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Original Paper

Ichtyocenoses of the Slaná River Polluted by the Mine Water

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In February 2022, the long-term pollution of the Slaná River by discharge of mine water began. The pollution is evident in a long stretch of the river, due to the bright orange colour of the water. According to media reports, there was significant damage ichthyocenoses, despite the fact that no mass death of fish was recorded. About a month before the survey was carried out, the concentration of dissolved substances was reduced by approximately 80%. The goal of the ichthyological survey was to determine the current state of ichthyocenoses affected by pollution. The presence, abundance and age representation of individual species of fish and roundmouths at three locations were examined. A decrease in species diversity, low densities in relation to the character of the flow and a distorted age structure in the affected locations were found. The condition of ichthyocenoses improved in direct proportion to the distance from the point of discharge of mine waters. When comparing results of current ichthyological survey with previous surveys and based on the absence of other significant sources that could negatively impact river ecosystems, it is possible to consider this pollution of the Slaná River to be the reason for degradation of ichthyocenoses.

Keywords: ichthyological survey, ichthyocenoses, water pollution, Slaná River, mine water

1 Introduction

The Slaná River is located in the south of the Slovakia and runs through area surrounded by hills that are relatively rich in minerals (Figure 1). As a result, this area was extensively used for mining in the past. Water that occurs naturally in active mining areas is usually pumped up to the surface and treated before discharging it into the recipient. However, most of the mines were used up and are nowadays closed. As a result, water is flooding the former mining tunnels and flushing out materials that were left behind in the abandoned mines. The untreated water from former mining area for iron started polluting the Slaná River in February 2022 (TASR, 2022c).

The ichthyological survey took place in the section of the Slaná river defined as the water body SKS0002, classified as type K2S, with a natural flow character, and water management significance (Ministry of environment of the Slovak republic, 2022). Type K2S corresponds to medium-sized rivers at elevations of 200-500 meters in the Carpathian Mountains (Makovinská et al., 2015).

Water pollution represents a deterioration of quality caused directly or indirectly by human activities (Tóth et al., 2020). Deterioration in water quality due to human activities is a significant phenomenon of the last century, which has fundamentally affected the quality of aquatic life. Changes of water quality when moving down the river continuum are expected. However, these changes are usually process influenced by natural causes typical for river ecosystems, such as erosion of the riverbed and banks, runoff from watersheds, leaf litter, and the decomposition of plants and animals within. These natural changes of water quality in longitudinal river continuum are sometimes called "natural pollution". On the other hand, pollution caused by human activity (also called anthropogenic) is a phenomenon that is causing more significant problems. Depending on how pollution enters the water, sources of pollution can be classified as point source (i.e. direct discharge of waste waters from single point) and diffuse source (flush out coming from areas, such as nutrients from land use). In terms of its temporal course, pollution can be chronic (long-term, continuous) or acute (sudden). Pollution can also be categorized by its

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origin – industrial, municipal (sewage), and agricultural. The toxic effects of pollution are distinguished based on their impact and can be either acute or chronic. Chronic effects involve the negative consequences of long-term exposure to sublethal (i.e., not causing sudden death) concentrations of toxic substances on the physiology. Such exposure of fish can have negative impacts on their reproduction, growth, and can negatively affect all other aspects of their life cycle (Adámek, 2013).

Based on these criteria, pollution of the Slaná River with contaminated mine waters can be classified as anthropogenic, industrial, point source of pollution with sub lethal effects.

The pollution of the Slaná River by mine waters was widely publicized, with information on the significantly negative impact of the pollution on the fish population in the river, despite the fact that no mass mortality of fish was observed (e.g., TASR, 2022d, TASR, 2022b). In June 2022, the amount of discharged waters was reduced, but it was not completely stopped up to this day (TASR, 2022a).

