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ORIGINAL PAPER

# The total Hg concentration in tissues of freshwater fish from northeastern Poland\*

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#### Abstract

There is a constant need to monitor mercury pollution in the environment, especially in water, due to the methylation of Hg and its increased bioavailability. Fish are a good indicator material, but the lethal method of sampling for testing could be replaced with a non-lethal one. The aim of the study was to determine the mercury content in selected tissues of freshwater fish species, commonly consumed in Poland, to check whether fish scale testing will be an alternative to lethal testing for mercury contamination. Crucian carp (Carassius carassius), tench (Tinca tinca), and perch (Perca fluviatilis) were obtained from a breeding pond in Knyszyn (Podlaskie Voivodeship, Poland). Mercury was analyzed in the skeletal muscles and liver (samples for the lethal method) and scales (samples for the non-lethal method). The AMA 254 mercury analyzer was used, and the method based on atomic absorption spectrometry with thermal decomposition of a sample in a flow of oxygen was applied. The results confirm the phenomenon of mercury biomagnification in the tissues of the tested fish and emphasize that even low concentrations in the natural environment can accumulate in living organisms. The highest mercury contents occurred in the species occupying a higher link in the trophic chain (perch). However, the mercury content in the muscles of the fish tested for consumption did not exceed the mercury content limits specified in the law (Commission Regulation (EU) 2023/915), i.e., up to 0.50 mg kg<sup>-1</sup>. Scales can be an interesting indicator of environmental pollution with Hg, but their Hg content was significantly lower compared to the concentration of this element in the muscle and liver of perch, tench, and crucian carp.

Keywords: fish, mercury, biomagnification, muscles, liver, scales

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### INTRODUCTION

Today, environmental pollution is a serious problem for animal species and humans. Hazardous heavy metals, including highly toxic mercury, are entering ecosystems through extensive chemical and mining industries and fuel combustion, and in Asian countries also through the use of fertilizers and pesticides containing mercury (Mania et al. 2012). At present, the proportion of natural sources of mercury to sources of anthropogenic origin, which together contribute to the total atmospheric burden, is not precisely determined (Zillioux 2015, Gworek et al. 2020). According to Seigneur et al. (2003), natural emissions and direct anthropogenic emissions are approximately equal. Penetrating into the environment, mercury accumulates in various chemical forms, undergoes transformations, and then returns to the atmosphere, thus creating a cycle of transformations (Jedruch et al. 2021, Blanchfield et al. 2022, Wu et al. 2024). Bioaccumulation occurs in plant and animal organisms along the path of these transformations. If an organism at a lower level in the trophic chain absorbs mercury, then each subsequent organism at a higher level is able to accumulate a higher concentration of mercury in its body. This phenomenon is called biomagnification. Mercury accumulates to a large extent in aquatic organisms and bottom sediments. For this reason, three species of freshwater fish were selected for the study: crucian carp (Carassius carassius) and tench (Tinca tinca) as benthic fish of the Cyprinidae family, and a piscivore, common perch (Perca fluviatilis). The crucian carp (Carassius carassius) has no size limit or closed season in Poland; it reaches up to 40 cm in length and up to 2 kg in weight, but in unfavorable conditions, crucian carps can become dwarfed, and in the presence of piscivores such as pike, crucians are deep bodied (Olsén, Bonow 2023). It occurs in shallow waters of standing or slowly flowing reservoirs, as well as in overgrowing ponds and coastal zones of larger lakes. The food of the common crucian carp is benthic fauna and aquatic vegetation. Tinca tinca is an important gastronomic and angling fish, living in shallow areas of lakes and ponds, preferring muddy or sandy substrates with submerged vegetation that favors their reproduction (Martín-Vertedor et al. 2025, Skibniewski et al. 2025). It has no closed season in Poland, but it has a protective size of up to 25 cm. It reaches up to 50 cm in length and up to 4 kg in body mass. Like Carassius carassius, tench feeds at the bottom. It eats insect larvae, snails, and mussels. Perch is a predatory fish that hunts invertebrates, smaller species of fish, and eats the roe of other species, but they shift their diets and feeding behaviors as they increase in body size (Ning et al. 2025). The protective size is up to 15 cm, and there is no protective period in Poland. It reaches up to 45 cm in length and 2.5 kg in body mass. It occurs in all types of water bodies, and is most abundant in lakes (Lyach, Remr 2019).

