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CONTENT OF SELECTED ESSENTIAL AND TOXIC METALS IN MEAT OF FRESHWATER FISH FROM WEST POMERANIA, POLAND

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Key words: macroelements, microelements, heavy metals, freshwater fish, diet

The aim of this study was to assess concentrations of essential and toxic metals in fish meat from north-west Poland. Selected metals (Al, Fe, Mn, Zn, Cu, Pb, Cd, Hg, Ni, Mg, Cr, V, Li, K, Ca, Na) were determined in the muscle tissue of freshwater fish species: pike, bream, perch and common carp, harvested from natural waters of West Pomerania, Poland. Levels of Cd and Pb were determined by Graphite Furnace Atomic Absorption Spectrometry (GF-AAS), Hg by the cold vapour technique after Hg ions reduction with $SnCl_2$ (CV-AAS), and the other metals by Inductively Coupled Plasma – Atomic Emission Spectrometry (ICP-AES). The accuracy and precision of methods applied were examined with certified reference materials: Fish-paste 2 and Dolt-2. The results of the research were processed using statistical methods (ANOVA); the Duncan's test was performed at a significance level of $p \leq 0.05$. The data indicate that the examined fish were not polluted with metals, and the mean concentrations of toxic metals (Pb, Cd, Hg) were within the limits for fish and fishery products specified by EU legislation, so the fish were safe for human consumption.

INTRODUCTION

Fish are located at the end of the aquatic food chain, and many accumulate metals and pass them to human beings through food. This way they enrich human diet in essential elements, but also cause chronic or acute diseases when deliver metals in excess [Eisler, 1981]. Human demand for minerals depends on age (which is related to body mass), physiological condition (pregnancy, lactation) and, in the case of Mg, Fe and Zn, also on gender. Fish meat contains more P, K and Mg, comparing to meat from livestock, and in the case of pickled fish or fish with edible bones (esp. *Clupeiformes*) – also more Ca. Accumulation of metals in fish depends on concentrations of metals in water and food organisms, on physiochemical factors, and exposure duration. Toxic effect of metals depends on the site of their deposition within the body [Witeska & Jezierska, 2001]. Trace element levels are known to vary in fish depending on various factors, such as habitat, feeding behavior and migration even in the same area [Roméo *et al.*, 1999; Canli & Atli, 2003]. Having entered animal body, metals are not evenly distributed, but accumulate in particular organs. In fish, metals accumulate mainly in kidney, liver, intestine epithelium, and in other organs in much smaller amounts. [Protasowicki, 1987]. Differences in metal concentrations in various tissues may result from their different capability to induce metal-binding proteins such as metallothioneins [Tuzen & Soylak, 2007]. Fish muscles, comparing to the other tissues, usually contain low levels of metals but are often examined for metal content due to their use for human consumption [Witeska & Jezierska, 2001].

In Poland, a majority of freshwater fish for consumption offered at local markets are harvested from local lakes and rivers. The aim of this study was to determine selected essential and toxic metals in the muscles of four fish species commercially harvested in the West Pomeranian region. Pike, bream and perch were obtained from commercial catches in randomly chosen lakes. Most of the lakes have basins used for agricultural or forestry purposes, and the lakes are not affected by industrial wastes. Potential sources of their pollution are surface run-off, and – as in the case of lakes Wisola, Mętno, Miedwie, also tributaries bringing in pollutants from a wastewater treatment plant, garbage dump site or from discharge of untreated wastewater. Carp fish were collected from a warm channel receiving discharges of cooling water (previously taken from the Oder River) from the "Dolna Odra" Power Station.

MATERIALS AND METHODS

The concentration of metals was determined in muscle tissue of four fish species, predatory fish: pike (*Esox lucius* L.), typical benthophagous: bream (*Abramis brama* L.), common carp (*Cyprinus carpio* L.), and benthophagous with substantial portion of fish in food: perch (*Perca fluviatilis* L.). The fish were obtained from commercial catchments from natural waters of West Pomerania, Poland (Figure 1). A total of 496 fish were caught and transported in cooler bags to the laboratory, where samples of dorsal muscles were dissected, placed in polyethylene bags and frozen at -20°C. For Hg determina-

