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BIOACCUMULATION OF ARSENIC, CHROME, MANGANESE, AND NICKEL IN THE GILLS OF SEA TROUT (*SALMO TRUTTA* M. *TRUTTA* L.) FROM THE SOUTHERN BALTIC SEA (CENTRAL POMERANIAN REGION)

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Abstract

This study aimed to assess the concentrations of arsenic (As), chrome (Cr), manganese (Mn), and nickel (Ni) in the gills of the sea trout (*Salmo trutta* m. *trutta* L.) collected in the Baltic Sea (northern region of Poland). The results were then compared to permissible limits to detect whether the metal contamination levels in sea trout from the Baltic sea exceed the values of limits admissible. Frequently consumed sea trout (25 samples) were purchased from local fisherman in January-February 2014, from Ustka (54°34'43"N 16°52'09"E), Pomeranian voivodship, Poland. The element contents in the gills were analyzed using an inductively coupled plasma mass spectrometry (ICP-MS) technique. Metal contents in fish samples were found 0.00313-0.02069 mg·kg^{-1} for As (the mean value was 0.01294±0.0009 mg·kg^{-1}), 0.0118-0.02161 mg·kg^{-1} for Cr (0.01449±0.0003 mg·kg^{-1}), 0.01895-0.14216 mg·kg^{-1} for Mn (0.04137±0.0046 mg·kg^{-1}), 0.00664-0.01528 mg·kg^{-1} for Ni (0.00811±0.00034 mg·kg^{-1}). According to these data, the ranking order of the mean concentration of the heavy metals in fish gills was Mn (0.04137 mg·kg^{-1}) > As (0.01294 mg·kg^{-1}) > Cr (0.01449 mg·kg^{-1}) > Ni (0.00811 mg·kg^{-1}). The mean concentration of arsenic in gill samples (0.01294±0.0009 mg·kg^{-1}) was much below the permissible limit of USFDA (1993b) and FAO/WHO (1976). The mean Cr content in the gill of sea trout samples was well within the toxic limit of USFDA (1993a). The samples had lower Cr concentration as compared to the limits of 0.200 set by FSANZ (2002) and 0.100 by EUROPA (2004). Our study reported that the accumulation of Mn was exceeding the maximum permissible limit (by 8.27-16.55-fold)

according to WHO/EPA standard (0.0025-0.005 mg·kg^{-1}) (FAO/WHO 1976). The proposed limit of Ni concentrations in marine fish species as recorded by FAO (1983) is about 10 µg/g and 0.5-0.6 µg/g according to WHO Guidelines for drinking water quality (1985). In general, it can be seen that the concentrations of Ni found in gills of the sea trout in this study are still considered as those of uncontaminated fish. In conclusion, the assessment of element contents in the organs of the sea trout appears to be a useful biomarker to evaluate the toxic effects of heavy metal pollution as well as for human consumption.

Key words: (*Salmo trutta* m. *trutta* L.), gills, Baltic Sea, arsenic (As), chrome (Cr), manganese (Mn), nickel (Ni)

INTRODUCTION

A diet high in seafood has been linked to beneficial outcomes for an increasing number of diseases and medical conditions. Seafood and nutrients derived from a seafood diet have been proven to prevent and protect against many of epidemic lifestyle diseases. Eventually, fish is the most important single source of high-quality protein contributing about 17% of animal protein and 6.7% of all protein consumed by the world population (FAO 2016). In addition to being a rich source of protein, fish provides high contents of essential fats, vitamins, and minerals (Larsen et al. 2011, Kaya and Turkoglu 2017).

As Larsen and colleagues (2011) mentioned, the health benefits of seafood consumption have primarily been associated with protective effects against cardiovascular diseases (CVD). The intake of seafood has also been associated with improved fetal and infant development, as well as several other diseases and medical conditions. The health-promoting effects have chiefly been attributed to the long-chain n-3 polyunsaturated fatty acids (n-3 PUFA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). In addition, the general fatty acid profile is considered favorable. On the other hand, recent and emerging research on seafood proteins and other seafood derived components suggest that these nutritional components contribute to the health effects (Larsen et al. 2011). Epidemiological studies have presented inconsistent evidence of the correlation between a fish-oriented dietary intake (FDI) and the risk of cognitive decline. Zeng and co-workers (2017) have confirmed that a higher intake of fish could be correlated with a reduced risk of dementia of Alzheimer type. Further research, especially prospective studies that specifically quantify FDI, will help find a more accurate assessment of the different levels of dietary intake (Zeng et al. 2017).