In aquatic ecosystems, mercury occurs in elemental (Hg<sup>0</sup>), inorganic

(HgII), and organic forms. In the case of aqueous organic mercury, it can be divided into two categories. The first is the covalent bonding of mercury with methyl groups (-CH<sub>2</sub>). These compounds include methylmercury (CH<sub>2</sub>Hg/MeHg) and dimethylmercury ((CH<sub>2</sub>)<sub>2</sub>Hg). Dimethylmercury is less important in the transport and transformation of mercury than methylmercury. The second category includes mercury complexes with organic matter, e.g., humic substances (Zhu et al. 2018, Aaseth et al. 2020, Wu et al. 2024). In the methylation process, aquatic microorganisms play a key role – sulfur and iron-reducing bacteria and methanogenic bacteria - present in sediments and wetland catchments. The methylation process produces methylmercury (the most common organic mercury compound in the aquatic environment and characterized by very high toxicity) and then dimethylmercury (Charkiewicz et al. 2025). Dimethylmercury is insoluble in water and highly volatile. Thanks to this, they easily penetrate the atmosphere through the process of diffusion. This is part of the mercury transformation cycle because after penetrating the atmosphere, they return to the aquatic environment with precipitation. Mercury absorption in fish occurs through food (then bioaccumulation is combined with biomagnification) and absorption through gills and body surfaces (US EPA 2000, Ravichandran 2004, Gworek et al. 2020, Donadt et al. 2021, Blanchfield et al. 2022). Methylmercury is taken up by small organisms floating in the water (phytoplankton and zooplankton). These organisms are then eaten by others, e.g., fish, which in turn are eaten by larger predatory species. Taking biomagnification into account, humans are exposed to the absorption of high concentrations of mercury, especially very toxic methylmercury, by consuming fish and crustaceans. This is dangerous because mercury in the human body leads to organ damage, mainly of the kidneys, and disorders related to the nervous system (Rice et al. 2014, Bradley et al. 2017, Kimáková et al. 2018, Ma et al. 2019, Charkiewicz et al. 2025). Therefore, it is important to know the concentration levels of this element in fish organisms in order to roughly assess the level of contamination of water bodies and estimate the risk of human exposure (de Almeida Rodrigues et al. 2019).

The aim of the study was to determine the total mercury concentration in the organs of freshwater fish representing three common species inhabiting Polish waters. An attempt was made to determine the relationship between the Hg content in the various organs analysed and whether its concentration in scales could be considered as a marker of the whole organism's burden by this toxic metal, which could serve as an indicator tissue taken from live individuals.

#### MATERIALS AND METHODS

10 crucian carp (*Carassius carassius*), 8 tench (*Tinca tinca*), and 10 common perch (*Perca fluviatilis*) were captured from breeding ponds — Ełk-Knyszyn fishing farm (Podlaskie Voivodeship) in Poland in 2023. The area of the facility is about 108 ha. At a distance of about 300 meters, there is a provincial road, and at a distance of about 700 m, national road 65. The ponds are surrounded by arable fields and forests. The fish were fed with grain. Data on basic morphological parameters of the fish studied are shown in Table 1. All individuals studied were obtained from fishermen; hence, according to Polish law, the study did not require the approval of the ethics committee.

Fish were killed by blunt head injury, destroying central nervous system activity. The gill arch was then cut for bleeding. Before the material collection, the individuals were weighed (model Radwag E425) with an accuracy of 0.01g and measured by means of a ruler – the maximum total length of the body was determined with an accuracy of 1 mm. Then, the material for analysis was collected: skeletal muscles and scales from the left side, from the area of the body over the lateral line under the dorsal fin. In the case of scales, an additional procedure was used, consisting of preliminary cleaning to remove surface contamination, mucus, skin residues, and other tissues on the scales and rinsing the scales in deionized water for about 5 minutes. All samples were stored at -20°C until analysis in sealed glass petri dishes.

The collected tissues were examined using the AMA 254 mercury analyzer (ALTEC, Czech Republic). The spectrometer, equipped with a mercury lamp and a silicon UV diode, detects mercury by atomic absorption at a wavelength of 254 nm. During this process, free mercury atoms absorb radiation produced by a mercury lamp, the cathode of which is made of Hg. As a result, the original intensity of radiation emitted by the lamp is reduced, which is recorded by the spectrometer. Total mercury (THg) content in muscles, livers, and scales was determined and expressed as mg kg-1 wet weight. The research material did not require prior mineralization. The accuracy of the results was verified using a standard reference material (BCR-CRM 464, Tuna, IRMM, Belgium). The analytic technique is based on generating mercury vapors, regardless of the form of the sample, and the device is characterized by high sensitivity. The advantage of this method is the possibility of analyzing small samples (20 - 60 mg). Such weights were prepared to assess the mercury content in the tested material. Samples were analyzed as quickly as possible after defrosting to minimize the moisture loss that could cause Hg concentrations to be overestimated.