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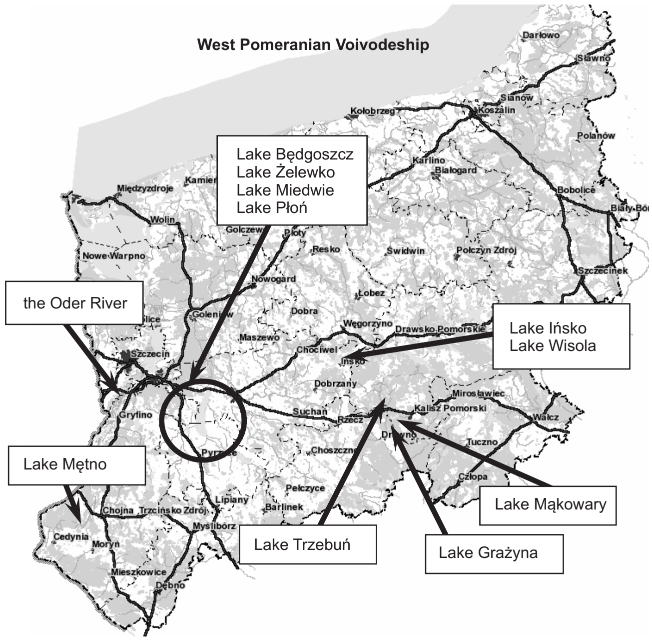

FIGURE 1. Localization of the study area.

tion, subsamples of 5 g (±0.001 g) wet weight (w.w.) were digested in a mixture of concentrated HNO_3 and $HClO_4$ (v/v, 4:1) [Protasowicki, 1985]. For the other metal analyses (Ca, Mg, Na, K, Fe, Mn, Zn, Cu, Ni, Cr, V, Li, Pb, Cd), subsamples of 2 g (±0.001 g) w.w. were digested 3 mL HNO_3 (65%) in Teflon bombs in a microwave oven (CEM MDS 2000). Mercury content was analysed by the cold vapour atomic absorption technique after Hg ions reduction with $SnCl_2$ (CV-AAS). Cadmium and lead were determined by graphite furnace atomic absorption spectrometry (GF-AAS), and the other metals by inductively coupled plasma – atomic emission spectrometry (ICP-AES). All analytical samples and blanks were prepared in triplicate. Accuracy and precision of methods applied were checked with certified reference materials (Fish-paste 2 and Dolt 2). The results obtained were subjected to analysis of variance (ANOVA, Duncan's test). In all cases statistical significance was estimated at $p \leq 0.05$ [Babiak, 1998]. The results were also compared with tables of food composition and nutritional value [Kunachowicz *et al.*, 2005] and nutritional recommendations for population in Poland [Ziemlański, 2001].

RESULTS AND DISCUSSION

Macroelements

Ca content in carp muscles averaged 384 mg/kg w.w. (Table 1). Falandysz [1994] reported Ca content in turbot (*Psetta maxima*) to range within 87-260 mg/kg w.w. Lower muscle content of Ca was reported in roach, bream and perch from lakes of the Olsztyn Lake District [Łuczyńska *et al.,* 2006]. Ca values of 463-854 mg/kg were reported by Orban *et al*. [2007]. According to Kunachowicz *et al.* [2005], a 100-g edible portion of carp tissue contains 10 mg of Ca, and only about 30-40% of dietary Ca is assumed to be assimilated [Ziemlański, 2001]. In our study, Ca content averaged 38 mg/100 g edible portion. The recommended daily allowance (RDA) of Ca for women and men aged 25 years and younger is 1200 mg per day and for women over 60 years of age 1100 mg per day (due to progressing loss in bone mineral mass) [Ziemlański, 2001]. From the above it appears that a 100-g edible portion of the examined carp meat would satisfy *ca.* 3.3 percent of human daily demand for Ca.

TABLE 1. Content of essential macroelements in the muscle tissue of common carp (*Cyprinus carpio* L.) from the Oder River, Poland.

Element	Concentration (mg/kg w.w.)	
	$\bar{X}$	±SD
Ca	384	402
Mg	252	100
Na	367	112
K	1354	629

$\bar{X}$±SD – mean ± standard deviation, n=70.

