49.84
	S	0.47	5.51	11.31	0.54	1.87	3.06	0.51	2.89	4.08
	x_{min}	1.66	13.23	25.48	1.52	9.22	20.52	0.95	10.24	16.71
	x_{max}	9.31	65.11	82.17	9.85	45.80	68.41	8.85	54.05	87.54

The copper content in bream muscles, gills and the liver decreases with weight. This relationship between copper content and specimen weight was characterized by a correlation coefficient of 0.89 and was curvilinear. It was described by the regression equation shown in Table 3. Much higher correlation coefficients were obtained for gills and the liver. The relationship between copper level in those tissues and fish mass was rectilinear. The liver contained almost fourteen times more copper than the muscles. A similar relationship was observed by Kostecki [13] in fish from the anthropogenic reservoir Dzierżno Duże and Usero et al. [28] in fish from salt marshes on the southern Atlantic.

Generally, the observed concentrations of copper were comparable with those described in the literature. To compare, the copper content in the muscles of bream from Szczecin Bay in 1984 ranged from 0.28 to 0.54 $\mu g\ g^{-1}$ [19]; in the muscles of bream from the Odra estuary in 1995 it amounted to the average 0.459 $\mu g\ g^{-1}$ [12], whereas in the muscles of bream from ponds of Barycza river it oscillated between 0.24 and 0.50 $\mu g\ g^{-1}$ [16]. Falandysz [6] determined the average content of copper in bream to be 0.26 $\mu g\ g^{-1}$, in perch 0.42 $\mu g\ g^{-1}$, whereas Kostecki [13] determined it to be 0.98 $\mu g\ g^{-1}$ in perch and 1.12 $\mu g\ g^{-1}$ in roach.

The cadmium content in the muscles of bream ranged from 0.019 to 0.076 $\mu g\ g^{-1}$ (Tab. 2) and was placed in the range 0,003–0,170 $\mu g\ g^{-1}$ observed for bream by Hłyńczak et al. [12]. Weight groups up to 200 g and from 200 to 400 g were characterized by the least cadmium concentration – on average 0.034 $\mu g\ g^{-1}$ and weight group 1800–2000 g had the highest cadmium concentration – 0.068 $\mu g\ g^{-1}$. The average Cd content in the muscles of bream (0.044 $\mu g\ g^{-1}$) is comparable to the average Cd content in the muscles of carp examined by Protasowicki and Chodyniecki [20] but it is lower than in the muscles of cod from the south part of the Baltic Sea (0.072 $\mu g\ g^{-1}$, [19]). A slightly lower concentration of cadmium was determined in the muscles of roach (0.033 $\mu g\ g^{-1}$) and perch (0.038 $\mu g\ g^{-1}$). A considerably higher level of that metal was observed by Kostecki [13] in perch muscles (0.067 $\mu g\ g^{-1}$) and similar concentration in roach muscles (0.033 $\mu g\ g^{-1}$). Kremer et al. [15] found too high level of cadmium (0.071 $\mu g\ g^{-1}$) in perch muscles.

An increase in the content of that metal with mass was also observed in gills and the liver; however the changes were not so unequivocal as it was for muscles. At the same time no statistically significant differences between Cd concentration in gills (on an average 0.085 $\mu g\ g^{-1}$) and in the liver (on an average 0.103 $\mu g\ g^{-1}$) were observed. The relationship between cadmium concentration and weight was curvilinear in gills and rectilinear in the liver (Tab. 3). Correlation coefficients were also statistically significant. The smallest average cadmium concentration in both gills and the liver was determined in the group of 400–600 g weight and amounted to about 0.058 $\mu g\ g^{-1}$. The highest concentration was determined in the group of 800–1000 g weight (gills) and amounted to 0.117 $\mu g\ g^{-1}$ and in the1000–1200 g group (the liver) and amounted to 0.143 $\mu g\ g^{-1}$. The average content of that metal in the liver of studied bream amounted to 0.119 $\mu g\ g^{-1}$ (0.045–0.168 $\mu g\ g^{-1}$) and was in the lower parts of the range of its concentration 0.051–0.400 $\mu g\ g^{-1}$ in the liver of fish of the same species in Szczecin bay [12] and in the liver of fish in the Eastern English Channel [11] and in Kolleru lake [5]. The Cd content in gills and the liver of roach and perch was similar. The only statistically significant difference was observed in roach gills where the Cd level was considerably lower (0.061 $\mu g\ g^{-1}$) than in bream and perch (Tab. 2).

