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Investigation of the hyporheic zone of two gravel-bed rivers after reservoir draining

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With 5 figures and 3 tables

Abstract: Dams and reservoirs are a common and globally widespread anthropogenic disturbance with documented negative effects on riverine and riparian habitats. The two most well-known impacts of river damming are longitudinal fragmentation of surface running waters and a shift from lotic habitats towards habitats with lentic characteristics that affect the benthic and pelagic communities. However, there is very little empirical evidence about the effects of damming on the aquatic fauna inhabiting interstitial habitats extending in and alongside the river-bed (i.e., hyporheic zone). In this study, we investigated the patterns in the interstitial community composition upstream, downstream and within the reservoir that was formed 80 years ago, when the river was dammed for the hydropower production. We used the rare opportunity to directly access the bottom of the reservoir drained due to dam maintenance in January 2018, to compare physical, chemical and faunistic data from the reservoir area, with those from downstream and upstream reaches of the two gravel bed rivers that are flowing into the reservoir. We sampled the interstitial invertebrate communities at seven locations, using a Bou-Rouch pump at two depths (30–60 cm and 60–90 cm within the river bed) and at three sampling points within each location. At the same sampling points we measured also physical and chemical parameters (temperature, conductivity, oxygen and pH). The interstitial water from the deepest point of the drained reservoir had substantially lower oxygen concentration, lower pH, and higher conductivity than water from the other sampling localities. This was also the site where taxa richness was lowest, and only one obligate groundwater species (i.e. stygobiont) was found. Most probably, the changes in morphology of the river channel and speed of water flow due to damming, which increased sedimentation rate and clogging of interstitial habitat, resulted in such large differences in environmental conditions and invertebrate community composition. This study provides rare empirical evidence of the effects of damming on the river interstitial habitats and fauna within the reservoir area. We recommend that environmental impact assessments conducted prior dam constructions should include also assessment of the effect of river damming on the interstitial communities. These organisms are playing important role in driving important ecosystem processes, such as organic matter degradation on one hand, and on the other hand, are composed of many rare and endangered species that need to be protected.

Keywords: interstitial habitats; subterranean fauna; stygobionts; dam; hydropower

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Introduction

Dams are man-made barriers on streams and rivers built for a variety of purposes, including power generation, flood control, water storage, and/or recreation. It has been estimated that dams impact approximately 48 % of global river water volume (Grill et al. 2015) and that reservoirs contribute to a 7.3 % increase in the Earth's naturally occurring surface freshwater (Lehner et al. 2011). The effects of dams extend beyond the reservoir and can be demonstrated both upstream and downstream (Ligon et al. 1995; Richter et al. 2010) in aquatic and riparian biotopes (Poff & Zimmerman 2010). Dams alter habitat characteristics by fragmenting the river continuum and severely modifying river flow, resulting in lower peak flows, changed flooding regimes, reduced inundation, and reduced transport of particles (Ligon et al. 1995; Poff et al. 1997; Nilsson et al. 2005; Brenna et al. 2021). Reservoirs transform river systems into habitats that have lost their lotic (i.e. flowing) character, while gaining partially or completely lentic (i.e. stagnant) functions (Schmutz & Moog 2018). Within the reservoirs, a substantial sediment deposition is taking place, trapping mostly sand and silt (Ansellmeti el al. 2007). These habitat changes have negative consequences for aquatic life, both in surface as well as in interstitial habitats, including loss of species richness, loss of endemic or specialized species, alteration of species communities, disrupted or blocked migration routes, and homogenization of species richness across continental scales (Poff et al. 2007; Poff & Zimmerman 2010). Restoration of ecological functions of rivers has also been among reasons for increasing trend of dam removal in North America and Europe in the last decades (Habel et al. 2020).

Dams and reservoirs affect all four dimensions of the lotic ecosystems: longitudinal, lateral, vertical and temporal (Ward 1989), but the effects in the vertical dimension are the least studied. These include the connection between the river bottom and the underlying groundwater, and the effects on organisms living in the interstitial habitats, i.e. the water-filled spaces between particles of unconsolidated river sediment. Surface water communicates with groundwater through the hyporheic zone – an ecotone between the river bottom and the underlying groundwater (Orghidan 1959; Boulton et al. 1998; Krause et al. 2022). Due to the absence of light, primary production in groundwater is extremely limited, and nutrient inputs are almost entirely dependent on surface habitats (Gibert et al. 1994; Foulquier et al. 2010; Culver & Pipan 2019). Therefore, the condition of groundwater, its inhabitants and its ecosystem services depend heavily on surface aquatic habitats and the intact connection of surface water to groundwater (Griebler et al. 2014; Griebler & Avramov 2015).

In reservoirs, standing water and sedimentation lead to the formation of thick lacustrine sediments deposited over gravel beds on the bottom. This eventually disrupts the vertical continuum between surface flow and groundwater (Schmutz & Moog 2018). In such conditions, the fine sediments clog the interstitial spaces in a process called colmation (Brunke 1999). The colmated riverbed is characterized by low porosity and reduced hydraulic connectivity and can even lead to a drop in groundwater level (Brunke 1999). Such river reaches no longer function as fish nurseries and lack refugial habitats for aquatic insects. Studies examining the effects of colmation on benthic and interstitial fauna have found reductions in taxonomic richness and species density of 50 % and 30 %, respectively (e.g. Descloux et al. 2013). The communities are dominated by species that can cope with smaller pore size and reduced oxygen concentration, such as oligochaetes and nematodes (Descloux et al. 2014).

Colmation in reservoirs negatively impacts also species that live exclusively in interstitial habitats, so-called stygobionts (Claret et al. 1999). Stygobionts are highly endemic (Malard et al. 2009; Trontelj et al. 2009; Zagmajster et al. 2014), sensitive to changes in abiotic conditions (Dole-Olivier et al. 2009; Mori et al. 2012) and have low dispersal potential (Asmyhr et al. 2014). Colmation changes the natural interstitial habitat by clogging the pores and consequently lowers the oxygen concentration. Major changes in hyporheic and groundwater communities, or complete loss of stygobionts, can have a severe negative effect on river and groundwater ecosystem services, including processes of water self-purification (Griebler & Avramov 2015).

Empirical evidence of the effects of reservoir formation on interstitial invertebrate communities is scarce. Investigation of the effects of such river modifications on interstitial fauna is hardly possible when reservoirs are filled with water, as the bottom of reservoirs is not accessible for sampling. The bottom of the reservoir can be reached only when the water is drained out, which is a rare event related to the maintenance of the reservoirs and dams (Kondolf et al. 2014). Such a rare opportunity arose in the reservoir of the Soča River (Western Slovenia, Europe) in 2018 due to maintenance works on the hydropower plant downstream. Emptying of the reservoir due to the opening of the dam had allowed access to interstitial habitats within the reservoir. We performed a comparison of taxonomic diversity, community composition and spatial distribution of interstitial invertebrates within, upstream and downstream from the drained reservoir. Since it is known that sedimentation processes alter environmental conditions in the interstitial habitats (Descloux et al. 2013), we were able to test this by comparing abiotic parameters of the reservoir with upstream and downstream localities. Due to thick sediment layer accumulating over decades within the reservoir area and consequent colmation of the interstitial spaces, we expected impeded hydrologic connectivity within the reservoir, and consequently higher conductivity, and lower oxygen contents as well as lower pH values as compared to other localities. Such conditions are typical for hyporheic zones with increased residence times of water due to smaller hydraulic conductivity (Malcolm et al. 2004). Further, the taxonomic diversity and community composition in the interstitial zone of the reservoir was expected to be lower and less diverse from the other sampled sites upstream and downstream.

2 Methods

2.1 Study area

The study was conducted within the area of reservoir called Doblarsko jezero (Jezero = lake) or lake near Most na Soči village (Western Slovenia, Southeastern Europe). The reservoir backwater is extending on the Soča River approximately 7 km upstream from a 40 m high dam located on the same river near the Podsela village. In the area of the reservoir backwater, it is also the confluence of the Soča River with smaller tributary, the Idrijca River, where the reservoir backwater is extending approximately 2 km upstream (see Fig. 1). Overall, the whole area of the reservoir extends for about 80 ha and is reaching a maximum depth of 32 m (Firbas 2001). The dam was built in 1938 to facilitate the operation of the downstream hydropower plants and is occasionally opened to release the sediment deposits from the reservoir.