Statistical analysis of the results was conducted using the Statistica 13 program. Data distribution was analyzed using the Shapiro-Wilk W test. Due to the distribution of values, nonparametric data analysis was per-

Data on the fish studied

Table 1

Species	n	Specification	Mean	$^{\mathrm{SD}}$	Median	Min	Max	$Q_{25}$	$\mathrm{Q}_{75}$
Perca	0	body mass (kg)	160.70	21.44	153.50	130.00	190.00	143.00	183.00
fluviatilis	01	body length (mm)	24.15	2.17	24.25	21.00	27.50	22.00	26.00
Carassius	10	body mass (kg)	251.00	60.42	234.50	188.00	358.00	200.00	301.00
carassius	10	body length (mm)	23.90	3.32	23.00	20.00	30.00	21.00	26.00
	G	body mass (kg)	530,38	94.20	503.00	448.00	720.00	466.00	568.50
דווכם וווכם	0	hody length (mm)	32.75	2.19	32.00	30.00	36.00	31.50	34.50

n-number of individuals, Q25-lower quartile, Q75-upper quartile, SD-standard deviations, Min. -minimum, Max. -maximum

formed. In order to compare intergroup differences, the Mann-Whitney U test was performed at the significance level of  $p \le 0.05$  and  $p \le 0.01$ .

#### RESULTS AND DISCUSSION

The highest Hg concentrations were recorded in the tissues of common perch (Perca fluviatilis). The highly statistically significant difference in mercury content in the tissues of perch compared to crucian carp and tench, both representing lower levels of the food chain, clearly reflects the phenomenon of biomagnification (Table 2). Our observations do not diverge from the results of other research teams. The tissues of apex predators in the food chain accumulate toxic elements. Łuczyńska et al. (2006) observed the highest concentration of mercury in predatory fish (pike and perch), and these findings are in agreement with the results of the present study. Piscivores like pike and studied perch accumulate mercury in their bodies with food, but Hg distribution in their tissues may vary depending on the degree of pollution of the reservoir. Havelková et al. (2008) reveal that in non-polluted reservoirs or reservoirs with low contamination, mercury is accumulated to a greater extent in the muscles, while in reservoirs with a high level of contamination, the liver is the main organ of mercury accumulation. This relationship is not only reserved for piscivorous fish; this fact has also been reported in studies on common carp (Cyprinus carpio) – Maršálek et al. (2007). Carassius carassius and Tinca tinca in the present study also belong to Cyprinidae family, and in their livers, the Hg content was also lower than in their muscles, by approximately 82% and 73% (based on the median value), respectively (Table 2). The mercury content of perch liver (based on the median value) was about 36% lower than its content in muscle tissue (Table 2).

 $\label{thm:thm:thm:concentration} Table~2$  Mercury concentration in selected tissues of piscivore and benthic fish in mg kg  $^{-1}$  wet weight

Species	Organs	Mean	SD	Median	Min.	Max.	$\mathrm{Q}_{25}$	$Q_{75}$
Perca fluviatilis	muscle	0.0805A	0.0318	0.0696	0.0474	0.1400	0.0574	0.0985
	liver	0.0459A	0.0273	0.0443	0.0154	0.0951	0.0175	0.0601
	scales	0.0053A	0.0038	0.0033	0.0014	0.0114	0.0024	0.0091
Carassius carassius	muscle	0.0356B	0.0084	0.0369	0.0259	0.0465	0.0265	0.0449
	liver	0.0068B	0.0016	0.0065	0.0050	0.0097	0.0053	0.0078
	scales	0.0002B	0.0001	0.0001	0.0001	0.0003	0.0001	0.0002
Tinca tinca	muscle	0.0249B	0.0077	0.0238	0.0151	0.0353	0.0189	0.0316
	liver	0.0065B	0.0009	0.0064	0.0050	0.0075	0.0058	0.0074
	scales	0.0003B	0.0003	0.0002	0.0000	0.0007	0.0001	0.0005

 $Q_{25}$  – lower quartile,  $Q_{75}$  – upper quartile, SD – standard deviation, Min. – minimum, Max. – maximum, A, B, – statistically significant differences at  $p \le 0.01$ 

A high correlation between mercury content in muscle and that in the liver (0.83) and scales (0.75), and in relation to body mass (0.76) was found (Table 3).