The average lead concentration in the muscle tissue of bream amounted to 0.41 $\mu g\ g^{-1}$ for specimens in the weight group of up to 200 g, whereas for specimens of the same species in the weight group of 1800–2000 g it was smaller and amounted to 0.24 $\mu g\ g^{-1}$ (Tab. 1).

The average concentration of that metal amounted to 0.29 μg g^{-1} in the muscles of studied bream, while in gills it was 1.29 μg g^{-1} and was situated in the range 0.68–2.06 μg g^{-1}; in the liver it amounted to 1.74 μg g^{-1} and was in the range 0.78–3.03 μg g^{-1} (Tab. 2). Differences between those averages were statistically significant. The statistically important difference was also observed between the Pb concentration in gills of bream and gills of roach and perch. The average content was 0.88 μg g^{-1}. The lead concentration in the liver of studied fish was similar. The highest level was observed in the liver of bream (1.74 μg g^{-1}) and the lowest in the liver of roach (1.45 μg g^{-1}). The lead content increased according to the growth of the fish mass for both gills and the liver, just opposite to muscles. Exceptionally for lead, the above relationship was curvilinear in both muscles and gills (Tab. 3). The relationship was rectilinear in the liver, with the highest correlation coefficient obtained in our research.

Table 3. The relationship between the contents of heavy metals in muscles, gills and liver and the weight of bream (r – correlation coefficient, p = 0.001)

Metals	Organs	Equation of regression	r
Cu	Muscles	$y = 2.33\,x^{-0.263}$	-0.895
	Gills	$y = -1.21 \cdot 10^{-3}x + 4.69$	-0.947
	Liver	$y = -2.61 \cdot 10^{-3}x + 8.60$	-0.917
Cd	Muscles	$y = 4.44 \cdot 10^{-3}x^{0.309}$	0.841
	Gills	$y = 1.50 \cdot 10^{-5}x + 6.65 \cdot 10^{-2}$	0.830
	Liver	$y = 2.56 \cdot 10^{-5}x + 6.30 \cdot 10^{-2}$	0.732
Pb	Muscles	$y = 1.59x^{-0.252}$	-0.850
	Gills	$y = 0.0995x^{0.356}$	0.912
	Liver	$y = 0.985x + 0.614$	0.978
Mn	Muscles	$y = 2.61x^{-0.259}$	0.796
	Gills	$y = 1.28 \cdot 10^{-4}x + 0.998$	0.747
	Liver	$y = 3.14 \cdot 10^{-4}x + 1.678$	0.838
Zn	Muscles	$y = 96.8x^{-0.441}$	0.788
	Gills	$y = 6.70 \cdot 10^{-3}x + 14.42$	0.821
	Liver	$y = 4.91 \cdot 10^{-3}x + 8.07$	0.838

To compare, the lead content in muscles of bream from Szczecin bay amounted to 0.38 μg g^{-1} in 1984 [19], in muscles of bream from the Odra estuary in 1995 it amounted to the average of 0.47 μg g^{-1} [12], whereas the lead content was several times higher in muscles of bream from ponds of Barycza river and varied between 2.00 and 3.28 μg g^{-1} [16]. Farkas [9] found in muscles of studied breams mean content of that metal 0.42 μg g^{-1}. Perch and roach from Dzierżno Duże reservoir [13] had similar Pb levels in muscles (0.42 and 0.39 μg g^{-1} respectively) as fish from Lake Gardno. Fish from the southern Atlantic coast of Spain content mean 0.32 μg g^{-1} lead in muscles [28].

The average content of zinc was 4.47 μg g^{-1} in the muscle tissue of bream, 14.39 μg g^{-1} in gills and 23.55 μg g^{-1} in the liver. Differences between those averages were statistically significant. No statistically important differences in zinc levels in muscles among the species of studied fish were observed. Roach muscles had the highest zinc content (5.84 μg g^{-1}) and perch muscles – the lowest (3.98 μg g^{-1}). A similar relationship was observed between roach and perch in Dzierżno Duże reservoir [13]; however the given concentrations were two or three times higher. The zinc level in bream muscles decreased from the average of 7.65 μg g^{-1} for specimens in the 200–400 g weight group to 3.19 μg g^{-1} for specimens in the 1600–1800 g of the same species, according to the equation of curvilinear regression, given in the Table 3. In gills and the liver an increase in the content of that metal with fish weigh was observed. It was described by equations of rectilinear regression and high correlation coefficients. In gills and the liver the lowest content of zinc (8.79 and 14.87 μg g^{-1} respectively) was observed in the weight group of 400–600 g and the highest one (20.35 and 28.60 μg g^{-1} respectively) in the weight group of 1600–1800 g. The content of zinc in gills of roach and perch was smaller but the differences were not statistically significant, similar to differences between concentrations of that metal in the liver (Tab. 2). Livers of bream and roach contained the same quantity of zinc and the liver of perch – smaller than 17.31 μg g^{-1}.