The reservoir is located in the Soča River catchment in the Alpine area. The Soča River is a 138 km long alpine river that originates in the western part of the Julian Alps and flows from north to south near the Slovenian/Italian border (Fig. 1). After crossing the border and entering Italy, Soča River flows for another 40 km until it reaches the Adriatic Sea. The river has a pluvio-nival hydrological regime and a mean discharge of 91 $m^3 s^{-1}$ (Solkan gauging station, 1980–2019; SEA 2021). The Idrijca River is a tributary of the Soča River (Fig. 1), with a pluvio-nival hydrological regime and a mean discharge of 23 $m^3 s^{-1}$ (Hotešk gauging station, 1980-2019; SEA 2021). Geology of the catchment area of the studied rivers consists mainly of Mesozoic limestone and dolomite (Novak & Rman 2018).

2.2 Sampling

In early January 2018, the study reservoir was drained due to maintenance and repair works at the Podselo dam. The fieldwork was conducted on 24th and 25th January 2018, when Soča River had already washed away substantial parts of the fine sediments from the channel along the reservoir, exposing the underlying gravel beds (Fig. 2). This enabled access to sample in the gravel bed that had previously been inaccessible due to excessive lake depth and fine sediment thickness of up to 3 meters (Fig. 2).

In total, seven localities were sampled (Fig. 1, Table 1). Six of them were located upstream of the dam, of which one was within the deepest part of the reservoir (R), three upstream on the Soča River (U1 and U2 – river, U3 _ at the edge of the reservoir), and two on the Idrijca River (U4 – river, U5 – near the reservoir). One locality was located downstream from the dam (D), where the river water was still muddy during sampling due to fine sediments washed-out from the upstream reservoir. Deposits of fine sediments were also visible on the river banks (Fig. 2B).

At each locality, interstitial habitat was sampled using a Bou-Rouch pump (Bou & Rouch 1967). The wetted area with water depths between 10 and 30 cm was selected within the riverbed or reservoir bottom, at least 2 meters from the river bank or reservoir shore. Three spatial replicates (sampling points) with a distance of 10 m between each other were sampled at two depths: 30–60 cm and 60–90 cm below the surface to consider the spatial heterogeneity of the sampling locality. At each sampling depth, 30 L of water containing organisms and sediment were pumped out and filtered through nets with a mesh size of 500 µm to avoid collecting huge amounts of finer sediments. The samples were immediately stored in 96 % ethanol. Later, invertebrates (body size > 500 µm) were sorted out using a stereomicroscope (up to 40× magnification) in the laboratory. Animals were identified to the lowest possible taxonomic level and were stored in the Zoological collection of SubBioLab (Department of Biology, Biotechnical Faculty, University of Ljubljana).

During each sampling, the first 10 L of pore water were used to measure four abiotic parameters: temper-

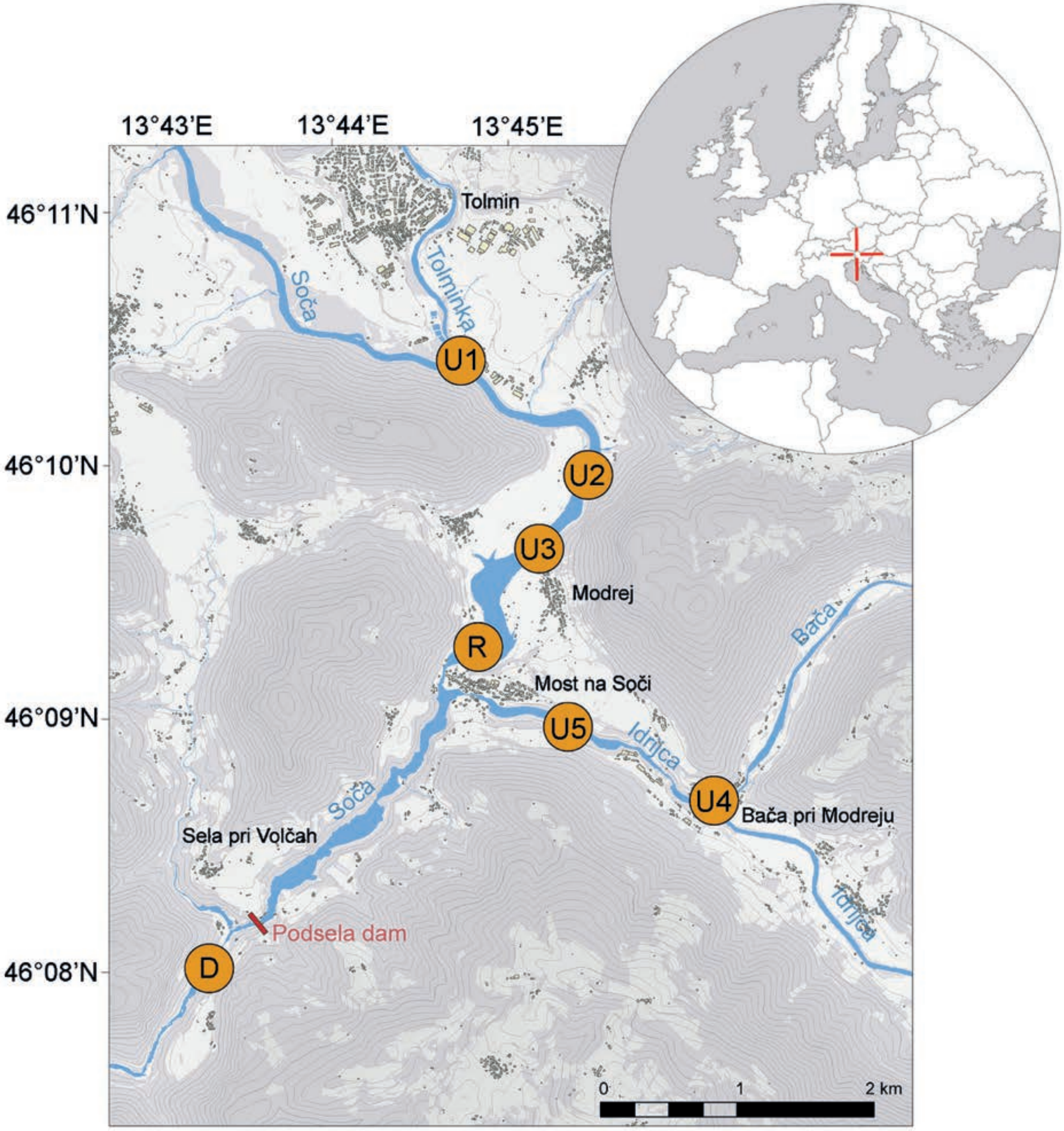

Fig. 1. The study area on the Soča and Idrijca Rivers in Western Slovenia (a red cross marks the position within Europe). Orange circles with codes denote sampling sites (see Table 1 for details), a red line marks the position of the dam in Podsela, due to which the reservoir lake upstream is formed.

ature, pH, oxygen saturation and conductivity, using a CyberScan 600 portable multimeter (Eutech Instruments).

2.3 Statistical analyses

To test for differences in abiotic parameters among localities, an analysis of variance (ANOVA) was performed for each abiotic parameter separately. Due to heteroscedastic variances, Welch's ANOVA was applied. For pairwise comparisons, Tukey's post hoc test was used. The effect of sample depth, i.e. 30–60 cm and 60–90 cm was tested separately, but as the samples were not significantly different, depth was omitted from the final model.

The statistical analyses of the invertebrate communities included only the presence/absence data of the taxa per site, while their abundances were not quanti-

Fig. 2. Consequences of damming visible after the draining of the reservoir on the Soča River in Western Slovenia. Canyons carved into the thick layers of sediments remained in the reservoir area after the water level dropped **(A)**, while the material was washed downstream **(B)** (photos: M. Zagmajster (A), T. Delić (B)).

fied (raw data available in Supplementary Material 2). Data on organisms from six samples were pooled for each locality, and differences in community composition among localities tested. The PERMANOVA, a nonparametric equivalent of MANOVA, was performed and calculated by permutations of the multivariate distance matrix. The Euclidean distance was used as the distance measure and 9999 permutations were run.