A cross-sectional consumer survey was carried out in 2004 with representative household samples from Belgium, the Netherlands, Denmark, Poland, and Spain (Pieniak et al. 2010). The sample consisted of 4,786 respondents, aged 18-84 years, who were responsible for food purchasing and cooking in the household. European consumers had a very strong belief that eating fish is healthy. Consumers' belief that eating fish is healthy, their interest in healthy eating and objective fish-related nutrition knowledge, positively, but only weakly, influenced fish consumption frequency. Subjective knowledge was found to be a stronger predictor of fish consumption than the previously noted factors. Age and education contributed, both directly and indi-

rectly through knowledge, to explain fish consumption behavior. However, the path coefficients in the estimated model were relatively low, which indicates that fish consumption frequency was also determined by factors other than health-related beliefs and consumers' knowledge (Pieniak et al. 2010).

As fish is an important constituent of the human diet, it is often deemed as the most suitable object among the bioindicators of the aquatic ecosystem (Katagi 2010, Mijošek et al. 2019, Vieira et al. 2019). The intense development of anthropogenic activities since the late 19th century has resulted in enhanced loads of pollutants (e.g., nutrients, metals) from a large densely populated catchment area to the Baltic Sea (Remeikaitė-Nikienė et al. 2018). The significant input of cadmium, lead, and mercury via atmospheric deposition was also reported by HELCOM (2010). For instance, 47.5 tonnes of cadmium and 274.2 tonnes of lead entered the Baltic Sea as waterborne pollutants, while the atmospheric deposition accounted for 7.1 tonnes of cadmium and 234 tonnes of lead (HELCOM 2010). Several elements, such as Zn and Cu, are known to be essential elements for life, while others, such as Pb and Cd, do not play any physiological role and are highly toxic to all organisms even at low concentrations (Jakimska et al. 2011, El-Sadaawy et al. 2013).

Heavy metals are elements that occur naturally in the environment. Concentrations of mercury, cadmium, and lead are usually naturally very low and vary between areas of different geological origin. These metals are toxic, even at low concentrations, and have no known beneficial biological effects. Cadmium and lead concentrate in the liver where high levels can cause liver malfunction. Mercury concentrates on muscles. High metal concentrations can damage neurotransmitters and learning capability (mercury, lead) and bones or shell strength (cadmium). According to the integrated assessment, levels of lead in biota and sediment exceed the threshold levels in fish, bivalves and surface sediments in several basins of the Baltic Sea (HELCOM 2010).

Gills and skin are the first points of contact between waterborne metals and fish (Ahmed et al. 2016). The sensitive respiratory and ion regulatory membranes in the gills are the first target of the metal's adsorption onto the gill's surface for pollutants in water and could also significantly influence the total metal levels of the gills (Playle 1998, Bebianno et al. 2004). Gills are the first tissues to be exposed to metals and afterward distributed by the blood to important organs such as liver, skin, and muscle where they can exhort toxic effects causing metabolic stress situation and oxidative stress damage such as lipid peroxidation and can end up in the kidneys, as the main excretory organ (Barbier et al. 2005, Wepener et al. 2005, Ahmed et al. 2016). As Camusso and co-workers (1995) noted, Cd accumulated mostly in spleen and muscle; Hg in muscle and kidney; Pb in bone, spleen, and kidney; Cr in spleen, muscle, and gills; and Cu in kidney. The highest Zn levels were measured in gills of the rainbow trout.

This study aimed to assess the concentration of arsenic (As), chrome (Cr), manganese (Mn), and nickel (Ni) in the gills of the sea trout (*Salmo trutta* m. *trutta* L.) collected in the Baltic Sea (northern region of Poland). The results were then compared to permissible limits to detect whether the metal contamination levels in sea trout from the Baltic Sea exceed the values of limits admissible.

MATERIALS AND METHODS

Fish. Frequently consumed sea trout were purchased from local fisherman in January-February 2014, from Ustka town (54°34'43"N 16°52'09"E). It is a town in the Middle Pomerania region of northwestern Poland (Fig. 1). It is also part of Słupsk County in Pomeranian Voivodeship since 1999. Each of 25 fishes were collected and wrapped to the individual polyethylene bags for transport to the Department of Zoology and Animal Physiology, Institute of Biology and Earth Sciences, Pomeranian University in Słupsk and Department of Medical Biology and Biochemistry, Department of Ecology and Environmental Protection, Collegium Medicum in Bydgoszcz, Nicolaus Copernicus University in Toruń (Poland).

Fig. 1. Map of sampling location in Ustka (Pomeranian Voivodship, Poland)

Immediately after transportation to the laboratory, samples were washed with fresh water to remove the mud or other fouling substances. Then the gill tissues of each sample were removed and chopped into pieces by steam cleaned stainless steel knife. The gill tissues were then washed with deionized water and air-dried to remove the extra water.

Sample digestion. Microwave digestion with the use of concentrated nitric acid and hydrogen peroxide was used for the decomposition of the dried animal tissue, which was weighted into reaction vessels. The 8 ml of 69-70% of Baker Instra Analyzed

grade nitric acid was added together with 2 ml of 30% Chemical Lab. Analytical grade hydrogen peroxide. Then the samples were microwaved for 5 minutes in 190°C (ramping time 25 min.), then 5 minutes in 200°C (ramping time 5 min.) and finally 5 minutes in 210°C (ramping time 5 min.), to ensure the total decomposition of organic matter. The digested solution was moved to the disposable calibrated tubes and filled up to 50 ml with deionized 0.05 µS/cm water.