In the surveyed section of the Slaná River, several ichthyological surveys were conducted in the past to assess the ecological status of the waters (Mišíková Elexová et al., 2015, Kováč, 2021). According to the results of these surveys, the following fish species were found in this section of the river: *Eudontomyzon danfordi* (Linnaeus 1758), *Alburnoides bipunctatus* (Bloch 1782), *Barbatula barbatula* (Linnaeus 1758), *Barbus barbus* (Linnaeus 1758), *Barbus peloponnesius* (Kotlík, Tsigenopoulos, Ráb & Berrebi, 2002), *Cottus gobio* (Linnaeus 1758), *Gobio gobio* (Linnaeus 1758), *Leuciscus cephalus* (Linnaeus 1758), *Oncorhynchus mykiss* (Walabaum 1792), *Phoxinus phoxinus* (Linnaeus 1758), *Salmo trutta* m. *fario* (Linnaeus 1758). These surveys were conducted using the same methodology as our ichthyological survey in 2022 and were located at the same site as our site number 1, respectively upstream from Rožňava, approximately

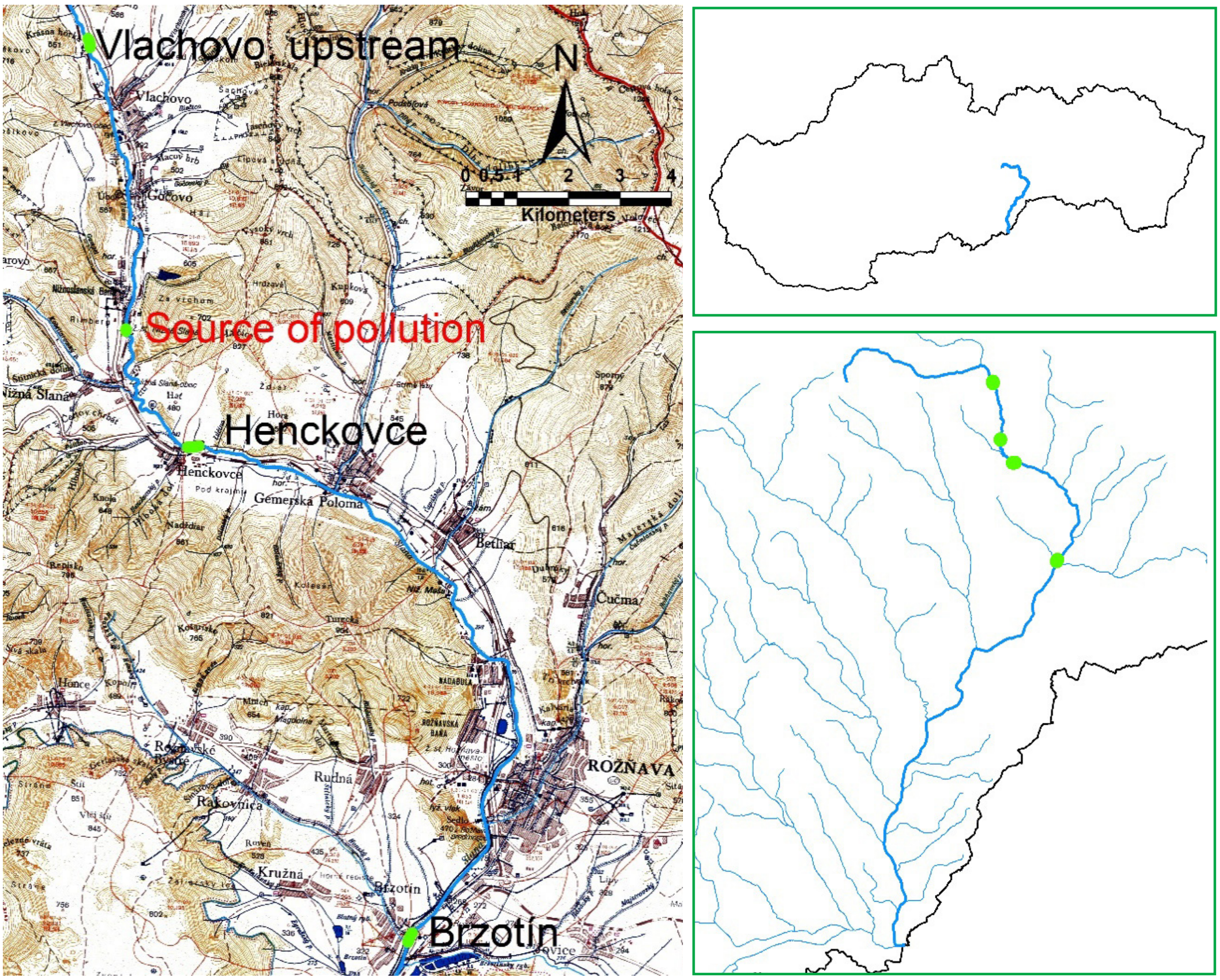

Figure 1 Location of sampling sites at Slaná River (Slovakia)

5 river kilometres higher from site 1. Several surveys were also conducted by various organizations. While their results were not published in the scientific literature, they were partially published in the daily press (Vargová, 2022) and confirmed through personal contact with the responsible, or representatives of the local organization of the Slovak angling association in Rožňava, which holds fishing rights in this section of the River. All information indicated a significant reduction to almost complete disappearance of fish in terms of their numbers and biomass due to pollution over tens of kilometres.

The ichthyological survey was carried out to assess the status of ichthyocenoses affected by the pollution of the Slaná River due to the discharge of polluted waters from the facility of the Rudné bane š.p. company.

2 Material and methods

