# Benthic macroinvertebrate community structure in Batušinac ponds (Serbia) relative to the distance from a river

### Abstract:

Batušinac ponds are situated in south-eastern Serbia, near the Južna Morava River. In this study we compared benthic macroinvertebrate community structures of the river and three ponds located at contrasting distances from the river in order to assess the impact of the river proximity on benthic macroinvertebrate community structures in Batušinac ponds. Only 7 out of 43 taxa were common for the river and the study ponds. Based on the NMDS analysis, the community structures of the river and the study pond situated closest to the river were the most similar. The results of an ANOSIM analysis showed that community structures of the river and all study ponds were significantly different (R=0.901; p=0.003). Given the observed differences between the river and pond macroinvertebrate communities, it can be concluded that Batušinac ponds contribute significantly to freshwater diversity of the study area.

#### Key words:

macrozoobenthos, species composition, lotic taxa, lentic taxa, spatial proximity

#### Apstract:

#### Struktura zajednice bentosnih makroinvertebrata Batušinačkih bara (Srbija) u odnosu na udaljenost od reke

Batušinačke bare su smeštene u jugoistočnoj Srbiji, u blizini reke Južne Morave. U ovoj studiji smo upoređivali strukturu zajednica bentosnih makroinvertebrata reke i tri bare smeštene na različitoj udaljenosti od reke, s ciljem da procenimo uticaj blizine reke na strukturu zajednica bentosnih makroinvertebrata u Batušinačkim barama. Od ukupno 43 taksona, svega je 7 bilo zajedničko za reku i istraživane bare. Rezultati NMDS analize su pokazali da su zajednice reke i bare smeštene najbliže reci najsličnije po strukturi. Rezultati ANOSIM analize su pokazali da se strukture zajednica reke i svih bara međusobno značajno razlikuju (R=0.901; p=0.003). Uzimajući u obzir uočene razlike između zajednica makroinvertebrata reke i bara, može se zaključiti da Batušinačke bare značajno doprinose slatkovodnom diverzitetu istraživanog područja.

Ključne reči:

makrozoobentos, sastav vrsta, lotički taksoni, lentički taksoni, prostorna bliskost

# Introduction

Ponds are small and shallow (surface area  $1 \text{ m}^2-2$  ha; max depth 8 m), natural or man-made water bodies that can be permanently or seasonally filled with standing water (Biggs et al., 2005). Despite their small size, ponds represent a valuable source of freshwater biodiversity, since they host more rare and threatened species than other freshwater ecosystems (Williams et al., 2004; Davies et al., 2008). Ponds

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contribute disproportionately to regional biodiversity due to their high  $\beta$  diversity, which is related to the existence of a wide variety of pond types that differ in their features (e.g., length of their hydroperiod, size, or physical and chemical properties), even on a small spatial scale (De Meester et al., 2005).

Macroinvertebrates constitute an important part of animal diversity in small water bodies, such as ponds (Davies et al., 2008). It has been recognized that community structure (species composition and



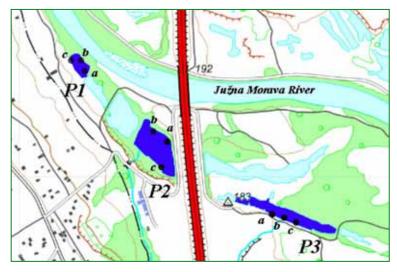


Fig. 1. Map of the study area with the position of the study sites

abundances) of macroinvertebrate communities in ponds is strongly driven by local environmental and biotic factors (e.g., water permanence, water chemistry, macrophyte cover, and fish predation) (Trigal et al., 2007; Hentges & Stewart, 2010; Walker et al., 2013; Gleason & Rooney, 2018). In addition, evidence has accumulated that macroinvertebrate communities in ponds are structured by dispersal processes (Leibold et al., 2004; Van de Meutter et al., 2007). Therefore, besides local, regional factors are also of huge importance for structuring macroinvertebrate communities in ponds. Among structuring processes that operate at regional level, connectivity between habitats (e.g., small distance among habitats) is of huge importance for macroinvertebrate communities in ponds (Oertli et al., 2008).

In this study we compared the community structures of benthic macroinvertebrates between a part of the river and three ponds situated at contrasting distances from the river. We aimed to assess the impact of the river proximity on benthic macroinvertebrate community structures in ponds. We hypothesised that a pond situated closest to the river and the river have the most similar benthic macroinvertebrate community structures.

# Materials and Methods

# Study area

Batušinac ponds are located in south-eastern Serbia, about 10 km west of the City of Niš. Study area encompasses three permanent ponds situated on the left bank of the Južna Morava River, at distances of 17.5 m, 212 m, and 386 m (**Fig. 1**). Study ponds are the remains of the former river channel that were isolated, as a result of the redirection of the river flow during the construction of a nearby highway (Fig. 1). The ponds are connected to the current riverbed both by groundwater supplies and by river flooding (Ranđelović et al., 2007). Pond 1 is the smallest (surface area of 429.42 m<sup>2</sup>; maximum depth 120 cm). It is situated nearest to the river (Fig. 1), and it is consequently the most intensively flooded. Pond 2 has a surface area of 6340.88 m<sup>2</sup> and maximum depth of 250 cm. Due to their spatial proximity pond 1 and pond 2 become a connected system during the flood events (Fig. 1). Pond 3 is the largest (surface area of 14206.05 m<sup>2</sup>; maximum depth 200 cm) and it is farthest from the river (Fig. 1). In addition, pond 3 is isolated from the rest of the two study ponds by the nearby highway (Fig. 1).