Finally, to quantify the overall similarity between localities in abiotic parameters and taxonomic composition using cluster analysis, the UPGMA agglomeration algorithm was applied. The abiotic parameters were clustered based on the Euclidean distance matrix calculated from the mean values. Invertebrate community composition was clustered based on the Jaccard similarity index, calculated from the presence or absence of taxon per each locality. We calculated the distances between localities in the ordination space using non-metric multidimensional scaling (nMDS). Three dimensions were selected based on stress value being much lower (0.017) than in two dimensions (0.142) (Clarke 1993). The results were presented as ordination plots. All statistical analyses were performed in PAST 3.0 (Hammer et al. 2001).

3 Results

The abiotic parameters varied greatly between the localities. Mean temperature of interstitial water varied from 4.9 to 9.1 °C, mean oxygen saturation from 34.9 to 95.9 %, mean pH from 6.9 to 8.4 and mean conductivity from 248 to 825 µS cm^{-1} (Table 1). All localities differed significantly in all measured abiotic parameters: temperature: $F = 18.94$, $p < 0.001$; pH: $F = 27.25$, $p < 0.001$; oxygen saturation: $F = 16.9$, $p < 0.001$; conductivity: $F = 154.9$, $p < 0.001$.

Even though pairwise comparisons revealed differences in some parameters among localities, it was the locality in the deepest part of the reservoir (R) that differed significantly from all others in all parameters except in temperature (Fig. 3, Supplementary Material 1). Here the interstitial water was low in mean oxygen saturation, having low pH and the highest mean conductivity. Also, at locality R in the reservoir, the measured parameters exhibited much higher spatial heterogeneity than at other localities (Fig. 3 and Fig 4, Table 1). The samples had more fine sediments particles than at other localities indicating sediment colmation, but we did not quantify this observation. At two sampling points within the reservoir, the smell of H_2S was present. There were two additional localities with significantly higher conductivity, and lower oxygen saturation, one on the Soča River (D), and the other on the Idrijca River (U5), just before its confluence with Soča. The former is just below the dam and the latter is at the same distance from the dam as the locality R.

In all localities together, we identified 40 taxa, among which 11 are considered stygobionts (Table 2, Supplementary Material 2). The latter belonged to Gastropoda, Copepoda and Amphipoda. Taxonomic richness per locality varied from 5 to 22 taxa, while individual samples contained between 0 and 15 taxa (Table 2, Supplementary Material 2). The smallest number of taxa and the lowest number of stygobionts was recorded in the deepest part of the reservoir (R), i.e. five taxa altogether and 0–3 taxa per sample and only one stygobiotic taxon (Table 2). Community composition differed significantly among most of lo-

Table 1. A list of localities on the rivers in Western Slovenia, where the interstitial habitats were sampled in January 2018, with locality codes and coordinates (see also Fig. 1). At each locality, six samples were taken with Bou-Rouch pump, three from 30–60 cm, and three from 60–90 cm, and then mean values of abiotic parameters were calculated from all six measurements (N = 6), with standard deviation (SD) given in parentheses.

Code	Locality	Latitude, longitude (WGS84)	Date (2018)	T [°C]	pH	O_2 saturation [%]	Conductivity [µS cm^{-1}]
U1	Gravel bar near confluence of Soča and Tolminka Rivers	46.173300 13.740173	24.1.	4.9 (0.5)	7.6 (0.2)	95.2 (3.8)	248.3 (3.8)
U2	Gravel bar near the Soča River 800 m N from Modrej	46.166620 13.752913	24.1.	8.5 (0.8)	8.1 (0.1)	87.7 (6.8)	265.6 (6.9)
U3	Gravel bar at the left bank of the Soča River at Modrej	46.161200 13.748800	24.1.	6.4 (0.2)	8.3 (0.1)	95.8 (3.7)	262.6 (6.2)
R	Most na Soči – gravel bar within the reservoir on Soča River	46.154792 13.743817	24./25.1.	6.6 (1.4)	6.9 (0.4)	34.9 (27.0)	825.8 (444.2)
U4	Gravel bar at the right bank of Idrijca river, 2 km upstream from confluence to Soča	46.145008 13.765222	24.1.	5.8 (0.7)	8.4 (0.1)	79.5 (2.8)	287.9 (4.5)
U5	Gravel bar at the right bank of Idrijca river, 750 m upstream from confluence to Soča	46.149650 13.751489	24.1.	5.7 (0.3)	8.4 (0.1)	62.3 (13.1)	344.6 (7.2)
D	Gravel bar at the right bank of Soča river, 500 m downstream from the Podsela dam	46.133676 13.718742	24.1.	9.1 (1.1)	7.6 (0.2)	65.3 (11.2)	336.0 (8.0)

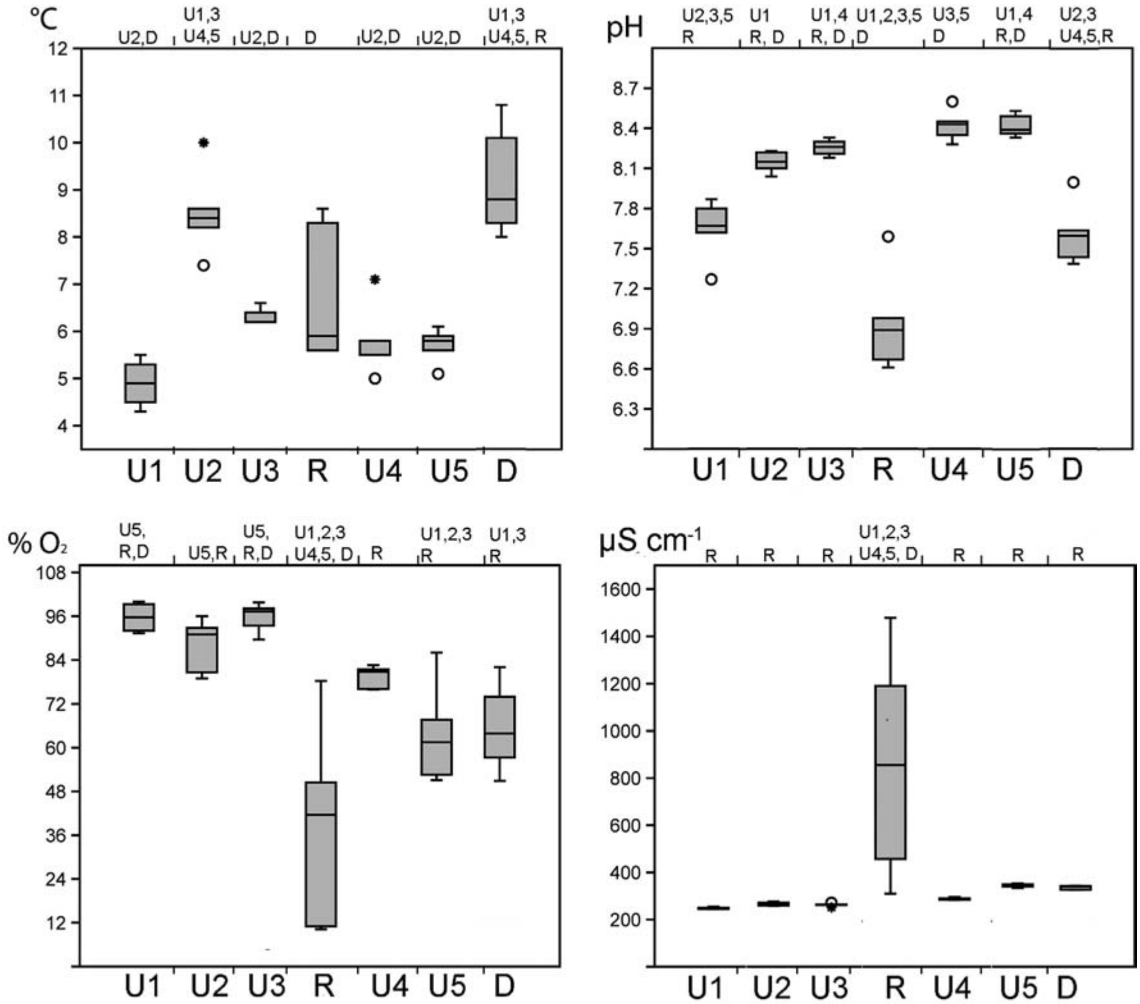

Fig. 3. Box plots of the abiotic parameters (top: temperature, pH, bottom: oxygen saturation, conductivity) measured at seven different locations, within the deepest point of the reservoir (R) and six other locations. See Table 1 and Fig. 1 for details on localities. Sites listed above each site boxplot are the ones statistically significantly different ($p < 0.05$) following Tukey's post-hoc tests (see also Supplementary Material 1).