The ichthyological survey was conducted following the Standard Operating Procedure of the Department of Hydrobiology and Microbiology NRL/HB-ŠOP/8 Ichthyological Survey and STN EN 14011 Water quality: Sampling of fish with electricity with consideration for the approved methodology for assessing the ecological status of waters based on fish (Kováč, 2010), which was also intercalibrated at the European level. Fish sampling was carried out using two certified electrofishing devices. The sampling team consisted of 8 members. The electrofishing operators worked simultaneously to cover the entire width of the stream. The captured fish were kept in mesh containers placed directly in the stream, out of the reach of the electric field.

After sampling, individual fish specimens were identified to the species level based on their morphological characteristics, and their total length (TL) was measured with an accuracy of the nearest five millimetres. After measuring a minimum of 150 individuals of one species, representing all size categories present in the sample, the remaining individuals belonging to the species were counted and recorded.

The ichthyological survey was conducted at three pre-selected sites with respect to source of pollution. The Brzotín and Henckovce (Figure 1, Table 1) sites were located below the source of pollution and represented sites affected by different intensity of pollution. Vlachovo site (Figure 1, Table 1) was located above the source of pollution and represented state of the River that is not affected by any pollution.

The obtained results were processed for each individual sampling site. The species composition, size spectrum of captured fish, abundance and domination were assessed (Losos, 1985). Fish were categorized into age groups of 0+ (individuals younger than 1 year) and 1+ (individuals older than one year) based on their size spectrum. The data were analysed using the statistical program Rstudio (2021) with the use of a comprehensive tool for wide-ranging data evaluation called tidyverse (Wickham et al., 2019). Size groups of captured individuals were created for all species from which more than one specimen was caught. Abundance is expressed in tables as the number of individuals captured per 100 square meters (Losos, 1985). The fishing effort is calculated per unit length of a section of 100 meters as CPUE.100 m^{-1} (catch per unit effort per 100 m), (Gandy & Rehage, 2017).