# Sampling

Macroinvertebrate sampling was performed in September 2016. Within each pond, we developed three sampling transects (study sites) starting from the shore towards the deeper water. Sampling transects were distributed across the ponds to cover all mesohabitats within a pond. One sampling transect was developed within the river, encompassing river banks and a midstream. The part of the river investigated represents the river's middle rich. Benthic macroinvertebrates were sampled using a kick-net of 250 µm mesh size. Three benthic subsamples were taken from the most common substrate types along each transect. All three benthic subsamples were merged into a single sample. The material collected was sorted out of sediment and preserved in 70% ethanol. Macroinvertebrates were identified to the lowest possible taxonomic level using relevant taxonomic keys (Elliot et al., 1988; Wallace et al., 1990; Edington & Hildrew, 1995; Nilsson, 1997; Waringer & Graf, 1997; Gerken & Sternberg, 1999; Timm, 1999; Pfleger, 2000; Bauernfeind & Humpesch, 2001; Glöer, 2002; Eiseler, 2005; Elliot & Humpesch, 2010). Most of the taxa were identified to species or genus level, except the families whose individuals were damaged: Aphelocheiridae (Hemiptera), Heptageniidae (Ephemeroptera), Hydrophilidae, Elmidae (Coleoptera), or whose identification requires taxonomic expertise, such as Chironomidae (Diptera).

## Data analysis

Family Chironomidae was present at all study sites, and due to its ubiquitous nature, was excluded from the analyses. 
 Table 1. List of benthic macroinvertebrate taxa recorded across the study sites. x indicates the presence of a taxon

Taxon	Abbreviation	P1a	P1b	P1c	P2a	P2b	P2c	P3a	P3b	P3c	River
Radix auricularia (Linnaeus, 1758)	Raur				X	x	X	x	X	x	
<i>Radix labiata</i> (Rossmässler, 1835)	Rlab							х			х
<i>Physella acuta</i> (Draparnaud, 1805)	Pacu			х	х	X	X	X	X	X	
Gyraulus sp.	Gyr				Х						
Acroloxus lacustris (Linnaeus, 1758)	Alac										Х
<i>Viviparus acerosus</i> (Bourguignat, 1862)	Vace										Х
Unio pictorum (Linnaeus, 1758)	Upic										х
Limnodrilus hoffmeisteri Claparède, 1862	Lhoff	х	х	х	х						Х
<i>Limnodrilus</i> claparedeanus Ratzel, 1868	Lcla	X			X						
<i>Limnodrilus undekemianus</i> Claparède, 1862	Lund			X							
<i>Tubifex tubifex</i> (Müller, 1774)	Ttub	X			x				x	X	х
Erpobdella octoculata (Linnaeus, 1758)	Eoct				х						х
Helobdella stagnalis (Linnaeus, 1758)	Hsta				х						
Haementeria costata (Fr. Müller, 1846)	Hcos				х						
Heamopis sanguisuga (Linnaeus, 1758)	Hsan						x				
Gammarus balcanicus Schäferna, 1922	Gbal										X
Pelocoris sp.	Pel						Х				
Aphelocheiridae	Aph										Х
Platycnemis pennipes (Pallas, 1771)	Ppen			х	х	х	х	х			
<i>Coenagrion puella</i> (Linnaeus, 1758)	Cpue						x	х	x		
Ischnura elegans (Vander Linder, 1820)	Iele			х					X		
Pyrrhosoma nymphula (Sulzer)	Pnym				X			X	X		
Sympetrum fonscolombii (Selys, 1840)	Sfon				X	х	X			X	
Anax imperator Leach, 1815	Aimp				X						
Calopteryx splendens (Harris, 1780)	Cspl										х

Gomphus vulgatissimus (Linnaeus, 1758)	Gvul										х
Onychogomphus forcipatus (Linnaeus, 1758)	Ofor										х
Ecnomus sp.	Ecn								х		
Athripsodes aterrimus (Stephens, 1836)	Aate							х			
Leptocerus sp.	Lep							х			
Hydropsyche contubernalis McLachlan, 1865	Hcon										х
Caenis sp.	Cae							х	х	х	х
Cloeon dipterum	Cdip									х	
Baetis rhodani (Pictet, 1843)	Brho										х
Heptageniidae	Hep										х
Potamanthus luteus (Linnaeus, 1767)	Plut										х
Hydrophilidae	Hydr										
<i>Peltodytes caesus</i> (Duftschmid, 1805)	Pcae						х				
<i>Dytiscus</i> sp.	Dyt							х			
Elmidae	Elm										х
Culex sp.	Cule					Х					
Culicoides sp.	Culic	х	х	х					х	х	х
Chironomidae		х	Х	Х	Х	Х	Х	х	х	х	Х

To visualise differences/similarities in the benthic macroinvertebrate community structures of study ponds and the river and spatial patterns of benthic macroinvertebrate taxa we used non-metric multidimensional scaling (NMDS) with a Bray-Curtis dissimilarity matrix. To test the statistical significance of the NMDS results on benthic macroinvertebrate community structures we used an Analysis of Similarities (ANOSIM). Prior to analyses abundance data were square-root transformed. All analyses were performed using PRIMER version 6 (Clarke & Gorley, 2006).