calities ($F = 3.16$, $p < 0.01$, Table 3). Cluster analyses of both community composition and abiotic parameters were concordant with the results of ANOVA and PERMANOVA. On both dendrograms, the locality from the deepest point of the reservoir (R) differed most from all other localities (Fig. 4). The communities from the closest point to the deepest point of the reservoir (U3) and just behind the dam (D) were more similar to each other than to any of the other localities. Similarly, the most up-stream localities U1 and U4, and U2 and U5, formed separate groups. In nMDS plot of the first two coordinates, R was most distant from all the other localities, with other localities roughly distributed in three groups: U1, U3 and D, a group of U5 and U2 and U4 (Fig. 5). The plot indicating the associated taxa, is given in Supplementary Material 1.

4 Discussion

The results presented in this study demonstrate significant differences in quality of interstitial habitats and community composition within the areas with normal river flow and permeable gravel-bed (Fig. 1; U1, U4), and area normally filled with slowly flowing reservoir water and covered with few meters deep layers of mostly sand and silt sediments deposited over decades (Fig. 1; R). This study was possible only due to the reservoir draining which was carried out during dam maintenance. As we found out about the draining from the public media, we sampled interstitial habitats a few weeks later the draining started. In the meantime, river water washed out thick deposits of fine sediments out of the reservoir, and exposed the gravel-bed, that ena-

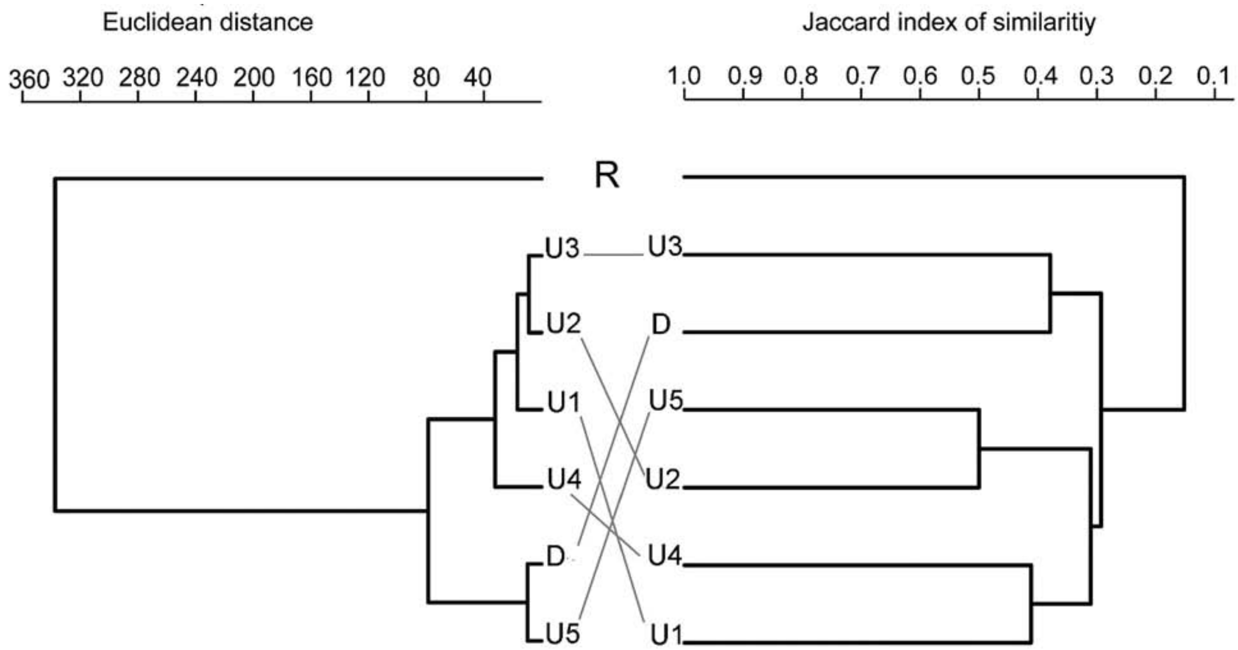

Fig. 4. Clustering diagrams showing results of the analysis of similarities of the sampling sites with respect to abiotic parameters (left) and invertebrate community composition (right). The locality from the reservoir (R) differed most from all other localities in both abiotic parameters and community composition. See Table 1 and Fig. 1 for details on localities.

bled direct access to interstitial habitats also within the deepest point of the reservoir (water column in filled reservoir of up to 32 meters depth).

The most evident environmental characteristics of the interstitial habitats in the area within the denudated zone of the reservoir (R) are significantly lower oxygen saturation and pH, and higher conductivity in comparison to upstream and downstream interstitial habitats (Fig. 3). Such differences are most likely related to the deposition of thick sediment layers within reservoir, colmation of the previously highly permeable gravel bed and consequently impaired hydrological exchange between surface and interstitial water (Brunke 1999). These conditions can be compared to lake profundal zones where oxygen content and interstitial permeability are low due to fine sediments and absence of hydrological exchange between surface and sediment subsurface (Hargrave 1972). Previous studies of the hyporheic zones in the Soča River catchment (a tributary of the Idrijca River – Bača River) indicated high rates of surface–subsurface exchange, both vertically (into the riverbed) and laterally (into gravel bars), as well as short retention times of stream water in the hyporheic zone due to the high permeability of sediments (Mori et al. 2011). In this study, the conductivity and oxygen saturation from the most distant upstream localities from the reservoir on both rivers (Fig. 1, 4U1 and U4;) were close to those measured in surface waters of Soča and Idrijca Rivers (data from Slovenian Environment Agency, https://www.arso.gov.si/vode/podatki/arhiv/kakovost_arhiv2018.html), indicating intensive surface–subsurface exchange at localities without impacts of reservoir.

Differences in environmental conditions between localities that reflect a gradient from lotic to lentic environment are also reflected in invertebrate richness and community composition. Total invertebrate richness was significantly lower in the deepest part of reservoir (R) previously covered with the thickest fine sediment layers compared to other sampling localities and had a significantly different community composition with only five taxa present. Among these we found gastropods, bivalves, ostracods, dipterans and stoneflies. All of these taxa are common interstitial inhabitants in this area and this type of gravel bed rivers (Mori et al. 2011) and were found also at other sampled locations during this study. The only stygobiotic species found here was a gastropod species *Hauffenia telinni* (Pollonera, 1898) with a known distribution within the Soča and Idrijca catchments, found mostly at interstitial or spring habitats, but also caves (Bodon et al. 2001). According to nMDS, the interstitial community of the Soča River from the upstream locality (Fig. 1; U2) was more alike that of its tributary, Idrijca

Table 2. A list of taxa from the interstitial habitats of two rivers in Western Slovenia, sampled in January 2018. The last two rows report on cumulative number of all taxa, and number of stygobionts per locality. See Table 1 and Fig. 1 for details on localities. * – stygobiont; 0 – not recorded; 1 – present.