3 Results and discussion

3.1 Sampling site: Brzotín

Sampling site is situated approximately 18 km below the source of pollution and is located upstream of the confluence of Honský creek with the Slaná River. The riverbed at the sampling site has a natural character with a suitable diversity of habitats for ichthyofauna. Turbidity caused by pollution at the time of the survey reached elevated levels but did not have significant impacts on the execution of the survey. The riverbanks at the sampling site were reinforced with rubble stones. The reinforcement is occasionally breached, allowing the flow to create more diverse conditions for

Table 1 Summary information about sampling sites and the source of pollution

Site number	Site	GPS coordinates	Wetted width of the river (m)	Pure catching time (min)	Sampled section of the river (m)	Sampling area (m^2)	River kilometre
1	Brzotín	48.631085 20.401316	10–12	35	115	1,380	49.0
2	Henckovce	48.714836 20.436205	8–10	30	160	1,600	63.7
0	source of pollution	48.734924 20.416142	–	–	–	–	67.4
3	Vlachovo, upstream	48.783493 20.401732	8–10	38	200	1,800	73.1

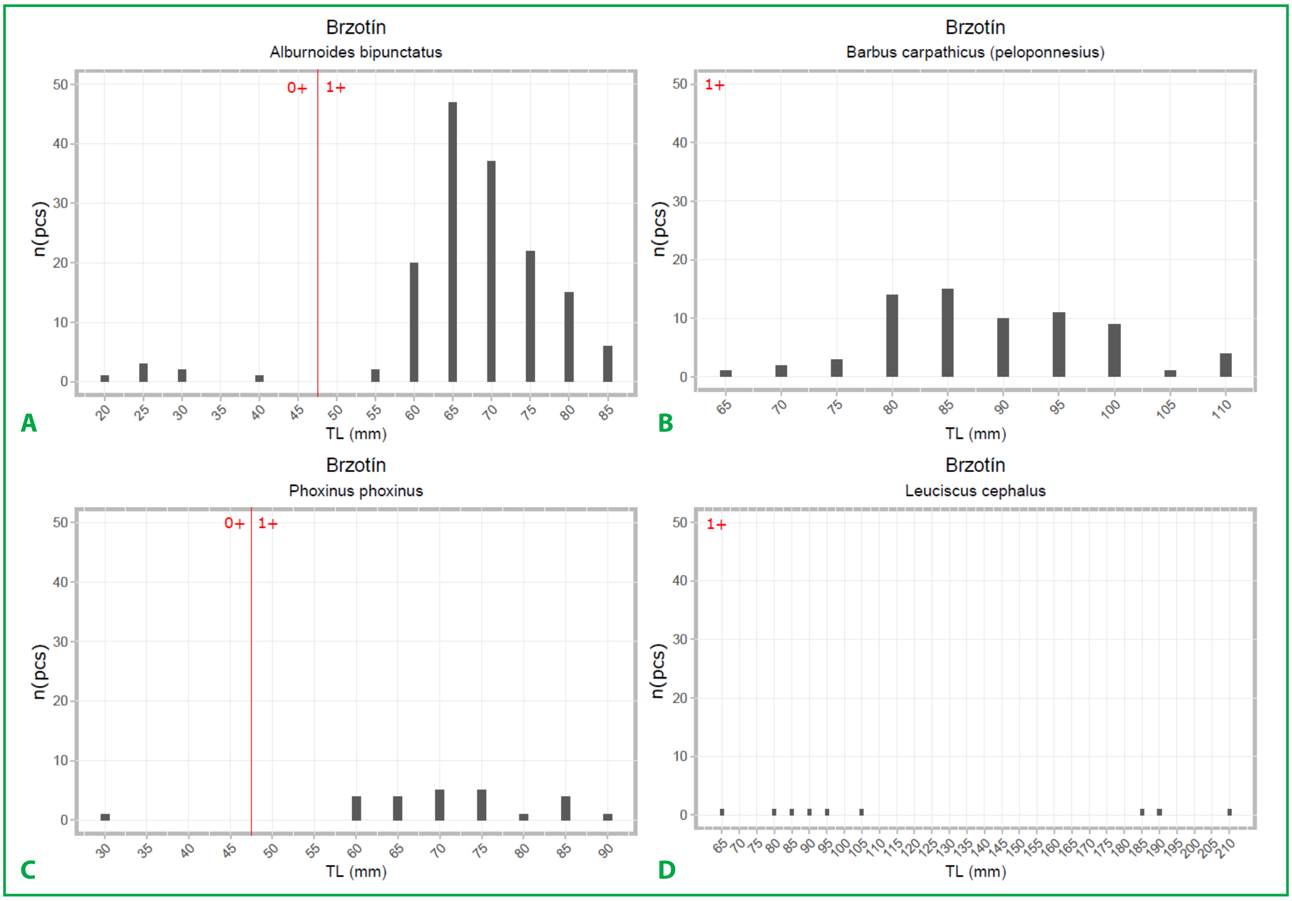

Figure 2 Size frequency of fish species at Brzotín location
A – *Alburnoides bipunctatus*, B – *Barbus carpathicus* (*peloponnesius*), C – *Phoxinus phoxinus*, D – *Leuciscus cephalus*

ichthyofauna. The wetted width of the river at the location averaged from 10 to 12 meters. Signs of pollution were still identifiable at the site.

In terms of species composition, the spirlin (*Alburnoides bipunctatus*) significantly dominated. The second most abundant species was the carpathian barbel (*Barbus carpathicus*), sometimes referred to by ichthyologists as *Barbus peloponnesius*. Finally, the third most abundant species was the eurasian minnow (*Phoxinus phoxinus*; for more details see Table 2).