Inductively coupled plasma mass spectrometry (ICP-MS) technique. ICP-MS technique was used for the quantitative analysis of trace elements. The Agilent 7500ce ISP-MS apparatus is fitted with a micro-mist nebulizer, Peltier cooled double pass spray chamber, a peristaltic pump. Argon 5.0 was (99.999% purity) was used as a carrier gas. Apparatus is also fitted with a torch with a "shield torch" system reducing so-called "secondary discharge", off-axis ion lenses, reaction/collision chamber with hydrogen 6.0 and helium 6.0 (purity 99.9999%) as a reaction/collision gasses for the elimination of interferences. The vacuum system consists of a rotary pump and a turbo-molecular pump. Quadrupole with hyperbolic rods is the mass separator. The detector gives the possibility to work in two modes: digital and analog which gives the possibility to work through nine orders of magnitude. All determinations were made in the presence of ^{45}Sc, ^{89}Y, ^{159}Tb as an internal standard to minimize the matrix effect and ensure long term stability.

Quality control programs. The above procedure was also performed for the blank samples for the control of contamination. Simultaneously, for every series of samples, the certified reference material (NCS ZC73016 chicken) from China National Analysis Center for Iron and Steel was used to ensure quality control requirements. Recoveries ranging from 90-110% were achieved for this material and uncertainty of measurement was established at 10%.
The ICP-MS 7500ce apparatus from Agilent was used for the determination, equipped with a micro-mist type nebulizer, thermoelectrically cooled by means of the Peltier effect, double-pass type fog chamber. The sample pumping speed was between 0.01 and 05 rpm. Argon 5.0 with a high purity of 99.999% was used as the carrier gas. The camera is equipped with a quartz burner with the "shield torch" option to prevent the formation of so-called "Secondary discharge", i.e. electrical discharges arising between plasma generated from Argon and camera cones. Standard nickel sampler and skimmer cones were used as well as CE type ion lenses misaligned to better eliminate polyatomic interference.
The apparatus is also equipped with an ORS (octopole reaction system) type reaction chamber to eliminate interference both polyatomic and those derived from double-charged ions. In the reaction chamber, hydrogen 6.0 and helium 6.0 (purity 99.9999%) were used as reaction gases to eliminate interference. The apparatus vacuum system consists of an oil pre-pump and a turbo-molecular pump. The mass splitter is a quadrupole equipped with hyperbolic rods to create the correct electric field. The detector, i.e. electron duplicator with the possibility of working in plus and analog modes, which allows achieving a 9-row dynamic range. In order to maintain apparatus stability and minimize matrix effects, all determinations were made in the presence of 45Sc, 89Y, 159Tb as an internal standard.

Statistical analysis. Statistical analysis of the data obtained was performed by employing the mean ± standard error of the mean (S.E.M.). All variables were tested for normal distribution using the Kolmogorov–Smirnov test ($p > 0.05$). All statistical analyses were performed using Statistica 8.0 software (StatSoft, Poland).

RESULTS AND DISCUSSION

The concentration of heavy metals in the gills of sea trout from the Baltic Sea was listed in Table 1. All metal concentrations were determined on a weight basis. Metal contents in fish samples were found 0.00313-0.02069 mg·kg^{-1} for As (the mean value was 0.01294±0.0009 mg·kg^{-1}), 0.0118-0.02161 mg·kg^{-1} for Cr (0.01449±0.0003 mg·kg^{-1}), 0.01895-0.14216 mg·kg^{-1} for Mn (0.04137±0.0046 mg·kg^{-1}), 0.00664-0.01528 mg·kg^{-1} for Ni (0.00811±0.00034 mg·kg^{-1}).

Table 1

The concentrations of heavy metals in the gills of sea trout from the southern Baltic Sea (n = 25)

Elements	The mean value, M±m, mg·kg^{-1}	The range of values	
		min, mg·kg^{-1}	max, mg·kg^{-1}
Arsenic, As	0.01294±0.0009	0.00313	0.02069
Chrome, Cr	0.01449±0.0003	0.0118	0.02161
Manganese, Mn	0.04137±0.0046	0.01895	0.14216
Nickel, Ni	0.00811±0.00034	0.00664	0.01528

According to these data, the ranking order of the mean concentration of the heavy metals in fish gills was Mn (0.04137 mg·kg^{-1}) > As (0.01294 mg·kg^{-1}) > Cr (0.01449 mg·kg^{-1}) > Ni (0.00811 mg·kg^{-1}) (Table 1).