Group	Family	Taxon	U1	U2	U3	R	U4	U5	D
Nematoda		Nematoda	1	0	1	0	0	1	1
Oligochaeta		Oligochaeta	0	1	1	0	0	0	1
Gastropoda	Hydrobiidae	*Hauffenia tellinii* *	0	1	1	1	0	0	1
Gastropoda	Hydrobiidae	*Hauffenia subpiscinalis* *	0	0	0	0	0	0	1
Gastropoda	Hydrobiidae	*Belgrandiella kusceri* *	0	1	1	0	0	0	1
Gastropoda	Hydrobiidae	*Iglica forumjuliana* *	0	0	0	0	0	0	1
Gastropoda	Lymnaeidae	*Galba truncatula*	0	0	0	0	0	0	0
Gastropoda	Ellobiidae	*Carychium tridentatum*	0	0	0	0	0	0	1
Gastropoda	Ancylidae	*Ancylus fluviatilis*	1	0	0	0	0	1	0
Bivalvia	Sphaeriidae	*Pisidium* cf. *casertanum*	0	1	1	1	0	1	1
Acarina		Acarina sp. 1	0	0	0	0	1	0	0
Acarina		Acarina sp. 2	1	0	0	0	0	0	0
Acarina		Acarina sp. 3	0	0	0	0	0	0	1
Acarina		Acarina sp. 4	0	1	0	0	0	0	0
Acarina	Unionicolidae	*Neumania* sp.	0	0	0	0	1	0	0
Ostracoda	Candonidae	Candoninae	0	0	0	0	0	0	1
Ostracoda	Candonidae	cf. *Candona* sp.	0	0	1	0	0	0	0
Ostracoda	Candonidae	cf. *Fabaeformiscandona* sp.	0	0	1	0	0	0	1
Ostracoda	Candonidae	cf. *Pseudocandona albicans*	0	0	1	1	0	0	0
Ostracoda	Candonidae	cf. *Candona candida*	0	0	0	0	0	0	1
Ostracoda	Candonidae	cf. *Cavernocypris subterranea*	0	0	0	0	0	0	1
Ostracoda	Cyprididae	*Cavernocypris subterranea*	0	0	1	0	0	0	0
Copepoda: Cyclopoida	Cyclopidae	*Acanthocylops robustus*	0	1	0	0	1	0	1
Copepoda: Cyclopoida	Cyclopidae	*Diacyclops clandestinus* *	1	0	0	0	1	1	0
Copepoda: Cyclopoida	Cyclopidae	*Diacyclops zschokkei* *	1	1	1	0	1	1	1
Copepoda: Cyclopoida	Cyclopidae	*Graeteriella unisetigera* *	1	0	0	0	0	1	1
Copepoda: Harpacticoida	Ameriidae	*Nitocrella* sp.*	0	0	0	0	0	1	0
Copepoda: Harpacticoida	Parastenocarididae	*Parastenocaris gertrudae* *	0	0	1	0	0	0	0
Amphipoda		Amphipoda	0	0	1	0	0	1	0
Amphipoda	Niphargidae	*Niphargus* cf. *aberrans* *	0	0	1	0	0	1	0
Amphipoda	Niphargidae	*Niphargus* cf. *minor* *	0	0	0	0	0	1	1
Diptera	Chironomidae	Chironomidae	1	1	1	0	1	1	1
Diptera	Simuliidae	Simuliidae	1	0	0	1	0	1	0
Diptera	Tipulidae	Tipulidae	0	1	1	0	1	1	0
Diptera	Ceratopogonidae	Ceratopogonidae	0	0	1	0	0	0	1
Trichoptera	Glossosomatidae	Glossosomatidae	0	1	0	0	0	1	0
Plecoptera	Capniidae	Capniidae	1	1	1	1	1	1	1
Ephemeroptera	Baetidae	Baetidae	1	1	1	0	1	1	1
Ephemeroptera	Heptageniidae	Heptageniidae	0	0	0	0	1	0	0
Coleoptera	Elmidae	Elmidae	1	1	0	0	1	0	1
		ALL TAXA	11	13	18	5	11	16	22
		STYGOBIONTS (%)	3 (27.3)	3 (23.1)	5 (27.8)	1 (20.0)	2 (18.2)	6 (31.5)	7 (31.8)

Table 3. Results of post-hoc pairwise comparisons of species composition between locations after PERMANOVA: *p*-values of pairwise tests, adjusted with the sequential Bonferroni method. Statistically significant ($p < 0.05$) results bold. See Table 1 and Fig. 1 for details on localities.

Locality	U1	U2	U3	R	U4	U5	D
U1							
U2	**0.0304**						
U3	**0.0195**	**0.0023**					
R	**0.0023**	**0.0015**	**0.0022**				
U4	1	**0.035**	**0.0022**	**0.0031**			
U5	**0.0021**	**0.0163**	**0.0021**	**0.0022**	**0.0022**		
D	**0.0047**	**0.0021**	**0.0227**	**0.0020**	**0.0015**	**0.0023**	

(Fig. 1; U4, U5), than to the community within the reservoir and sites close to the reservoir (Fig. 1; U3, R, D). The exception was U1, which grouped closer to U3 and D, but on account of generally distributed surface species (Table 2, Fig. 5). Roughly, similarity in community composition among localities was in line with the distance from the dam. Interestingly, at those localities the number of taxa was slightly lower than at U3 and D). Most probably hydrogeological settings are the main drivers of differences between the communities in this study. Gayraud & Philippe (2003) clearly demonstrated the strong linkages between overall in-

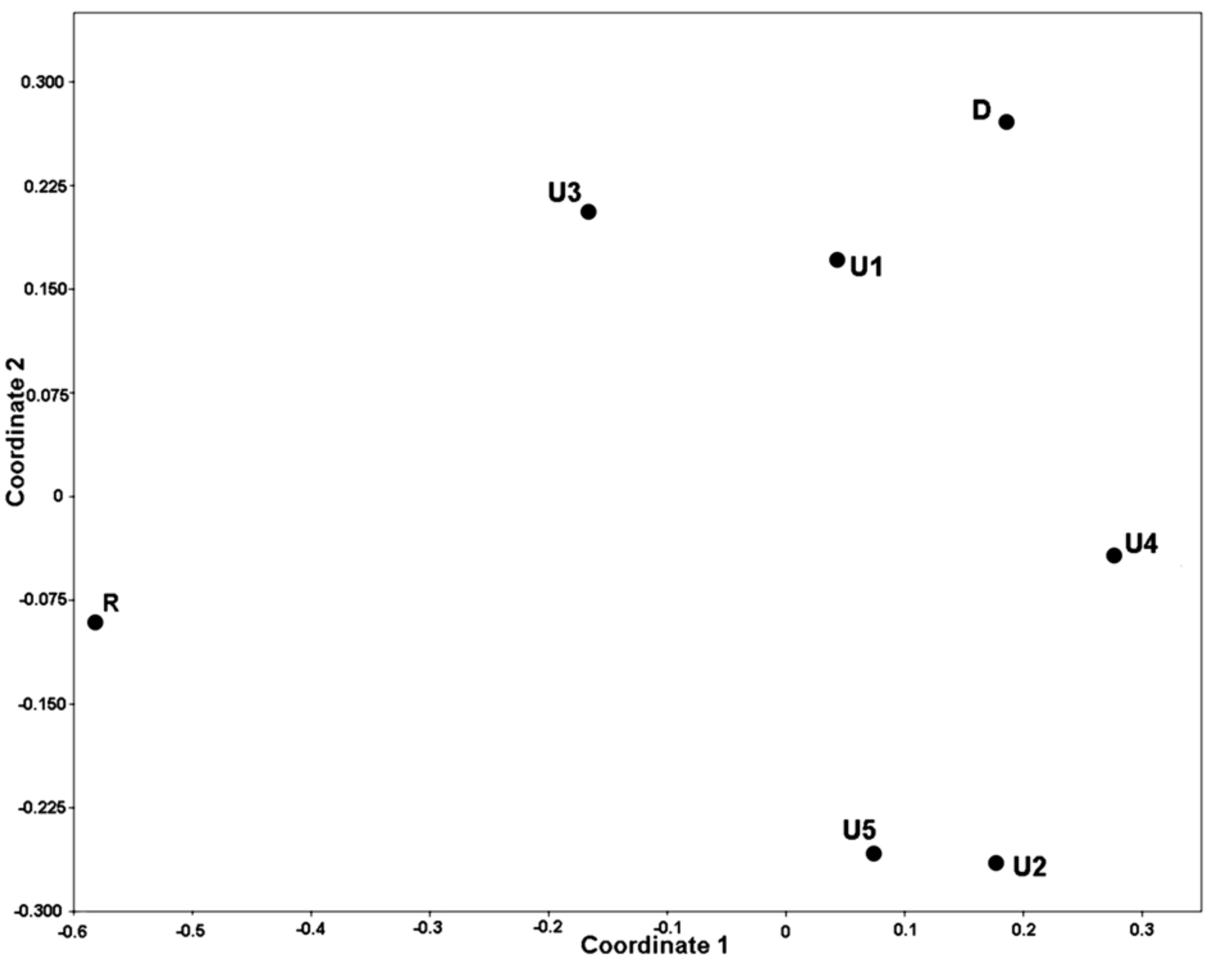

Fig. 5. The ordination diagram of the non-metric multidimensional scaling (nMDS) of invertebrate samples from seven localities calculated based on Jaccard similarity index using tridimensional space (for associated taxa, see Supplementary Material 1). See Table 1 and Fig. 2 for details on localities.