In comparison to the 2011 survey (Mišíková Elexová et al., 2015), the lamprey (*Eudontomyzon danfordi*) and the european bullhead (*Cottus gobio*) were no longer found at the site. In comparison to the 2015 survey (Kováč, 2021), the lamprey species (*Eudontomyzon* sp.) and the common barbel (*Barbus barbus*) were not found at the site, but the carpathian barbel (*Barbus carpathicus*), eurasian minnow (*Phoxinus phoxinus*), chub (*Leuciscus cephalus*), and gudgeon (*Gobio gobio*) were identified. Species that were not caught recently but were captured in the surveys from 2011 and 2015 are characteristic

Table 2 Fish species at the Brzotín location

Species	Abundance	Domination (%)	pcs.100 m^{-2}	CPUE.100 m^{-1}
Alburnoides bipunctatus	273	70.91	19.78	237.39
Barbus carpathicus (*peloponnesius*)	70	18.18	5.07	60.86
Phoxinus phoxinus	30	7.79	2.17	26.08
Leuciscus cephalus	9	2.34	0.65	7.82
Salmo trutta m. *fario*	1	0.26	0.07	0.86
Gobio gobio	1	0.26	0.07	0.86
Barbatula barbatula	1	0.26	0.07	0.86
Summary	385	100.00		

by living close to the riverbed or buried in sediments. These types of habitats are expected to be the most influenced by pollution, which could have contributed to the destruction of populations due to sediment coverage of the riverbed with precipitated iron deposits.

Based on the size frequency of the captured species (Figure 2), as well as their abundance and fishing effort (Table 2), there is a reasonable assumption that the individuals found at the time of sampling do not represent a permanent population. Given the recent decrease in pollution, it is possible to conclude that at this stage in the river, there is partial recolonization and occupation of free space by fish species that inhabited the location or withdrew to nearby tributaries before the pollution. This can be especially of importance because there are two significant tributaries as well as several smaller local tributaries that could provide refuge.