The content of zinc in muscles and the liver in studied bream was comparable to the concentration of that metal in muscles of bream caught in ponds of the Barycza river (3.00 to 4.48 μg g^{-1}) and in the liver (14.03 to 22.98 μg g^{-1}) [16]. The similar range of Zn concentrations was observed in muscles of bream from Szczecin bay in 1984 (2.95–5.75 μg g^{-1}, [19]) and from the Vistula River near Kraków (4.00–8.70 μg g^{-1}, [14]), whereas Hłyńczak et al. [12] noticed a little higher values for the same species of fish from the Odra estuary (the average content in 1995 in muscles was 7.13 μg g^{-1}, in the liver – 26.85 μg g^{-1}). Falandysz [7] recorded the zinc content of 7.7 and 19.00 μg g^{-1} in muscles and in the liver of studied perch respectively, thus it was higher than in the liver of the same species of fish of Lake Gardno. Kremer et al. [15] found high level of zinc in perch muscles (9.71 μg g^{-1}) and in the liver (28.68 μg g^{-1}).

The average content of manganese in the muscle tissue of studied bream was 0.21 μg g^{-1} (Tab. 2), whereas the average Mn content was 0.75 μg g^{-1} in gills and 1.20 μg g^{-1} in the liver. Differences among concentrations of manganese in muscles, gills and the liver were statistically significant. The level of manganese in bream muscles varied between 0.04 and 0.41 μg g^{-1}, in gills it ranged from 0.15 to 1.20 μg g^{-1}, whereas in the liver it ranged from 0.56 to 2.14 μg g^{-1}. Similar concentrations were observed in muscles, gills and the liver of roach and perch. The only major difference was observed in the content of manganese in the roach liver, where its level was much lower than in bream and perch.

A decrease in the manganese content with weight was observed in muscles, similar to previously described metals. The most rapid decrease was observed between groups of up to 200 g weight and 800–1000 g. The manganese content in gills of studied bream was similar in particular weight groups, with a small increase with weight, whereas in the liver, the level of manganese decreased with weight from 0.61 μg g^{-1} (weight group of up to 200 g) to 1.87 μg g^{-1} (weight group 1000–1200 g). The relationship between the level of that metal in muscles of studied fishes and their weight was curvilinear in gills and rectilinear in the liver (Tab. 3). Correlation coefficients were high and statistically significant.

The content of manganese in muscles of studied bream from Gardno lake was higher than in bream from the Vistula River near Kraków and bream from Fish Storehouse in

Kraków (0.109–0.146 μg g^{-1}, [14]) but it was similar to bream from the Baltic sea (0.27 μg g^{-1}, [6]). The similar concentration was observed for other fish species: pike – 0.23 μg g^{-1}, tench – 0.27 μg g^{-1}, carp – 0.13 μg g^{-1}, perch – 0.18 μg g^{-1}, roach – 0.29 μg g^{-1} [13], perch – 0.31 μg g^{-1} in muscles and 3.9 μg g^{-1} in the liver [7].

The iron content increased together with the weight of studied fish, in muscles, gills and the liver as well (Tab. 1). The average concentration of that metal was 5.35 μg g^{-1} in muscles, 38.84 μg g^{-1} in gills, and 57.38 μg g^{-1} in the liver of bream (Tab. 2). Differences between those averages were statistically highly significant. The lowest iron level in muscles of bream (1.88 μg g^{-1}) was observed in the weight group of 200–400 g and the highest (8.82 μg g^{-1}) in the group of 1600–1800 g. The lowest level of iron (10.10 μg g^{-1}) was in the group of up to 200 g and the highest (64.40 μg g^{-1}) in the group of 1800–2000 g. This group also had the highest content of that element in the liver (79.40 μg g^{-1}). The iron content in muscles, gills and the liver of roach and perch from Lake Gardno was similar to studied bream.

The levels of iron in muscles of studied fish were similar to the levels determined in other studies. The iron content in muscles in fish examined by Kostecki [13] varied between 5.33 and 9.58 μg g^{-1}. In cod examined by Falandysz and Ośmiałowski [8] it varied between 3.1 and 9.8 μg g^{-1}. Kretowska-Kulas [14] gives values of 5.84 μg g^{-1} in muscles of bream and 6.02 μg g^{-1} for roach. Falandysz [7] recorded the iron content of 2.9 μg g^{-1} in muscles of perch and 57.0 μg g^{-1} in the liver.