According to Kunachowicz *et al.* [2005], fish tissue contains 12 to 49 mg of Mg per 100-g edible portion. In our study, Mg content in carp muscles averaged 252 mg/kg w.w. (Table 1), which is equivalent to 25 mg/100 g. This corresponded to Mn levels in European perch (218-271 mg/kg)reported by Orban *et al*. [2007]. It is widely assumed that *ca.* 50 percent of dietary Mg is absorbed by humans. The recommended daily allowance of Mg for girls and adult women ranges within 300-380 mg per person, while for boys and adult men within 290-400 mg per person [Ziemlański, 2001]. Therefore, a 100-g portion of the examined carp meat would satisfy *ca.* 7 percent of human daily demand for Mg.

Carp muscle Na content determined in this study averaged 367 mg/kg w.w. (Table 1) and was similar to levels of the mineral reported by Adeyeye *et al.* [1996] in the muscle meat of Nile tilapia (*Oreochromis niloticus*) and by Łuczyńska *et al*. [2006] in fish from north-east of Poland. Falandysz [1994] determined Na content of turbot (*Psetta maxima*) to be 920 mg/kg. According to Kunachowicz *et al.* [2005], carp flesh contains 30 mg of Na per 100 g edible portion. In the examined carp meat, Na content averaged 37 mg/100 g, which corresponded to *ca.* 7% of human daily demand for Na, as the RDA for women and men ranges within 500-625 mg per person.

According to Kunachowicz *et al.* [2005], a 100-g edible portion of carp contains 387 mg of K. In this study, muscle K levels were lower and averaged 135.4 mg/100 g (Table 1), however they were significantly higher than those reported by Adayeye *et al.* [1996]. Falandysz [1994], Erkan & Özden [2006] found K contents to be higher (2993-4597 mg/kg). The recommended daily allowance of K for women and men ranges within 2000-3500 mg per person [Ziemlański, 2001]. Thus, it can be assumed that a 100-g portion of the examined fish would satisfy nearly 5 percent of human daily demand for K.

Essential microelements

Fish are considered to be some of the major sources of Fe in human diet. Fe content in the muscles of the examined fish varied within 0.6 in pike from Mętno lake to 3.2 mg/kg w.w. in carp from The Oder River (Table 2). For comparison, Alibabić &Wahèić [2007] found Fe concentrations of 5.2 mg/kg w.w. in the muscles of fish caught in the Una River basin. Türkmen & Ciminli [2007] reported 1.48 mg Fe/kg w.w. in fish from Lake Gölbaşi. In Nasser lake in Egypt, fish muscles contained 2.18 mg Fe/kg w.w. [Rashed, 2001]. Much higher concentration of Fe was observed in Mediterranean fish (16.6-78.4 mg/kg w.w.) [Canli & Atli, 2003]. On average, only 10 to 15 percent of Fe is absorbed from the diet, the absorption efficiency being dependant on food composition and an organism's demand. In the examined fish, Fe levels ranged within 0.1-0.3 mg per 100 g of muscle meat (Table 2) and were lower than those reported by Kunachowicz *et al.* [2005] for edible portions of fish (0.2-1.4 mg/100 g). The recommended daily allowance of Fe for girls and adult women ranges within 13-26 mg per person, while for boys and adult men – within 14-15 mg per person [Ziemlański, 2001], hence consumption of a 100-g portion of the examined fish would satisfy merely about 1.5 percent of human daily demand for Fe.

In our study, Mn content in fish meat ranged from 0.10 to 0.39 mg/kg w.w. (Table 2) and the lowest mean concentration was observed in pike. Łuczyńska *et al.* [2006] found Mn concentrations of 0.071-0.117 mg/kg in fish from northeast Poland. Higher Mn levels were reported by Alibabić & Wahèić (0.62-0.91 mg/kg w.w.) [2007], and Jurkiewicz-Karnakowska [2001] observed ten times higher levels than found in our study. Fish meat is assumed to contain 0.01-0.06 mg Mn per 100 g edible portion [Kunachowicz *et al.*, 2005], comparing to *ca.* 0.02 mg per 100 g observed in this study.