To conclude, the ichthyocenoses at this location cannot be considered stable, as the sample largely lacks larger, adult individuals, and the representation of other fish species is more limited. Destabilization can also be observed in terms of the fishing effort and the number of captured individuals. The natural recovery of the population at this moment cannot be confirmed. This is mostly due to multiple barriers on the Slaná River that are considered impassable for fish. Therefore, potential population recovery only through the migration of individuals from lower sections (with lower pollution intensity) upstream to occupy the free space can be a significant problem. Moreover, the surrounding smaller tributaries may not have the flow ratios or habitat possibilities to ensure the survival of larger fish species and individuals in sufficient numbers.

3.2 Sampling site: Henckovce

Sampling site is situated approximately 3.7 km below the source of pollution, therefore representing site that is potentially the most affected. The riverbed at the sampling site is characterized by areas with deposits of quarry stones. In most of its section, riverbank exhibits a natural character. At the time of sampling, there was a significant increase in turbidity caused by pollution, visually observed as an orange colour. Sampling

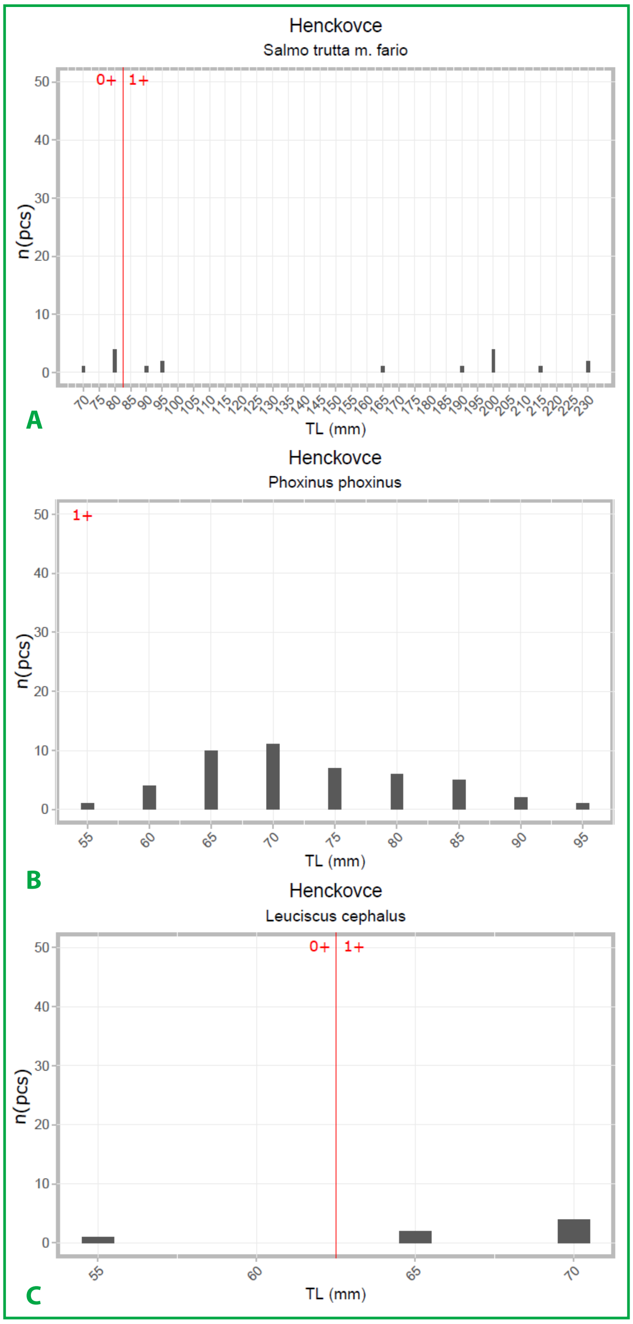

Figure 3 Size frequency of fish species at Henckovce location
A – *Salmo trutta* m. *fario*, B – *Phoxinus phoxinus*, C – *Leuciscus cephalus*

Table 3 Number and representation of caught fish species at the Henckovce location