vertebrate density, taxon richness and effective porosity that is strongly related to grain-size distribution. The latter is strongly shaped by hydrology, the other important driver of riverine invertebrates including interstitial community characteristics as demonstrated by Olsen & Townsend (2005). When the dam is in full function, the water flow slows down from lotic to almost lentic regime starting at the locality U3, while locality D is situated 1 km downstream from the dam and it is affected by distinct water level fluctuations during hydropower plant operation. However, the number of taxa at D was higher than those from all upstream localities. When dam is operational, this site does not experience the effect of colmation, compared to most upstream sites in our study, indicating good environmental conditions for interstitial invertebrates. Dam removal can lead to temporary degradation of downstream habitats by debris previously accumulated in the reservoir (Habel et al. 2020). At this site below the dam (D), we did observe the increased sediment in the river water and on its banks, as well as higher conductivity and lower oxygen saturation of interstitial waters, but it may be that negative effects on interstitial fauna have not yet been expressed. More samples, on both spatial and temporal scale, should be collected to more profoundly understand the patterns of interstitial community distribution in this area.

The study showed a different degree of degradation of the interstitial habitat within the reservoir area and indicated impacts of released sediments from drained reservoir downstream from the dam. Despite small distances between the most distant sampling localities in the study (i.e. approximately 5 km measured along the longitudinal river corridor), differences in invertebrate communities by means of number of taxa were substantial. It could be assumed that the dam and intensive sand and silt particles within reservoir that deposit in thick layers severely impair surface-subsurface hydrological exchange and decrease quality of environmental conditions important for interstitial invertebrates (e.g. oxygen conditions, size and connectivity of interstitial pores). However, the magnitude of this effect needs further investigation. The study was conducted in a short time window during cold period, and only a limited part of the alluvium was sampled, so additional sampling over a larger spatial and temporal scale, also using sampling nets with smaller mesh size (100 µm) for collecting meiofauna, could confirm or reject current observations. Many studies have demonstrated the high species richness and densities of stygobionts in the shallow layers of the hyporheic zone and groundwater at depths between 0.3–1.5 m (Danielopol 1976; Dole-Olivier & Marmonier 1992; Altmoos et al. 2009; Mori et al. 2011; Prevorčnik et al. 2019). However, heavy sedimentation, i.e. colmation, has been found to have the potential to prevent benthic fauna from accessing the interstitial habitats and its resources. This can ultimately lead to a reduction in diversity, thereby limiting the overall productivity and resilience of the lotic ecosystem (Mathers et al. 2014).

While this study clearly showed negative effects of river damming, reservoir formation and reservoir drainage on interstitial invertebrate fauna, it also opened many questions that require further investigation. How deep does the effect of sedimentation in the reservoir bottom extend? Do groundwater layers at greater depths represent a continuum along the river? How does the colmation of the hyporheic zone affect river functioning in general? How rapid is the colmation process? How does the composition of species in the alluvium and their interactions affect ecosystem services, particularly self-purification processes and water quality? It is highly recommended to monitor these processes regularly, with the clear goal of ensuring connectivity between the river and the underlying groundwater and maintaining the full functionality of the river. Even though the use of smaller mesh size could result in catching more smaller animals (microcrustaceans), this could result in higher abundances, and not necessarily additional species since we still collected several meiofaunal taxa despite using mesh size of 500 µm.

Although the European Groundwater Directive has recognized groundwater as an ecosystem, its biological component has been largely neglected (Fišer et al. 2022). As a consequence, deep knowledge gaps and lack of evaluations in environmental impact assessments could lead to losing an important share of European biodiversity and its endemic species. This is particularly relevant amidst the challenges of the climate change era and the emerging societies shifting towards energy sources that emit less carbon dioxide. Although originally perceived as such, hydropower has not proven to be a sustainable energy source, as large construction projects can destroy biodiversity and ecosystem services (Poff et al. 2007; Poff & Zimmerman 2010). Our results show that artificially formed reservoirs disable an important part of the river ecosystem below the riverbed, which is crucial for the normal functioning of the river. This should be taken into account when any planning of new dams and corresponding reservoirs is made, as they severely affect all four dimensions of lotic habitats (Ward 1989), impact river biodiversity and disrupt river continuity.

Authors' contributions: The sampling strategy was developed by MZ and CF and the fieldwork was carried out by MZ, GB, ŠB, TD, ŽF, KK, SP, VZ and CF. Sample sorting and taxonomic identification was carried out by GB, SP, NM, AB and CF. CF, MZ and NM analysed the data and prepared the first draft the manuscript. All authors contributed equally to the final version of the manuscript.

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Supplementary Material 1

Table S1. Tukey's post-hoc tests, *p*-values of pairwise comparisons for temperature between localities. Statistically significant ($p < 0.05$) results bold.

Locality	U1	U2	U3	R	U4	U5	D
U1							
U2	**0.0001**						
U3	0.2288	**0.0205**					
R	0.0962	0.0591	0.9994				
U4	0.1884	**0.0267**	1	0.9999			
U5	0.8301	**0.0012**	0.9345	0.7428	0.901		
D	**0.0001**	0.9959	**0.004**	**0.0129**	**0.0053**	**0.0003**	

Table S2. Tukey's post-hoc tests, *p*-values of pairwise comparisons for pH between localities. Statistically significant ($p < 0.05$) results bold.

Locality	U1	U2	U3	R	U4	U5	D
U1							
U2	**0.0172**						
U3	**0.002**	0.9821					
R	**0.0003**	**0.0001**	**0.0001**				
U4	0.9588	1	**0.0008**	0.9566			
U5	**0.0002**	0.4718	0.9167	**0.0001**	**0.0013**		
D	1	**0.0099**	**0.0011**	**0.0004**	**0.0001**	**0.0002**	

Table S3. Tukey's post-hoc tests, *p*-values of pairwise comparisons for oxygen saturation between localities. Statistically significant ($p < 0.05$) results bold.

Locality	U1	U2	U3	R	U4	U5	D
U1							
U2	0.9492						
U3	1	0.9291					
R	**0.0001**	**0.0001**	**0.0001**				
U4	0.2236	0.7946	0.1942	**0.0002**			
U5	0.0017	**0.0248**	**0.0014**	**0.0124**	0.4272		
D	**0.0057**	0.0714	**0.0046**	**0.0038**	0.7044	0.9994	

Table S4. Tukey's post-hoc tests, *p*-values of pairwise comparisons for conductivity between localities. Statistically significant ($p < 0.05$) results bold.