Zn content in the muscles of the examined fish species ranged within 2.8-6.8 mg/kg w.w. (Table 2). Muscle levels of Zn were significantly higher ($p \leq 0.005$) in pike and perch than in bream and carp. Zn concentrations observed by Szefer *et al*. [2003] in the Pomeranian Bay and Szczecin Lagoon and by Protasowicki [1991] are in agreement with those found in this study. For comparison, slightly higher Zn levels were reported for fish from the Dnieper River [Sapozhnikova, 2005]. Much higher concentrations of this metal were observed by other authors [Kuznetsova *et al.*, 2002; Liang *et al.,* 1998; Farkas *et al.*, 2002]. However, Türkmen & Ciminli [2007] reported lower Zn concentration (0.456 mg/kg w.w.) in fish from Lake Gölbaşi. On average, 20 to 40 percent of dietary Zn is absorbed in healthy people [Rao, 1980]. According to Kunachowicz *et al.* [2005], a 100-g edible portion of fish contains from 0.30 to 1.75 mg of Zn. In our study, Zn levels ranged from 0.28 to 0.68 mg/100 g edible portion. The recommended daily allowance of Zn for girls and adult women ranges within 13-21 mg per person, while for boys and adult men it is 16 mg per person [Ziemlański, 2001]. Assuming an average RDA for Zn to be 19 mg per person, a 100-g edible portion of the examined fish would satisfy *ca.* 2.6 percent of human daily demand for this mineral.

Mean muscle concentration of Cu in the examined fish ranged from 0.08 to 0.17 mg/kg w.w. (Table 2). Al-Yousuf *et al.* [2000] reported similar Cu levels in muscles of fish from the Arabian Gulf along the western coast of the United Arab Emirates. Higher Cu concentrations (0.2-0.4 mg/kg w.w) were reported by Szefer *et.al*. [2003] and also by Karadede & Ûnlü [2000] in carp (2.23 mg/kg w.w.). Cu assimilability ranges within 35-70 percent, being lower in adults and higher in young people. According to the tables of food composition and nutritional value [Kunachowicz *et al.*, 2005], a 100-g edible portion of fish contains from 0.02 to 0.23 mg of Cu. Copper levels in the fish examined in our study varied from 0.01 to 0.02 mg/100 g edible portion. The recommended daily allowance of Cu for both women and men varies within 1.5-2.5 mg per person [Ziemlański, 2001]. Therefore, 100 g of fish meat would satisfy *ca.* 1 percent of human daily demand for this mineral.

Dietary Cr usually satisfies human demand for the element, which varies from 50 to 200 μg per day in adult people. Muscles of the examined fish contained from 0.02 mg Cr/kg w.w. in perch from Lake Miedwie to 0.15 mg Cr/kg w.w. in perch from Lake Płoń (Table 2), which corresponded to 5.5 μg Cr per 100-g edible portion of the examined fish. Sapozhnikowa *et al.* [2005] and Rashed [2001] reported similar chromium levels in fish, and Ikem *et al.* [2003] observed 0.01 mg Cr per kg w.w. in fish muscle. Much higher Cr concentrations ab. to 1.08 mg/kg, were observed in fish from the Una River basin, by Alibabić & Vahèić [2007].

Average V content in fish meat examined in this study ranged within 0.02-0.36 mg/kg w.w. (Table 2), the most V being found in perch. For comparison, Ikem *et al.* [2003] observed no V in fish from Tuskegee Lake, while Sapozhnikowa *et al.* [2005] reported V levels in fish exceeding 0.4 mg/kg which is similar to levels observed in perch from lakes Miedwie and Płoń. Türkmen & Ciminli [2007] observed V to be 0.013 mg/kg w.w.in fish (*Clarias gariepinus*) from Lake Gölbaşi – concentration similar to found fish from lakes Grażyna and Trzebuń.

Levels of the Li in the examined fish averaged from 0.01 mg/kg w.w. for carp to 0.13 mg/kg w.w. for perch (Table 2). We found no information on Li content in fish meat in the available literature.

In our study, Ni levels in fish muscles averaged from 0.03 mg/kg w.w. in pike to 0.13 mg/kg w.w. in perch (Table 2). Similar levels of this metal in fish muscles were observed by Liang *et al.* [1999]. Türkmen & Ciminli [2007] observed 0.009 mg/kg w.w. in fish from Lake Gölbaşi, however Mendli & Uluözlü [2007] reported higher levels of nearly 5.5 mg/kg in fish from lakes in Tokat. Human demand for this mineral is assumed within 25-35 μg per day [Anke *et al.*, 1992]. Hence, a 100-g edible portion of the examined fish would satisfy *ca.* 3.6 percent of human daily demand for Ni.