Species	Abundance	Domination (%)	pcs.100 m^{-2}	CPUE.100 m^{-1}
Phoxinus phoxinus	51	63.75	3.18	31.87
Salmo trutta m. *fario*	22	27.50	1.37	13.75
Leuciscus cephalus	7	8.75	0.43	4.37
Summary	80	100.00		

conditions were challenging in this regard, and sampling was therefore conducted with great caution due to low visibility conditions to minimize escaping of fish. The shores at the sampling site are reinforced with quarry stones, while area upstream from sampling site is without noticeable reinforcement. A species selective barrier – a stabilizing weir was identified above the sampling area. The width of the water in the location ranged from 8 to 10 meters. Strong signs of pollution are visible at the location – pronounced orange colouring on both the banks and the bottom, as well as persistent turbidity due to the proximity of the pollution source.

The species composition at this location was significantly less diverse despite the fact that this site is similar to sampling site 1 based on observed abiotic parameters (Table 3). Differences in the representation of the number of individuals are lower in this case, but considering the greater fishing effort, the density of fish on this site can be considered very low. In this case as well, the ichthyocenoses cannot be considered stable due to the low number of recorded individuals in relation to the fishing effort and the significant absence of natural representation in several age and size categories (Figure 3). Low numbers of fish are therefore most likely caused by stronger effect of pollution due to the shorter distance from the source of pollution and the absence of more substantial tributaries in the vicinity that could serve as refuges.

3.3 Sampling site: Vlachovo, upstream

Sampling site is situated approximately 5.7 km above the source of pollution. Both the stream and its banks exhibit a natural character with significant variations between shallow sections and deep pools, creating diverse habitats for the ichthyofauna. However, the stream at this location differs in type from the previous two sites. The stony substrate is deposited within a gravel-sand bed, with the absence of larger stones, and fine sediment is freely deposited at the edges of the banks, which are without any reinforcement. Turbidity at the time of survey was nearly non-existent, with high water clarity even in deep areas. The width of the stream at the sampling site ranged from 8 to 10 meters.