Łuczyńska et al. (2016) also showed that the concentration of total mercury in muscles was positively correlated with the body mass of perch. In our study, a high correlation in the crucian carp was noted only in the mercury content between muscles and the liver (0.87) (Table 4).

In turn, in tench, a high correlation between the mercury content in muscles and scales (0.83) was noted (Table 5). Furthermore, a strong correlation was found between the body mass of tench and the mercury content in muscles (0.95) and scales (0.79).

Table 3 Correlations between body mass and mercury concentration in organs of the *Perca fluviatilis* 

Perca fluviatilis	Muscle	Liver	Scales
Liver	0.8303*	-	
Scales	0.7454*	0.7818*	-
Body mass	0.7575*	0.6242	0.3575

<sup>\*</sup> statistically significant differences at *p*≤0.05

Carassius carassius	Muscle	Liver	Scales
Liver	0.8666*	-	
Scales	0.2392	0.4110	-
Body mass	0.2969	0.5878	0.2760

<sup>\*</sup> statistically significant differences at *p*≤0.05

Tinca tinca	Muscle	Liver	Scales
Liver	0.3809	-	
Scales	0.8333*	0.3095	-
Body mass	0.9523*	0.4285	0.7857*

<sup>\*</sup> statistically significant differences at  $p \le 0.05$ 

The high correlations obtained indicate significant dependencies in the distribution of mercury in fish organisms, to a greater extent in piscivorous (perch) than in the studied benthic fish (tench and crucian carp).

The problem of Hg residuals in the food of animal origin, especially fish, is particularly important due to the increase in aquaculture production in

recent decades. Global fisheries and aquaculture production reached a record 223.2 million tonnes in 2022, according to the FAO's latest SOFIA report (FAO, 2024). A 4.4% increase from the year 2020 was noted. For the first time, aquaculture production (130.9 million tonnes) surpassed capture fisheries, accounting for 51% of aquatic animal output. Poland contains the largest area of ground ponds among European Union countries. In 2023, freshwater fish catches in Poland amounted to 59.5 thousand tonnes, 1.6 kg per capita. The average monthly consumption of fish and seafood per person in households in Poland in 2023 was 0.22 kg (Statistical Yearbook of Agriculture, 2024). According to the FAO report (2022), regarding global aquaculture production, it is estimated that aquatic animal production will increase by another 14% by 2030. In accordance with Resolution No. 2/2024 of the Management Committee of the Fish Promotion Fund of 5 June 2024 on the adoption of a promotion strategy for the fish industry for 2025, it is recommended to increase the consumption of fish bred by Polish aquaculture. Fish are a source of polyunsaturated fatty acids, vitamins A, D, and B, and minerals such as iodine, phosphorus, selenium, magnesium, and potassium (Khalili Tilami, Sampels, 2017). On the other hand, it is necessary to supervise and monitor the levels of substances hazardous to health (e.g., Hg) accumulated by fish (meat - muscles) for human consumption (de Almeida Rodrigues et al. 2019, Skibniewski et al. 2025). Implementation of monitoring programs for freshwater fish may require the sacrifice of large numbers of fish each year. In addition to skeletal muscles, the gills, liver, kidneys, and olfactory epithelium are also used to study mercury content. However, these are still invasive methods that end in the death of the fish. Sampling such as blood drops, tissue biopsy, or fin samples can limit the number of animals used (Baker et al. 2004, Červenka et al. 2011, Debska et al. 2023). Among the tissues mentioned above, a sample of scales seems to be the simplest choice, which does not require special equipment or procedure. Moreover, it does not significantly burden the fish's body and does not leave a large wound that would be a gateway for microorganisms. It has been suggested that sampling by scale is probably a less harmful technique than taking biopsy plugs, especially for small fish (Cervenka et al. 2011). In addition, it is possible to use archived fish scales as markers of past contamination (Jovičić et al. 2023), and rare or threatened species can be sampled without causing mortality (Baker et al. 2004). Moreover, the growth rate of fish could be calculated from the scales (Lampart-Kałużniacka, Heese 2017). However, not all scales are the same, and knowledge on how to collect and analyze them is needed. Most modern fish species have elastic scales, which take the form of flexible plates deeply embedded in the skin. There are two types of scales of this type: cycloid scales with smooth and rounded edges of the visible part (ring-like scales) that occur, for example, in carps (Carassius carassius) and (Tinca tinca), and ctenoid scales whose surface is serrated (comb-like scales) and which occur, for example, in perches (Perca fluviatilis) - Rawat et al. (2021). Cervenka et al. (2011) reported that the method of sampling scales is unsuitable for fish species with small scales, such as brown trout.