Considering the above mentioned information on metal levels in fish meat and human demand for nutrients we assume that fish are a significant source of essential macro- and microelements for people in Poland, even regarding relatively low proportion of fish meat in the average Polish diet (on average 5.6 kg per year). On the other hand, it is necessary to determine toxic metal levels in fish, as maximum limits for such metals in foodstuffs are set by legislation in Poland, EU and other countries in order to protect consumer's health.

Non-essential microelements

Pb content in the examined fish meat ranged within 0.01-0.02 mg/kg w.w. Similar levels were observed by Szefer *et al.* [2003] in perch from the Pomeranian Bay and Szczecin

TABLE 2. Content of essential microelements and toxic metals in the muscle tissue of pike (*Esox lucius* L.), bream (*Abramis brama* L.), perch (*Perca fluviatilis* L.) and common carp (*Cyprinus carpio* L.) from commercial catches in randomly chosen lakes and rivers of West Pomerania, Poland.

Fish species Lake/River	n	Length (cm)		Weight (g)		Metal concentration (mg/kg w.w.)																					
						Fe		Mn		Zn		Cu		Cr		V		Li		Ni		Pb		Cd		Hg	
		$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD	$\bar{X}$	SD
PIKE	236					0.9		0.19		6.8		0.13		0.04		0.02		0.05		0.03		0.02		0.003		0.09	
L. Ińsko	64	46.8	3.2	683	135	1.4	1.8	0.24	0.12	9.4	5.2	0.14	0.06	0.06	0.03	0.01	0.03	0.05	0.05	0.01	0.03	0.02	0.02	0.003	0.004	0.01	0.00
L. Wisola	52	46.5	4.5	665	224	0.8	1.2	0.22	0.19	6.2	4.4	0.19	0.16	0.04	0.02	0.05	0.03	0.02	0.01	0.04	0.03	0.02	0.01	0.003	0.003	0.03	0.06
L. Mętno	60	52.7	5.6	1119	394	0.6	0.5	0.20	0.10	6.9	3.0	0.09	0.03	0.03	0.01	0.00	0.00	0.05	0.04	0.01	0.03	0.02	0.01	0.002	0.001	0.19	0.05
L. Będgoszcz	60	50.1	7.5	978	543	0.8	1.1	0.10	0.10	4.6	2.2	0.11	0.04	0.04	0.03	0.00	0.00	0.07	0.06	0.07	0.06	0.02	0.01	0.002	0.001	0.13	0.03
BREAM	140					1.4		0.23		2.8		0.17		0.05		0.02		0.04		0.05		0.01		0.002		0.01	
L. Miedwie	10	43.2	2.7	1044	181	1.3	1.0	0.11	0.05	2.3	1.0	0.14	0.07	0.03	0.02	0.00	0.01	0.05	0.03	0.04	0.04	0.01	0.00	0.000	0.001	•	•
L. Płoń	10	43.2	3.6	990	160	1.4	0.9	0.27	0.40	2.5	1.5	0.11	0.05	0.05	0.04	0.01	0.01	0.05	0.03	0.01	0.02	0.01	0.01	0.003	0.003	•	•
L. Żelewko	10	29.8	3.3	330	112	1.4	0.6	0.18	0.04	3.0	0.8	0.18	0.08	0.04	0.02	0.00	0.00	0.05	0.02	0.00	0.00	0.00	0.00	0.001	0.000	•	•
L. Wisola	48	37.7	2.7	599	137	1.3	1.1	0.23	0.17	3.0	1.6	0.24	0.15	0.02	0.02	0.03	0.01	0.01	0.02	0.06	0.07	0.02	0.01	0.002	0.003	0.01	0.01
L. Ińsko	62	39.4	2.7	708	160	1.5	1.8	0.39	0.43	3.2	1.9	0.18	0.15	0.11	0.06	0.06	0.05	0.05	0.03	0.13	0.06	0.01	0.01	0.002	0.003	0.01	0.00
PERCH	50					1.7		0.22		5.2		0.16		0.06		0.36		0.13		0.13		0.01		0.003		0.03	
L. Trzebuń	10	18.6	3.2	77	47	1.7	0.6	0.24	0.34	7.5	2.3	0.16	0.10	0.06	0.04	0.02	0.01	0.02	0.01	0.11	0.08	0.02	0.03	0.003	0.003	0.02	0.05
L. Mąkowary	10	19.1	1.7	88	28	1.7	0.6	0.25	0.48	6.1	2.0	0.20	0.06	0.03	0.03	0.01	0.01	0.02	0.01	0.08	0.10	0.03	0.01	0.006	0.003	0.06	0.04
L. Grażyna	10	18.9	4.6	102	83	1.6	0.7	0.22	0.45	4.8	1.4	0.18	0.05	0.03	0.03	0.01	0.02	0.02	0.01	0.08	0.04	0.01	0.02	0.004	0.006	0.05	0.14
L. Miedwie	10	24.8	1.2	201	24	1.2	1.2	0.18	0.04	4.6	1.2	0.14	0.12	0.02	0.06	1.17	0.52	0.21	0.03	0.11	0.04	0.00	0.01	0.001	0.001	0.01	0.01
L. Płoń	10	22.1	1.0	148	10	2.1	1.7	0.19	0.52	3.2	4.4	0.13	0.24	0.15	0.05	0.60	0.62	0.38	0.36	0.27	0.21	0.00	0.01	0.001	0.001	0.02	0.01
CARP	70					3.2		0.20		3.3		0.08		0.06		0.05		0.01		0.10		0.01		0.001		0.01	
Oder River	70	23.4	4.4	286	149	3.2	2.0	0.20	0.18	3.3	1.9	0.08	0.06	0.06	0.04	0.05	0.04	0.01	0.01	0.10	0.06	0.01	0.01	0.001	0.001	0.01	0.00