Arsenic is one of the most potentially toxic heavy metals present in the environment and originates from both natural and anthropogenic processes (Saha et al. 2016, Ullah et al. 2017). According to USFDA (1993b), about 90 percent of total human exposure of As instigates from fish and other seafood. According to the results of our study, the maximum arsenic concentration was 0.02069 mg·kg^{-1} (Table 1). As for inorganic arsenic, WHO established the provisional tolerable weekly intake (PTWI) value as 0.015 mg·kg^{-1} weekly per body mass (WHO 1998). As can result from this legal limit, a person weighing 70 kg can take a maximum of 1.05 mg of inorganic arsenic in a week (Kaya and Turkoglu 2017). Consistent with several standards, the permitted limit of arsenic in fish and seafood for human consumption is 1 mg·kg^{-1} of wet weight (Australia New Zealand Food Authority 1998). The US Environmental Protection Agency (USEPA) has considered an upper limit of arsenic equal to 1.3 mg·kg^{-1} of wet weight in fish as a safe criterion for human health (Eisler 1994). Considering these legal limits, the results of this study demonstrate that total arsenic concentrations in gills did not exceed the permitted limit of arsenic. Moreover, the mean concentration of arsenic in gill samples (0.01294±0.0009 mg·kg^{-1}) was much below the permissible limit of USFDA (1993b) which is 76 mg·kg^{-1} and FAO/WHO (1976) which is 0.1-0.15 mg·kg^{-1}.

Usydus and co-workers (2005) determined the total arsenic (As) contents in the muscle tissue of the major Baltic fish species: herring, sprat, cod, and flounder, caught in different areas of the Baltic Sea. It was also evaluated fish size-dependent differences in As content on flounder as an example. Flounder is a fairly stationary species and therefore may be a good model for comparisons of the area-dependent degree of pollution. The maximum acceptable As content in fish and fish products in 2003 was 4.0 $mg \cdot kg^{-1}$ wet weight (w.w.). In none of the samples was this level exceeded. The highest mean As contents (0.78 $mg \cdot kg^{-1}$ w.w.) was typical of sprat, the lowest (0.26 $mg \cdot kg^{-1}$ w.w.) being recorded in flounder. The species studied differed significantly ($p < 0.05$) in their As contents in the muscle tissue, except for cod and flounder that showed no significant interspecific differences ($p > 0.05$). The Eastern Baltic flounder showed significantly ($p < 0.05$) higher As contents than their conspecifics harvested in both the Central and the Western Baltic. As contents in the 30-40 cm long flounder were significantly ($p < 0.05$) higher than those recorded in smaller fish, i.e., those measuring 20-30 cm and those smaller than 20 cm (Usydus et al. 2005).

Two oxidation states of chromium are considered to be biologically and environmentally relevant based on their stability in the presence of water and oxygen. Compounds containing hexavalent chromium (Cr^{6+} are mutagenic and carcinogenic when inhaled and potentially when ingested orally in large quantities as well (Vincent and Lukaski 2018). Numerous critical reviews have evaluated exposure to toxic and carcinogenic hexavalent chromium Cr^{6+} from a number of pathways; including workplace air, cement, and packaging materials. The contribution of foodstuffs to dietary Cr^{6+} has been increasingly under investigation also (Hamilton et al. 2018). The main sources of Cr in the human diet are meat, dairy products, bread, and tea (Lendinez et al. 2001). Dietary chromium apparently is absorbed *via* passive diffusion and the extent of absorption is low (~1%). Chromium is maintained in the bloodstream bound to the protein transferrin. It is generally believed to be delivered to tissues by transferrin *via* endocytosis (Vincent and Lukaski 2018). Chromium has been postulated to be involved in regulating carbohydrate and lipid (and potentially also protein) metabolism by enhancing insulin's efficacy (Vincent and Lukaski 2018). Trivalent chromium has also been proposed as a therapeutic agent to increase insulin sensitivity and affect lipid metabolism. Recent case studies examining intravenous infusions of chromium (generally 3 μg/h) as a treatment for glucose intolerance have found that chromium reduced insulin requirements for subjects with hyperglycemia (Vincent 2017). It is likely that any ingested Cr(VI) will be reduced to Cr^{3+} in the acidic conditions of the stomach (Milacic and Stupar 1994), and any Cr^{6+} transferred from soil to plant will also be reduced (Hamilton et al. 2018). In our study, the minimum and maximum chromium values were found as 0.0118 $mg \cdot kg^{-1}$ and 0.02161 $mg \cdot kg^{-1}$ in the gills of sea trout, respectively (Table 1). The mean Cr content in the gill of sea trout samples (0.01449±0.0003 $mg \cdot kg^{-1}$) was well within the toxic limit of USFDA (1993a). The samples had lower Cr concentration as compared to the limits of 0.200 set by FSANZ (2002) and 0.100 by EUROPA (2004).