Luczyńska and Krupowski (2009) showed that the mercury content in the skeletal muscles of freshwater fish is more than twice as high as in marine fish. Other authors showed that the concentration of this element in the liver in both cases was similar, while the content in the gills was higher in marine fish (Langauer-Lewowicka, Pawlas 2017). For those reasons, in this study, the Hg concentration in muscle and liver in freshwater fish was determined, using scales as a non-lethal method of sampling. Moreover, the liver was selected for analysis because it is a good indicator of environmental pollution, has the ability to accumulate pollutants, and also plays an important role in their storage, redistribution, detoxification, and transformation (Havelková et al. 2008), while the choice of muscles was dictated by the fact that they are a valuable source of protein in our diet.

Assessment of biomagnification of mercury in fish is a valuable indicator of contamination with this element of the aquatic environment, and it ensures the safety of consumers of fish occupying different trophic levels or other aquatic animals. Based on the results obtained, it was confirmed that muscles and the liver are good indicators of environmental pollution with mercury, but scales turned out to be an equally interesting indicator, although their Hg values were much lower. In perch scales, the Hg value was 20-fold reduced relative to the content in muscles and 13-fold relative to the content in liver (based on median values). In turn, in the crucian carp, this value was 300-fold smaller in comparison to muscles and 54-fold compared to the liver, and in tench, 113-fold and 30-fold (based on median values), respectively. It follows from the above that scales may constitute a valuable material for Hg detection in the body, mainly for fish from higher levels in the food chain, because in non-piscivorous fish their content is drastically decreasing compared to other, typically analyzed tissues, like muscle and the liver (Červenka et al. 2011).

It should be emphasized that the mercury content in perch scales was 26-fold higher than in crucian carp scales, and 17-fold higher than in scales of tench (Table 2), based on the mean values. These results confirm the phenomenon of bioaccumulation and biomagnification of mercury in the trophic chain. Moreover, it indicates that the studied fish samples do not pose a risk to human health in accordance with the current limits of mercury content in fish meat (Commission Regulation (EU) 2023/915). Each sample of muscle of the studied fish species accumulated mercury at a level of 0.0805, 0.0356, and 0.0249 mg kg<sup>-1</sup> in perch, common carp, and tench, respectively, i.e., the percentage reduction was almost 56 and 69 in crucian carp and tench in relation to the muscle of perch. Łuczyńska et al. (2016) showed a higher mercury content in the muscles of perch, i.e., 0.173 mg kg<sup>-1</sup> wet weight, but in their study, the fish were heavier and their weight was in the range of 159.8-559.5 g. Červenka et al. (2011) reported even higher mercury con-

tents in the perch muscle: from ~3 to ~7 mg kg¹ and for scales: from ~0.05 to ~0.15 mg kg¹, but the weight, length, or age of the perch caught was not given. More recent studies showed the content of total mercury in the tissues decreased as follows: muscle>liver>gonads>scales in carp, pike, zander, and catfish from Czech ponds, and these results are consistent with our results. The cited authors concluded that the highest content of THg was observed in the muscle of  $Esox\ lucius\ (0.1517\pm0.0176\ mg\ kg¹)$ , the lowest content was determined in samples of scales of  $Cyprinus\ carpio\ (0.0010\pm0.0001\ mg\ kg¹)$  – Pavla et al. (2022). Similar observation was reported in the present study, but for perch, which is a piscivore, and for  $Carassius\ carassius\ and\ Tinca\ tinca\ which, like\ Cyprinus\ carpio\ belong to\ Cyprinidae.$ 

#### CONCLUSIONS

The accumulation of mercury in fish organisms is related to their species and feeding behaviour. Higher mercury content in fish tissues is noted in species located higher in the trophic chain (perch). The mercury content in fish tissues examined in the present study can be arranged, from highest to lowest, in the following order: muscles > liver > scales. Predicting Hg concentrations in the muscle tissue, especially of perch, which is a piscivore, from measured Hg concentrations in their scales may be useful for assessing Hg contamination in fish muscle as a form of screening. The mercury content in fish muscles did not exceed the limits for mercury content specified in legal regulations (Commission Regulation (EU) 2023/915), i.e., up to 0.50 mg kg<sup>-1</sup>.

#### **Author contributions**

IL, MS – conceptualization, IL, ID, MK – methodology, IL, BS, MS, ID, ES, MK – investigation, IL, BS, MS, ES – writing original draft preparation, MS – data curation, MS, BS – visualization, MS, ES – supervision, MS – writing review & editing. All authors have read and agreed to the published version of the manuscript.

#### Conflicts of interest

The authors declare no conflict of interest.

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