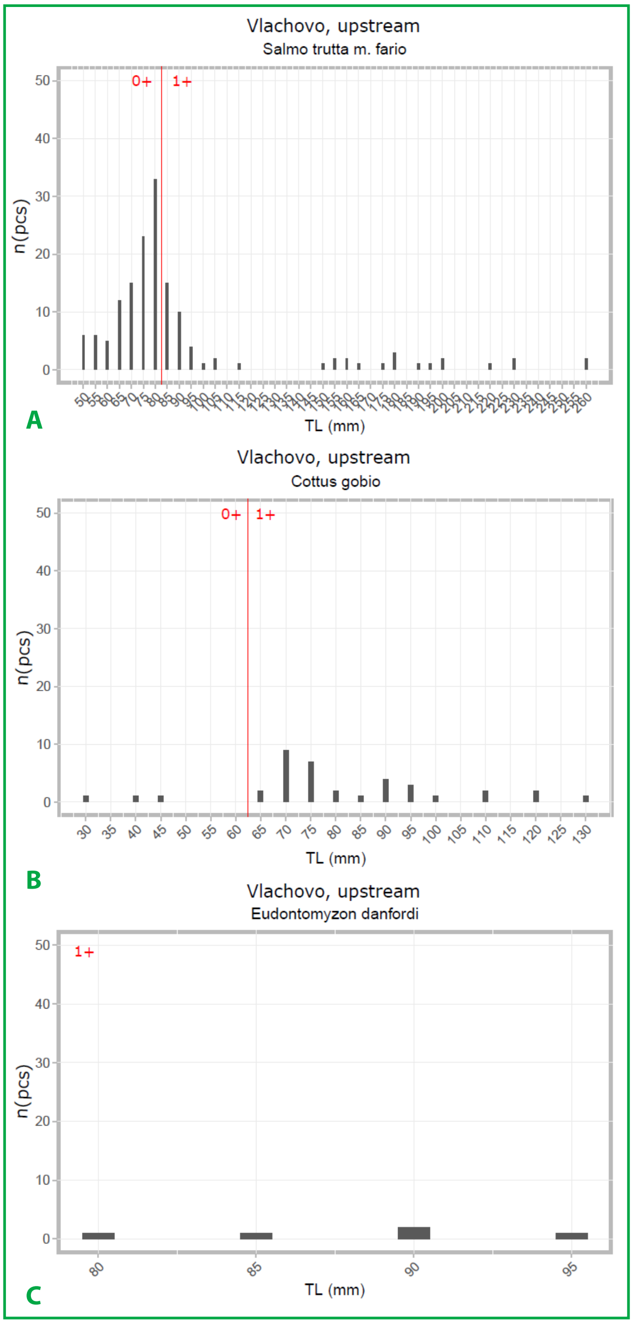

Figure 4 Size frequency of fish species at Vlachovo, upstream location
A – *Salmo trutta* m. *fario*, B – *Cottus gobio*, C – *Eudontomyzon danfordi*

Table 4 Number and representation of caught fish species at the Vlachovo, upstream location

Species	Abundance	Domination (%)	pcs.100 m^{-2}	CPUE.100 m^{-1}
Salmo trutta m. *fario*	299	84.94	16.61	149.5
Cottus gobio	48	13.64	2.66	24.0
Eudontomyzon danfordi	5	1.42	0.27	2.5
Summary	352	100.00		

In terms of ichthyofauna, three fish species were found in the surveyed section (Table 4). The lower species diversity in this case corresponds to the changing nature of the river, but it is also influenced by the predation pressure of the significantly dominant brown trout. The studied location is at a higher elevation (approximately 150 meters above sea level than location Brzotín). At this particular location, a healthy and self-sustaining population of the identified species can be assumed based on their mobility and the presence of both juvenile and adult individuals of brown trout (*Salmo trutta* m. *fario*) and european bullhead (*Cottus gobio*; Figure 4). Five juvenile individuals of Carpathian lamprey were also recorded (Figure 4). These individuals were in the process of metamorphosis, which occurs in this species at the age of 4–5 years (Dungel & Řehák 2005).

4 Conclusions

Based on the results of the ichthyological survey, it can be concluded that fish populations in the affected sections of the Slaná River show significant signs of destabilization and are in poor condition. In the affected locations, there is a lack of diverse representation of size groups representing various age categories, as well as a less abundant representation of species compared to what can be observed at site above pollution. Furthermore, in the affected locations, adult individuals are significantly absent. Species found in the sampling site that was most distant from the source of pollution are most likely recolonizing free habitats from the tributaries of the Slaná River. This might be due to pollution effect falling off caused by dilution in longitudinal spectrum of the river. These tributaries could have served as refuges for the ichthyofauna, which is a relatively mobile component of aquatic ecosystems, depending on the species. However, it is questionable whether the habitat conditions in the Slaná River tributaries were able to provide refuge for larger species or adult fish in adequate numbers that would be enough for restoring populations on its own. Moreover, the presence of numerous migration barriers on the Slaná River may hinder the recolonization of river sections by fish from lower, possibly unaffected sections.

Considering that no negative factors that could lead to the deterioration of the ichthyocenoses in the affected locations (Brzotín, Henckovce) were recorded before pollution, and given the good condition of the unaffected location (Vlachovo, upstream), the current state can be highly likely caused by contamination from discharging polluted mining waters.

Based on the findings, it is evident that ichthyological monitoring is needed to track population developments over time. Monitoring should be carried out on multiple sections of the river with varying pollution intensities to obtain more comprehensive results about the state of the ichthyofauna. Through a comprehensive evaluation of long-term ichthyological monitoring, it will be more likely to assess in the future whether the populations in the river are capable of recovery. Monitoring the state of ichthyocenoses is considered important, especially considering that several of the identified species are subject of protection.

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