In the study of Yin and co-workers (2019), bighead carp (*Aristichthys nobilis*) were exposed to waterborne Cr^{6+} of 0.01, 0.1, 1 and 5 mg/L for 14 days and subsequently transferred to clean water for another 14 days. The Cr^{6+} contents in some edible parts, such as dorsal muscle, ventral muscle, and head were detected. The Cr^{6+} concentra-

tions in the three parts were in the order of head > ventral muscle > dorsal muscle with a significant increase during the exposure period and a remarkable decrease when kept at clean water during the deputation stage. The head contained higher fat than that of muscle and the Cr^{6+} levels of these parts showed a significantly positive correlation with fat content, however, the Cr^{6+} contents in the separated fat were extremely low. Therefore, the head of fish poses a higher potential health risk than muscle due to heavy metals pollution (Yin et al. 2019).

Manganese (Mn) is an essential element for humans, animals, and plants and is required for growth, development, and maintenance of health (Santamaria and Sulsky 2010). Upon fast absorption into the body *via* oral and inhalation exposures, Mn has a relatively short half-life in blood, yet fairly long half-lives in tissues. Recent data suggest Mn accumulates substantially in bone, with a half-life of about 8-9 years expected in human bones (O'Neal and Zheng 2015). High-dose oral, parenteral, or inhalation exposures are associated with increased tissue Mn levels that may lead to the development of adverse neurological, reproductive, or respiratory effects. Manganese-induced clinical neurotoxicity is associated with a motor dysfunction syndrome commonly referred to as manganism (Santamaria and Sulsky 2010). In the present study, the minimum concentration of Mn was 0.01895 $mg \cdot kg^{-1}$, while the maximum concentration was 0.14216 $mg \cdot kg^{-1}$ (the mean level was 0.04137±0.0046 $mg \cdot kg^{-1}$) (Table 1). Our study reported that the accumulation of Mn was exceeding the maximum permissible limit (by 8.27-16.55-fold) according to WHO/EPA standard (0.0025-0.005 $mg \cdot kg^{-1}$) (FAO/WHO 1976).

In the case of nickel presence in gills, the minimum and maximum levels were 0.00664 and 0.01528 $mg \cdot kg^{-1}$ (the mean level was 0.00811±0.00034 $mg \cdot kg^{-1}$) (Table 1). The proposed limit of Ni concentrations in marine fish species as recorded by FAO (1983) is about 10 $mg \cdot kg^{-1}$ and 0.5-0.6 $mg \cdot kg^{-1}$ according to WHO Guidelines for drinking water quality (1985). In general, it can be seen that the concentrations of Ni found in gills of the sea trout in this study are still considered as those of uncontaminated fish.

Nickel is known to exert its toxic effects on aquatic biota *via* three key mechanisms: inhibition of respiration, impaired ion regulation, and stimulation of oxidative stress. Blewett and co-workers (2016) have studied the nickel effects on respiration, ion regulation and oxidative stress in the galaxiid fish, *Galaxias maculatus*. Inanga (*Galaxias maculatus*) is a euryhaline and amphidromous Southern hemisphere fish species inhabiting waters highly contaminated in trace elements such as nickel (Ni). Inanga acclimated to freshwater (FW), 50% seawater (SW) or 100% SW were exposed to 0.150 or 2,000 μg Ni per L, and tissue Ni accumulation, metabolic rate, ion regulation (tissue ions, calcium (Ca) ion influx), and oxidative stress (catalase activity, protein carbonylation) were measured after 96 h. Ni accumulation increased with Ni exposure concentration in gill, gut and remaining body, but not in the liver. Only in the gill was Ni accumulation affected by exposure salinity, with lower branchial Ni burdens in 100% and 50% SW inanga, relative to FW fish. There were no Ni-dependent effects on respiration, or Ca influx, and the only Ni-dependent effect on tissue ion content was on gill potassium. Catalase activity and protein carbonylation were affected by Ni, primarily in FW, but only at 150 μg Ni per L (Blewett et al. 2016).

The concentrations of heavy metals in the water, sediment, and muscle of various fish species in Europe have been reported in the literature. For example, the moni-

toring survey to assess the environmental pollution status of the river Morava was carried out in 2014 by Dvořák and co-workers (2015). The study of these researchers was presented the concentrations of heavy metals (Hg, Cr, Zn, Pb, and Cd) in the water, sediment and muscle tissue of European chub (*Squalius cephalus*) from the middle and lower reaches of the Morava River basin (Bečva, Dřevnice, Haná, Kyjovka and Morava rivers), in the Czech Republic. Results showed a positive significant correlation between the concentration of Hg, Pb, Cd, Cr and Zn in muscles and age of fishes ($p < 0.05$). The contents of the analyzed metals in European chub muscles were low Hg 0.049-0.402, Pb 0.005-0.035, Cd 0.006-0.026, Cr 0.016-0.042 and Zn 5.59-64.31 $mg \cdot kg^{-1}$ wet weight basis. The contents of the analyzed metals in European chub muscles were low at monitoring sites and did not exceed the values of limits admissible in the Czech Republic.