Iron and zinc were dominant metals in all tissues of studied fish. The content of studied metals accumulated in bream was as follows: Cd < Mn < Pb < Cu < Zn < Fe. That record is in principle consistent with results published by other authors and concerning the content of trace elements in fish organs [3, 4, 9, 12, 13]. Small differences concerning the quantity of accumulated metals in tissues of bream, perch and roach were recorded. Marek [16] noted similar relationships, however no relationship between accumulation of studied metals and sex of studied fishes was observed, similarly to Al-Yousuf et al. [1].

The content of studied metals in muscles of examined fish from Lake Gardno did not exceed permissible values, given by regulations of the Minister for Health [23], so they do not pose a threat for consumers. Taking into consideration the fact that fish in Lake Gardno are caught for potential consumers, studies of this kind should be carried out from time to time in order not to allow to trade with fish polluted with metals.

Results presented in this paper show that concentration of copper, lead, manganese and zinc in muscles of studied breams decreased with their age and more exactly with their mass. It shows that fish probably develop a mechanism removing heavy metals from their muscles. Therefore, we can say that given concentrations in muscles of studied fish are natural. Falandysz [7] and Farkas [9] gave a similar conclusion basing on their research. That means that Lake Gardno is not polluted with those metals or polluted to a small degree. They examined the content of heavy metals in water and bottom sediments of that lake. However, the cadmium concentration in muscles of bream increased slightly with weight. It does not mean yet that heavy metals present in that lake are of natural origin only. Values of enrichment factors deny this fact (Tab. 4). Indirectly they show the origin of metals accumulated by organisms. Obtained factors are normalized in relation to iron and related to bottom sediments, for which some necessary values are taken from Trojanowski (unpublished data). Enrichment factors calculated that way do not include specific enrichment of organism with a given metal, which is caused by physiological parameters or efficiency of detoxication of organisms from different metals. It is assumed that enrichment factors higher than 10 shows an anthropogenic origin of metals, whereas EF values lower than 3 show the natural origin of metals [24]. Referring to the above, manganese and lead are the only metals of natural origin in fishes in Gardno Lake, whereas zinc and copper are partially of anthropogenic origin. Particularly when we consider zinc, enrichment factors in muscles are very close to 10 and the same is the case for copper in the liver. That shows that the majority of metals originate from unnatural sources. The origin of cadmium in gills and the liver of studied fish is natural and may be also thought of as natural in muscles because the values obtained from enrichment factors are very close to the value of 3 (Tab. 4).

Table 4. Enrichment coefficients of heavy metals in muscles (A), gills (B) and liver (C) of studied fish in Lake Gardno

Metals	Bream			Roach			Perch		
	A	B	C	A	B	C	A	B	C
Cu	5.52	6.03	7.02	5.52	6.14	8.09	5.91	6.09	8.69
Cd	4.90	1.30	1.24	3.12	2.58	2.46	4.11	2.76	1.23
Pb	1.27	2.09	0.71	1.67	1.06	0.74	1.41	0.92	1.05
Mn	0.10	0.05	0.06	0.03	0.06	0.08	0.11	0.05	0.07
Pb	8.43	3.74	4.14	9.34	4.88	6.54	7.29	4.27	3.54

The greatest changes in the concentration of heavy metals in muscles were observed for small specimens of studied fish (up to 600 g). The higher the weight the smaller changes were observed. Only cadmium and iron reacted in a different way. The largest increase in cadmium level was observed for specimens with weight above 1200 g. Iron is a metal needed by organisms on a large scale. Therefore, the content of iron in bream muscles systematically increased, proportionally to their mass.

The concentration of studied metals was several times higher in the liver of examined fishes. It results from the fact that the liver has a function of a filter in organisms and heavy metals removed from muscles are accumulated in the liver. Much higher concentrations of metals in the liver than in muscles confirms a large usability of that organ for such purposes as monitoring of metal pollution in aquatic environments [10, 22]. Some differences in the concentration of those elements in the liver of bream, roach and perch probably result from more or less selective absorption of heavy metals, different degree of their regulation in tissues, the way the fish is supplied with food and the trophic level. Feeding on detritus and in sediments [4, 12] results in more intensive accumulation. Therefore the liver of bream contained more studied metals than the liver of roach and perch because bream feed on sedimental fauna.