Locality	U1	U2	U3	R	U4	U5	D
U1							
U2	1						
U3	1	1					
R	**0.0001**	**0.0002**	**0.0002**				
U4	0.999	0.9999	0.9999	**0.0002**			
U5	0.9535	0.9824	0.9787	**0.0005**	0.9986		
D	0.9713	0.9907	0.9884	**0.0004**	0.9996	1	

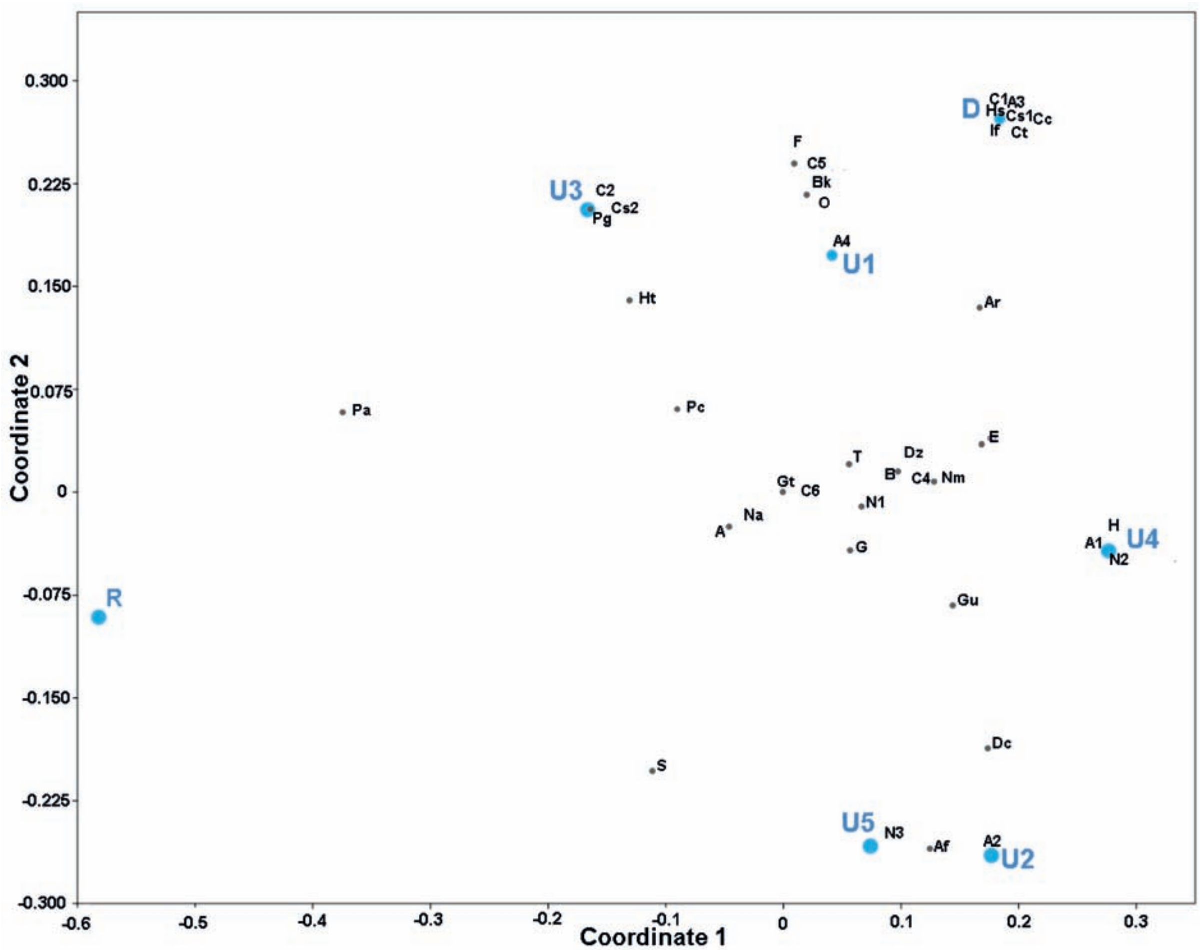

Fig. S1. NMDS ordination diagram indicating associated taxa. The plot of the first two coordinates of 3D nMDS ordination analysis, with sites and taxa shown. Site numbers are in blue, for details see Table 1 and Fig. 1. Abbreviations: N1 – Nematoda, O – Oligochaeta, Ht – *Hauffenia tellinii**, Hs – *Hauffenia subpiscinalis**, Bk – *Belgrandiella kusceri**, If – *Iglica forumjuliana**, Gt – *Galba truncatula*, Ct – *Carychium tridentatum*, Af – *Ancylus fluviatilis*, Pc – *Pisidium* cf. *casertanum*, A1 – Acarina sp1, A2 – Acarina sp2, A3 – Acarina sp3, A4 – Acarina sp4, N – *Neumania* sp., C1 – Candoninae, C2 – cf. *Candona* sp. , F – cf. *Fabaeformiscandona* sp., Pa – cf. Pseudocandona albicans, Cc – cf. Candona candida, Cs1 – cf. *Cavernocypris subterranean*, Cs2 – *Cavernocypris subterranean*, Ar – *Acanthocylops robustus*, Dc – *Diacyclops clandestinus**, Dz – *Diacyclops zschokkei* *, Gu – *Graeteriella unisetigera* *, N3 – *Nitocrella* sp.*, Pg – *Parastenocaris gertrudae* *, A – Amphipoda, Na – *Niphargus* cf. *aberrans* *, Nm – *Niphargus* cf. *minor* *, C – Chironomidae, S – Simuliidae, T – Tipulidae, C5 – Ceratopogonidae, G – Glossosomatidae, C – Capniidae, B – Baetidae, H – Heptageniidae, E – Elmidae (for details, see Table 2).

Supplementary Material 2

Sheet 1. Abiotic data raw.

Code	Locality	Longitude	Latitude	Date	Sample point	Depth [cm]	T [°C]	pH	O_2 [%]	conductivity [microS]
U1	Gravel bar near confluence of Soča and Tolminka Rivers	13.740173	46,173300	24.01.2018	1	30–60	4.5	7.27	100.0	244.2
U1	Gravel bar near confluence of Soča and Tolminka Rivers	13.740173	46.173300	24.01.2018	1	60–90	5.3	7.63	92.0	254.6
U1	Gravel bar near confluence of Soča and Tolminka Rivers	13.740173	46.173300	24.01.2018	2	30–60	4.8	7.62	91.3	247.4
U1	Gravel bar near confluence of Soča and Tolminka Rivers	13.740173	46.173300	24.01.2018	2	60–90	5.5	7.67	95.7	245.5
U1	Gravel bar near confluence of Soča and Tolminka Rivers	13.740173	46.173300	24.01.2018	3	30–60	4.9	7.80	92.9	250.6
U1	Gravel bar near confluence of Soča and Tolminka Rivers	13.740173	46.173300	24.01.2018	3	60–90	4.3	7.87	99.3	247.6
U2	Gravel bar near the Soča River 800 m N from Modrej	13.752913	46.166620	24.01.2018	1	30–60	10.0	8.10	86.8	264.4
U2	Gravel bar near the Soča River 800 m N from Modrej	13.752913	46.166620	24.01.2018	1	60–90	8.4	8.15	96.0	262.8
U2	Gravel bar near the Soča River 800 m N from Modrej	13.752913	46.166620	24.01.2018	2	30–60	8.6	8.10	79.0	271.7
U2	Gravel bar near the Soča River 800 m N from Modrej	13.752913	46.166620	24.01.2018	2	60–90	7.4	8.04	80.7	276.1
U2	Gravel bar near the Soča River 800 m N from Modrej	13.752913	46.166620	24.01.2018	3	30–60	8.2	8.22	92.8	258.1
U2	Gravel bar near the Soča River 800 m N from Modrej	13.752913	46.166620	24.01.2018	3	60–90	8.3	8.23	91.0	260.4
U3	Gravel bar at the left bank of the Soča River at Modrej	13.748800	46.161200	24.01.2018	1	30–60	6.6	8.18	97.3	271.0
U3	Gravel bar at the left bank of the Soča River at Modrej	13.748800	46.161200	24.01.2018	1	60–90	6.4	8.24	99.8	262.7
U3	Gravel bar at the left bank of the Soča River at Modrej	13.748800	46.161200	24.01.2018	2	30–60	6.2	8.33	96.4	264.0
U3	Gravel bar at the left bank of the Soča River at Modrej	13.748800	46.161200	24.01.2018	2	60–90	6.2	8.26	89.6	264.4
U3	Gravel bar at the left bank of the Soča River at Modrej	13.748800	46.161200	24.01.2018	3	30–60	6.4	8.21	93.4	251.9
U3	Gravel bar at the left bank of the Soča River at Modrej	13.748800	46.161200	24.01.2018	3	60–90	6.3	8.30	98.1	261.4
R	Most na Soči – within the reservoir on Soča River	13.743817	46.154792	24.01.2018	1	30–60	8.3	6.89	11.0	1478.0
R	Most na Soči – within the reservoir on Soča River	13.743817	46.154792	24.01.2018	1	60–90	8.6	6.67	10.2	1190.0
R	Most na Soči – within the reservoir on Soča River	13.743817	46.154792	25.01.2018	2	30–60	5.9	7.59	78.3	309.9
R	Most na Soči – within the reservoir on Soča River	13.743817	46.154792	25.01.2018	2	60–90	5.6	6.78	41.6	855.0
R	Most na Soči – within the reservoir on Soča River	13.743817	46.154792	25.01.2018	3	30–60	5.7	6.98	50.5	457.5
R	Most na Soči – within the reservoir on Soča River	13.743817	46.154792	25.01.2018	3	60–90	5.6	6.61	17.9	664.6
U4	Gravel bar at the right bank of Idrijca river, 2 km upstream from confluence to Soča	13.765222	46.145008	24.01.2018	1	30–60	5.8	8.60	82.6	290.0
U4	Gravel bar at the right bank of Idrijca river, 2 km upstream from confluence to Soča	13.765222	46.145008	24.01.2018	1	60–90	5.5	8.43	76.1	285.5
U4	Gravel bar at the right bank of Idrijca river, 2 km upstream from confluence to Soča	13.765222	46.145008	24.01.2018	2	30–60	5.0	8.45	80.8	284.9
U4	Gravel bar at the right bank of Idrijca river, 2 km upstream from confluence to Soča	13.765222	46.145008	24.01.2018	2	60–90	5.6	8.35	81.4	283.3
U4	Gravel bar at the right bank of Idrijca river, 2 km upstream from confluence to Soča	13.765222	46.145008	24.01.2018	3	30–60	5.8	8.28	80.2	295.8
U4	Gravel bar at the right bank of Idrijca river, 2 km upstream from confluence to Soča	13.765222	46.145008	24.01.2018	3	60–90	7.1	8.37	76.0	287.8
U5	Gravel bar at the right bank of Idrijca river, 750 m upstream from confluence to Soča	13.751489	46.149650	24.01.2018	1	30–60	5.7	8.49	52.6	353.2