The concentrations of heavy metals (Cr, Cd, Hg, Zn, and Pb) in the water, sediment, and muscle of two fish species from the middle and lower reaches of the Dyje River basin (Dyje, Jihlava, Svratka, Svitava and Trkmanka rivers), in the Czech Republic were analyzed by Dvořák and co-workers (2014). The potential ecological risk analysis of heavy metal concentrations in the sediments indicated that nine sites in the middle and lower reaches posed a moderate or considerable ecological risk. The health risk analysis of individual heavy metals in fish muscle indicated safe levels for the general population, but there was a possible risk in terms of the total target hazard quotients. The contents of the analyzed metals in fish muscles were low at seven sites and did not exceed the values of limits admissible in the Czech Republic (Dvořák et al. 2014).

Valová and co-workers (2010) have demonstrated that the Morava River basin (the Czech Republic, Danube basin) does not represent a threatening source of mercury, cadmium or lead for the Danube River downstream. These researchers have assessed the mercury (Hg), cadmium (Cd) and lead (Pb) contamination in muscle tissue of fishes over the longitudinal profile of the Morava River (the Czech Republic, Danube basin) and to detect any temporal trends over the past 18 years before the study period. Fish samples were collected in 1992, 1998, 2000, 2003 and 2009 at 6 study sites situated just downstream of important pollution sources. A catastrophic flood in 1997 resulted in an increase in metal concentrations, especially cadmium and lead, in the following 1998 season. Chub (*Leuciscus cephalus* L.) were selected as indicator species at 5 sites, and brown trout (*Salmo trutta* m. *fario* L.) at the uppermost site where chub does not occur. In total, the muscle tissue of 175 specimens of chub and 19 specimens of brown trout was analyzed. Concentrations of heavy metals ranged as follows: mercury 0.015-0.369 $mg \cdot kg^{-1}$; cadmium 0.001-0.254 $mg \cdot kg^{-1}$ and lead 0.006-1.505 $mg \cdot kg^{-1}$. Mercury levels did not exceed the maximum allowed concentration in the Czech Republic (0.5 $mg \cdot kg^{-1}$). Content of cadmium and lead in fish muscle exceeded the maximum allowed levels (0.05 and 0.3 $mg \cdot kg^{-1}$ respectively) in 11 and 4 samples, respectively. On average, the order of metal concentration in fish muscle was: Hg > Pb > Cd. No significant differences were found between sites along with the longitudinal profile of the river. Significant differences were found, however, for the interannual comparison of cadmium and lead (but no mercury) at different sites ($p < 0.05$) (Valová et al. 2010).

In Serbia, Milošković and co-workers (2016) have monitored the contamination of fish muscle tissue by elements Al, As, Cd, Co, Cr, Cu, Fe, Hg, Mn, Ni, Pb, and Zn at 17 sampling sites, in order to assess the pollution status of the main rivers. Of the six commercially important fish species included in the study (pikeperch *Sander lucioperca*, catfish *Silurus glanis*, bream *Abramis brama*, barbel *Barbus barbus*, chub *Squalius cephalus*, nase *Chondrostoma nasus*), the bioconcentration factor (BCF) indicated that benthivore bream and barbel and predatory catfish have the highest tendency toward the accumulation of elements. The estimated metal pollution index (MPI) showed the Tisa River to be unaffected by direct pollution (with an MPI value of 0.31) and the West Morava and Pek rivers to be affected (with MPI values of 1.92 and 0.73 for the WM1 and WM2 sampling sites and 0.65 for the Pek sampling site). Hg concentrations exceeded the maximum permitted concentrations (MPCs) only in catfish samples (0.62 $mg \cdot kg^{-1}$) from the Danube (D3 sampling site) and barbel (0.78 $mg \cdot kg^{-1}$) from the West Morava (WM1 sampling site), while Cd concentrations exceeded the MPC in catfish samples (0.09 $mg \cdot kg^{-1}$) from the Danube (D1 sampling site) and chub samples (0.1 $mg \cdot kg^{-1}$) from the South Morava (SM2 sampling site). The average concentrations of Pb exceeded the MPC in chub and barbel samples (0.32 and 0.82 $mg \cdot kg^{-1}$, respectively) from the West Morava (WM1 sampling site); chub, barbel, and nase samples (0.35, 0.32, 0.31 $mg \cdot kg^{-1}$, respectively) from the West Morava (WM2 sampling site); chub and barbel samples (0.35 and 0.3 $mg \cdot kg^{-1}$, respectively) from the Ibar; chub samples (0.39 $mg \cdot kg^{-1}$) from the Drina; chub and barbel samples (0.59 and 0.4 $mg \cdot kg^{-1}$, respectively) from the Great Timok; and nase samples (0.33 $mg \cdot kg^{-1}$) from the Pek (Milošković et al. 2016).