$\bar{X}$±SD – mean ± standard deviation, • – no data

Lagoon (Pomeranian Bay 0.007-0.033 mg/kg w.w. and in Szczecin Lagoon 0.013 mg/kg w.w.). Türkmen & Ciminli [2007] also found low Pb concentrations in fish from Lake Gölbaşi (*Clarias gariepinus* – 0.014 mg/kg w.w., and *Carasobarbus luteus* – 0.008 mg/kg w.w.). Some other authors reported values over 0.100 mg/kg w.w. [Has-Schön *et al.*, 2006]. Much higher concentration was reported by Jurkiewicz-Karnakowska [2001] in fish from Zegrzyński Reservoir (from 8.9 to 12.7 mg/kg w.w.).

Mean Cd content in the muscle tissue of the examined fish ranged within 0.001-0.003 mg/kg w.w., and was below the maximum allowable level of 0.050 mg/kg w.w. [O.J. L 364, 2006]. Similar situation was reported by Szefer *et al.* [2003] in fish from Szczecin Lagoon. Also Türkmen & Ciminli [2007] found Cd levels below 0.001 mg/kg w.w. in fish (*Clarias gariepinus*) from Lake Gölbaşi, whereas Dobicki & Polechoński [2003] observed ten times higher Cd level in pike from Wojnowskie lake (0.01 mg/kg w.w.). Sapoznikowa *et.al* [2005] observed 0.038 mg Cd/kg w.w. in roach from the Dniester River, while Liang *et al.* [1999] and Farkas *et al.* [2002] reported Cd levels in fish that exceeded the allowable maximum level of 0.050 mg/kg w.w.

Mercury levels in the muscles, depending on the examined fish species, varied between 0.01 and 0.19 mg/kg w.w. (Table 2), while the maximum allowable level is 1.0 mg/kg w.w. [O.J. L 364, 2006]. Similar mercury levels in West Pomeranian fish (on average 0.015-0.030 mg/kg w.w.) were reported by Szefer *et al.* [2003], and by Perkowska & Protasowicki in pike muscle from Świdwie lake [1999]. In contrast, higher Hg levels were found by Has-Schön *et al.* [2006] in muscles of Croatian fish and by Dušek *et.al* [2005] in fish from the Elbe River.

CONCLUSIONS

Apart from many other diet components, fish meat may be an important source of human diet supplementation in macroelements and microelements. In the case of the examined freshwater fish from the north west Poland this was especially true for macroelements such as: magnesium, sodium, potassium, calcium, and microelements such as: zinc, nickel and iron. Metal concentrations found in the muscle tissue of West Pomeranian fish were low – similar or lower than those reported for fish by other authors. Regarding low levels of toxic metals *i.e.*, lead, cadmium and mercury, the fish were found to be safe for consumers' health.

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