For comprehensive description of accumulation of studied metals in some fish an analysis of the coexistence of those elements was carried out. Table 5 contains correlation coefficients between the content of particular pairs of studied metals in muscles, gills and livers. The positive essential correlation in muscles with $p = 0.001$ was observed for following pairs of metals: Cu-Pb, Cu-Mn, Cu-Zn, Pb-Mn, Pb-Zn, in gills: Cd-Pb, Cd-Zn, Cd-Fe, Pb-Zn, Pb-Fe, whereas in livers: Cd-Pb, Cd-Zn, Cd-Fe, Pb-Fe, Zn-Fe. Similar relationships in muscles

Table 5. Correlation coefficients between concentration of respective metals in organs of studied fish ($p = 0.001$)

Metals	Muscles	Gills	Liver	Metals	Muscles	Gills	Liver
Cu/Cd	-0.732	-0.866	-0.860	Cd/Fe	0.460	0.652	0.612
Cu/Pb	0.906	-0.816	-0.373	Pb/Mn	0.647	0.044	0.419
Cu/Mn	0.689	-0.133	-0.286	Pb/Zn	0.603	0.844	-0.538
Cu/Zn	0.723	-0.802	-0.831	Pb/Fe	-0.820	0.777	0.625
Cu/Fe	-0.872	-0.719	-0.870	Mn/Zn	0.462	0.050	0.296
Cd/Pb	-0.587	0.744	0.711	Mn/Fe	-0.341	0.073	0.193
Cd/Mn	-0.431	0.314	0.161	Zn/Fe	-0.706	0.637	0.494
Cd/Zn	-0.322	0.740	0.842				

and the liver were shown by Falandysz [7]. The statistical analysis does not give the reason for univocal opinion on interactions in which those metals might participate in fish, but it may suggest that during accumulation one metal may be accompanied by others under favorable circumstances.

Table 6. Correlation coefficients (r) between the content of a metal in gills (Mc_s) and its content in muscles (Mc_m) and in liver (Mc_w)

	Me_m	Me_w
Cu_s	0.875**	0.925**
Cd_s	0.437*	0.932**
Pb_s	-0.702**	0.860**
Mn_s	-0.003*	-0.377*
Zn_s	-0.720**	0.309*
Fe_s	0.598**	0.629**

**p = 0.001, *p = 0.05

Fish gills are the organ through which water constantly passes, together with metals that are contained in it. They are just one of the organs, through which metals can penetrate into fish organism. Because of this, the relationship between the concentration of studied metals in gills and their content in muscles and the liver was examined. Table 6 shows calculated correlation coefficients. A positive, statistically significant correlation on the significance level of 0.001 for muscles was received for copper and iron and on significance level of 0.05 for cadmium. That means that accumulation of those metals in muscles is probably related to their content in gills. A similar situation was determined for copper, cadmium, lead and iron in the liver; that means that the presence of those metals in gills has a real effect on their contents in the liver.

CONCLUSIONS

The results presented in this paper show that the quantities of accumulated metals (Cu, Pb, Cd, Zn, Mn, Fe) in different fish tissues vary distinctly, irrespective of the species. Among compared tissues, the smallest quantities of metals were determined in muscles, much larger in gills, and the largest in the liver. Their content in the liver was several to about a dosen times higher than in muscles. Zinc was the most abundant metal in all tissues, irrespective of fish species. Cadmium was present in the lowest concentrations.

The effect of fish weight on the content of metals in studied tissues is very noticeable, particularly in younger fish of a small weight. At that time fish absorb much food together with metals contained in it. The content of manganese, copper, zinc and lead in muscles of studied bream decreased with the increase in weight. It is possible that older fish ensue change of diet and start to produce a mechanism that removes metals from muscles. On the other hand, iron and cadmium content increased with increasing weight. Iron is very important for the metabolism in fish. Therefore its content increases systematically in fish organisms, together with the increase of their weight. Cadmium is a toxic element but it probably binds more durably in muscles than other studied metals and is difficult to remove.

Among studied metals, manganese, cadmium and lead in Lake Gardno are of natural origin and zinc and copper are also of partially antropogenic origin.

Some irregular differences in the content of studied metals in muscles, gills and the liver between bream, roach and perch are probably a result of different metal selection and accumulation capabilities, different nourishment way and different trophic levels.

It was determined that during copper accumulation in muscles, lead, manganese and zinc were able to co-accumulate with copper, whereas zinc and manganese co-accumulated with lead and copper. In gills, cadmium is able to co-accumulate with lead, zinc and iron, and lead co-accumulates with zinc, iron and cadmium. In the liver, similarly as in gills, cadmium co-accumulates with lead, zinc and iron; iron co-accumulates with zinc, and lead and cadmium are able to accumulate together.

The contents of copper, iron and cadmium in gills affects considerably the accumulation level of those metals in muscles. The more of those elements are present in gills, the higher concentration of them can be expected in muscles. A similar relationship has been determined between gills and the liver for such metals as copper, cadmium, lead and iron.

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