In Poland, Łuczyńska and co-workers (2018) have assessed the heavy metals content (Zn, Cu, and Hg) in gills, liver, gonads, and muscles of perch, *Perca fluviatilis* (L.) and roach, *Rutilus rutilus* (L.) from the Pluszne Lake (north-eastern Poland). As expected, muscles contained the significantly highest values of Hg ($p \leq 0.05$). The concentrations of Zn were significantly higher in gills of roach and gonads of perch ($p \leq 0.05$), while the liver of fish accumulated significantly more Cu than other organs ($p \leq 0.05$). In all organs of perch the higher content of mercury was found ($p \leq 0.05$). The value of Zn and Cu was highest in organs of roach ($p \leq 0.05$) (with the exception of Zn in muscles $p > 0.05$). The sequence of metals in both species was Zn > Cu > Hg. Only in muscle tissue, Hg was significantly positive correlated with weight of roach ($r = 0.811$, $p = 0.045$) and perch ($r = 0.652$, $p = 0.041$), and total length of roach ($r = 0.806$, $p = 0.005$). A positive relationship was also observed between Zn concentration in gills of perch and their weight ($r = 0.634$, $p = 0.049$). In contrary, Zn in gills of roach decreased with weight ($r = -0.693$, $p = 0.026$) and length ($r = -0.668$, $p = 0.035$). Cu concentration in liver of perch was statistically positively correlated with HSI ($r = 0.717$, $p = 0.020$), whereas Hg content in muscle tissue of roach with FCF ($r = 0.643$, $p = 0.045$). There was a negative relationship between Hg in perch gonads and GSI ($r = -0.808$, $p = 0.005$). Metal pollution index (MPI) in gills, liver, gonads, and muscles of roach was 7.68, 7.24, 6.77 and 3.13, respectively, whereas in these organs of perch was 3.25 (gills), 4.75 (liver), 5.84 (gonads) and 4.44 (muscles), therefore the contamination of each tissue ranged from very low contamination to low contamination. The concentration of mercury was lower than the maximum acceptable limit estimated by the Commission Regulation (EC) No 629/2008 of 2 July 2008. The values of HI

and THQ were below 1, which means that the consumption of these fish is not hazardous to the consumer (Łuczyńska et al. 2018).
Fish and fishery products available in Poland are safe for consumers. Winiarska-Mieczan and co-workers (2018) have verified whether fish and fishery products available on the Polish market were safe for consumers in terms of Cd and Pb content. Safety was evaluated according to the content of Cd and Pb in fishery products and based on the share of such products in supplying Cd and Pb in the weekly diet of an adult. Fish samples, of which 139 were smoked fish (26 samples of mackerel, 21 of salmon, 35 of sprat, 38 of eel and 19 of trout) and 117 samples of prepared fish-based dishes (20 of salads, 41 of spreads and 56 of marinated herring) were analyzed. The content of Cd and Pb was determined using the GF AAS method. The content of Cd per 1 kg of the analyzed product can be represented as follows: salads > smoked eel > smoked salmon and mackerel > smoked trout and spreads > marinated herring > smoked sprat. The content of Pb per 1 kg of the analyzed product can be represented as follows: smoked salmon and salads > smoked mackerel and spreads > smoked eel > smoked sprat and smoked trout. Most Cd was found in salads (on average 10.7 $\mu g \cdot kg^{-1}$; range 6.53-14.7 μg), whereas most Pb was recorded in salads (on average 56.8 μg per kg; range 32.6-78.9 μg) and marinated fish (on average 58.8 μg per kg; range 19.8-79.6 μg).
The concentrations of iron, manganese, zinc, and copper in selected tissues of two fish species: pike (*Esox lucius* L.) and bream (*Abramis brama* L.) living in lakes Ińsko and Wisola, Northwestern Poland were studied by Rajkowska and Protasowicki (2013). The lakes differ in their trophic status. The effect of gender and environmental conditions on metals accumulation was also investigated. The mean metal concentrations (micrograms per gram wet weight) in both lakes occurred in the following ranges: Fe 0.8-240.6, Mn 0.2-8.4, Zn 3.0-185.9, and Cu 0.14-7.76. The lowest levels of the studied metals were always detected in the muscles. The spleen, kidneys, and liver were found to accumulate the highest amounts of Fe. In the case of the other metals, the highest levels were found, as follows: Mn in skin, gills, and gonads, Zn in the digestive tract and gills, Cu in the liver. Heavy metal content in fish gonads was observed to be sex-dependent (Rajkowska and Protasowicki 2013).
It is very difficult to compare the concentration of metals in similar tissue from different species due to factors such as physiological tolerance, regulatory mechanisms, and body reaction of fish species as well as their tendency to bind to specific cellular molecular groups and to the metabolic characteristics of tissues (Mwashote 2003).