U5	Gravel bar at the right bank of Idrijca river, 750 m upstream from confluence to Soča	13.751489	46.149650	24.01.2018	1	60–90	5.9	8.36	51.1	348.5
U5	Gravel bar at the right bank of Idrijca river, 750 m upstream from confluence to Soča	13.751489	46.149650	24.01.2018	2	30–60	6.1	8.39	61.5	343.9
U5	Gravel bar at the right bank of Idrijca river, 750 m upstream from confluence to Soča	13.751489	46.149650	24.01.2018	2	60–90	5.8	8.36	55.0	349.1
U5	Gravel bar at the right bank of Idrijca river, 750 m upstream from confluence to Soča	13.751489	46.149650	24.01.2018	3	30–60	5.1	8.53	86.0	333.3
U5	Gravel bar at the right bank of Idrijca river, 750 m upstream from confluence to Soča	13.751489	46.149650	24.01.2018	3	60–90	5.6	8.33	67.7	339.6
D	Gravel bar at the right bank of Soča river, 500 m downstream from the Podsela dam	13.718742	46.133676	24.01.2018	1	30–60	8.0	8.00	82.0	325.5
D	Gravel bar at the right bank of Soča river, 500 m downstream from the Podsela dam	13.718742	46.133676	24.01.2018	1	60–90	8.4	7.64	74.0	326.3
D	Gravel bar at the right bank of Soča river, 500 m downstream from the Podsela dam	13.718742	46.133676	24.01.2018	2	30–60	8.3	7.60	63.9	341.5
D	Gravel bar at the right bank of Soča river, 500 m downstream from the Podsela dam	13.718742	46.133676	24.01.2018	2	60–90	8.8	7.44	57.3	340.0
D	Gravel bar at the right bank of Soča river, 500 m downstream from the Podsela dam	13.718742	46.133676	24.01.2018	3	30–60	10.8	7.39	50.9	343.8
D	Gravel bar at the right bank of Soča river, 500 m downstream from the Podsela dam	13.718742	46.133676	24.01.2018	3	60–90	10.1	7.44	63.7	339.1

Sheet 2. Community raw data. Taxonomic group (grey marks stygiobiont).

Code	Nematoda	Oligochaeta	Gastropoda	Gastropoda	Gastropoda	Gastropoda	Gastropoda	Gastropoda	Gastropoda	Bivalvia	Acarina	Acarina	Acarina	Acarina	Acarina	Ostracoda	Ostracoda	Ostracoda	Ostracoda	Ostracoda
	Nematoda	**Oligochaeta**	***Hauffenia tellinii***	***Hauffenia subpiscinalis***	***Belgrandiella kusceri***	***Iglica forumjuliana***	***Galba truncatula***	***Carychium tridentatum***	***Ancylus fluviatilis***	***Pisidium* cf. *casertanum***	**Acarina sp. 1**	**Acarina sp. 2**	**Acarina sp. 3**	**Acarina sp. 4**	***Neumania* sp.**	**Candoninae**	***Candona* sp. cf.**	**cf. *Fabaeformiscandona* sp.**	**cf. *Pseudocandona albicans***	**cf. *Candona candida***
U3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
R	0	1	1	0	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U4	1	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
U4	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0
U4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U4	1	0	1	0	1	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0
U4	1	0	1	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
U4	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U5	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U5	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1	1	0
U5	0	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U5	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
U5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U5	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
D	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
D	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0
D	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0
D	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
D	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0
D	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U3	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0
U3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
R	1	0	0	0	0	0	0	0	1	1	0	0	0	0	0	0	0	0	0	0
R	0	0	0	0	0	0	0	0	1	1	0	0	0	0	0	0	0	0	0	0
U4	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U4	0	1	1	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U4	1	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0
U4	1	1	1	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
U4	1	1	1	0	0	1	0	1	0	1	0	0	1	0	0	1	0	1	0	1
U4	1	1	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Ostracoda	Ostracoda	Copepoda	Copepoda	Copepoda	Copepoda	Copepoda	Copepoda	Amphipoda	Amphipoda	Amphipoda	Diptera	Diptera	Diptera	Diptera	Trichoptera	Plecoptera	Ephemeroptera	Ephemeroptera	Ephemeroptera		
cf. *Cavernocypris subterranea*	***Cavernocypris subterranea***	***Acanthocylops robustus***	***Diacyclops clandestinus***	***Diacyclops zschokkei***	***Graeteriella unisetigera***	***Nitocrella* sp.**	***Parastenocaris gertrudae***	**Amphipoda**	***Niphargus* cf. *aberrans***	***Niphargus* cf. *minor***	**Chironomidae**	**Simuliidae**	**Tipulidae**	**Ceratopogonidae**	**Glossosomatidae**	**Capniidae**	**Baetidae**	**Heptageniidae**	**Elmidae**	**SUM – ALL TAXA**	**SUM – STYGOBIOTIC**
0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	1	1	1	0	1	5	1
0	0	0	0	1	0	0	0	0	0	0	1	0	1	0	0	1	1	0	1	6	1
0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	1	1	0	0	4	1
0	0	1	0	1	0	0	0	0	0	0	1	0	1	0	0	1	1	0	0	6	1
0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	1
0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	1	1	0	0	9	3
0	0	0	0	1	0	0	0	0	0	0	1	1	0	0	0	1	1	0	0	6	1
0	0	0	0	0	1	0	0	0	0	0	1	0	0	0	0	1	0	0	0	3	1
0	0	0	1	0	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	3	1
0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	4	0
0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	1	1	0	1	6	1
0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	1	0
0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	9	3
0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	2
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0	0	0	0	1	0	0	0	0	0	0	1	0	0	0	0	1	1	0	0	9	2
0	1	0	0	1	0	0	0	1	0	0	1	0	0	0	0	0	0	0	0	6	2
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0	0	0	0	1	0	1	0	0	0	0	1	0	0	0	0	0	1	0	0	4	2
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0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	5	0
0	0	0	0	0	1	0	0	0	0	1	0	0	0	1	0	0	0	0	0	6	4
0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	2	0
0	0	0	0	1	1	0	0	0	0	0	0	0	0	0	0	0	1	0	0	7	4
1	0	0	0	1	1	0	0	0	0	0	0	0	0	0	0	1	0	0	1	15	4
0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	2