CONCLUSIONS

This study was performed to evaluate the metal concentrations (arsenic, chrome, manganese, and nickel) in well-known and most consumed fish species in the northern region of Poland. It is important to mention that the observed manganese concentrations exceeded their threshold level as described in international standards in whole fish (if utilized for any processing).
Metal contents in fish samples were found 0.00313-0.02069 $mg \cdot kg^{-1}$ for As (the mean value was 0.01294±0.0009 $mg \cdot kg^{-1}$), 0.0118-0.02161 $mg \cdot kg^{-1}$ for Cr (0.01449±0.0003 $mg \cdot kg^{-1}$), 0.01895-0.14216 $mg \cdot kg^{-1}$ for Mn (0.04137±0.0046 $mg \cdot kg^{-1}$), 0.00664-

0.01528 mg·kg^{-1} for Ni (0.00811±0.00034 mg·kg^{-1}). According to these data, the ranking order of the mean concentration of the heavy metals in fish gills was Mn (0.04137 mg·kg^{-1}) > As (0.01294 mg·kg^{-1}) > Cr (0.01449 mg·kg^{-1}) > Ni (0.00811 mg·kg^{-1}). The mean concentration of arsenic in gill samples (0.01294±0.0009 mg·kg^{-1}) was much below the permissible limit of USFDA (1993b) and FAO/WHO (1976). The mean Cr content in the gill of sea trout samples was well within the toxic limit of USFDA (1993b). The samples had lower Cr concentration as compared to the limits of 0.200 set by FSANZ (2002) and 0.100 by EUROPA (2004). Our study reported that the accumulation of Mn was exceeding the maximum permissible limit (by 8.27-16.55-fold) according to WHO/EPA standard (0.0025-0.005 mg·kg^{-1}) (FAO/WHO 1976). The proposed limit of Ni concentrations in marine fish species as recorded by FAO (1983) is about 10 µg/g and 0.5-0.6 µg/g according to WHO Guidelines for drinking water quality (1985). In general, it can be seen that the concentrations of Ni found in gills of the sea trout in this study are still considered as those of uncontaminated fish. In conclusion, the assessment of element contents in the organs of the sea trout appears to be a useful biomarker to evaluate the toxic effects of heavy metal pollution as well as for human consumption. It is recommended that additional monitoring plans be conducted to determine critical bioaccumulation levels in fish species of the southern Baltic Sea (central Pomeranian zone). Our results would also be helpful for the safe consumption of the selected indigenous fish species.

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BIOAKUMULACJA ARSENU, CHROMU, MANGANU I NIKLU W SKRZELACH TROCI WĘDROWNEJ (*SALMO TRUTTA* M. *TRUTTA* L.) Z POŁUDNIOWEJ CZĘŚCI MORZA BAŁTYCKIEGO (REGION POMORZA ŚRODKOWEGO)

Streszczenie

Celem badań była ocena stężenia arsenu (As), chromu (Cr), manganu (Mn) i niklu (Ni) w skrzelach troci wędrownej (*Salmo trutta* m. *trutta* L.) z Morza Bałtyckiego (północny region Polski). Wyniki zostały porównane z wartościami dopuszczalnych stężeń granicznych. Zawartość pierwiastków w skrzelach analizowano przy użyciu techniki spektrometrii mas sprzężonej z plazmą wzbudzaną indukcyjnie (ICP-MS). Zawartość arsenu w próbkach skrzeli ryb wynosiła 0.00313-0.02069 mg·kg^{-1} (średnia wartość 0.01294±0.0009 mg·kg^{-1}), chromu – 0.0118-0.02161 mg·kg^{-1} for Cr (0.01449±0.0003 mg·kg^{-1}), manganu – 0.01895-0.14216 mg·kg^{-1} (0.04137±0.0046 mg·kg^{-1}) oraz niklu – 0.00664-0.01528 mg·kg^{-1} (0.00811±0.00034 mg·kg^{-1}). Kolejność uszeregowania średniego stężenia metali w skrzelach ryb: Mn (0.04137 mg·kg^{-1}) > As (0.01294 mg·kg^{-1}) > Cr (0.01449 mg·kg^{-1}) > Ni (0.00811 mg·kg^{-1}). Średnie stężenie arsenu w próbkach skrzeli (0,01294±0,0009 mg·kg^{-1}) było znacznie poniżej dopuszczalnych wartości według USFDA (1993a) i FAO/WHO (1976). Średnia zawartość Cr w skrzelach znajdowała się w granicach limitu toksyczności zgodnie z USFDA (1993a). Wykazano niższe stężenie Cr w skrzelach troci wędrownej w porównaniu z dopuszczalnym stężeniem ustalonym przez FSANZ (2002) i EUROPA (2004). Analiza wykazała natomiast przekroczenie kumulacji Mn (8,27-16,55-krotnie) w porównaniu z wartościami dopuszczalnymi tego pierwiastka zgodnie ze standardami WHO/EPA (0,0025-0,005 mg·kg^{-1}) (FAO/WHO 1976). Dopuszczalne stężenie Ni w gatunkach ryb morskich zarejestrowane przez FAO (1983) wynosi około 10 µg/g i 0.5-0.6 µg/g (WHO 1985). Analiza stężenia niklu w skrzelach troci wędrownej wykazała wartości tego metalu poniżej dopuszczalnych stężeń granicznych.