# Results

Overall, 43 taxa (including the family Chironomidae) were recorded across the study sites (**Tab. 1**). A total of 31 taxa were recorded across the study ponds, of which 9 taxa were recorded in pond 1; 20 taxa in pond 2, and 17 taxa in pond 3. The recorded taxa belonged to 22 families, 10 orders, 3 classes and 3 phyla. Benthic macroinvertebrate community of the river was represented with 20 taxa, belonging to 18 families, 12 orders, 5 classes and 3 phyla. Insects were the taxonomically dominant class, both in the river and the study ponds, of which the order

Ephemeroptera was the taxonomically dominant in the river (represented with 4 taxa), while the order Odonata was the taxonomically dominant in the study ponds (represented with 6 taxa). *Limnodrilus hoffineisteri* Claparède, 1862 (Annelida, Oligochaeta) was the most abundant species both in the river and in pond 1. The most abundant species in pond 2 was *Radix auricularia* (Linnaeus, 1758) (Mollusca, Gastropoda), followed by *Pyrrhosoma nymphula* (Sulzer) (Insecta, Odonata) (Fig. 2). *Pyrrhosoma nymphula* was the most abundant species in pond 3 (Fig. 2).

Overall, 23 taxa were recorded exclusively in the study ponds, while 13 taxa were recorded exclusively in the river (**Tab. 1**; **Fig. 3**). Besides the Chironomidae family, common taxa of both pond and the river were: *Radix labiata* (Rossmässler, 1835) (Mollusca, Gastropoda); *Caenis* sp. (Insecta, Ephemeroptera); *Culicoides* sp. (Insecta, Diptera); *L. hoffmeisteri, Tubifex tubifex* (Müller, 1774) (Annelida, Oligochaeta), and *Erpobdella octoculata* (Linnaeus, 1758) (Annelida, Hirudinea) (**Tab. 1**; **Fig. 3**).

The study sites were grouped in the NMDS plot in two dimensions based on the similarities of their benthic macroinvertebrate community structures

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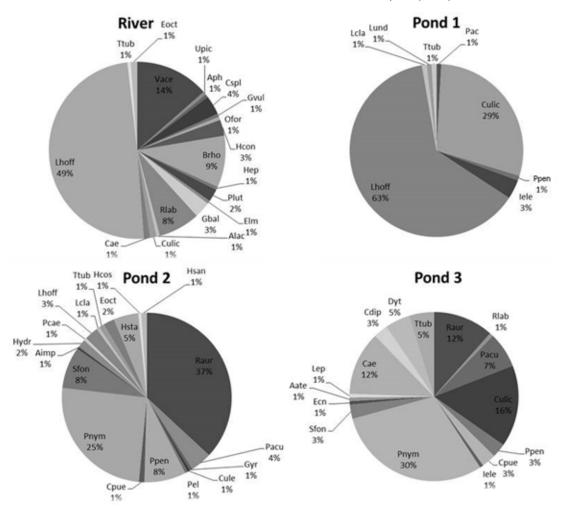


Fig. 2. Partitioning of the taxa recorded at the study ponds and the river by their abundances. For taxa name abbreviations see Tab. 1

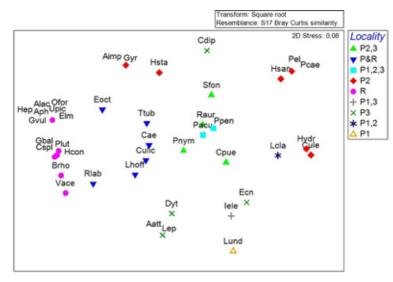
with stress value 0.06 (**Fig. 4**). The study sites from pond 1 and the river were grouped in the same cluster, while sites form pond 2 and pond 3 were clearly separated from the river and pond 1 (**Fig. 4**). The results of the ANOSIM analysis showed that benthic macroinvertebrate structures of the river and all three ponds were significantly different (R =0.901; p = 0.003).

# Discussion

As previously reported in the literature, ponds and rivers, as distinct freshwater habitats, usually support distinct invertebrate assemblages, even at the genusfamily level (Céréghino et al., 2008). This was the case in our study, since only 7 out of 43 recorded taxa were common for the river and the study ponds. Moreover, our results are similar to previous research showing that rivers make a higher contribution than ponds to the taxonomic richness of Ephemeroptera, Trichoptera, Plecoptera, and Mollusca, while ponds make a higher contribution to the taxa richness of Odonata, Heteroptera, and Coleoptera (Céréghino et al., 2008).

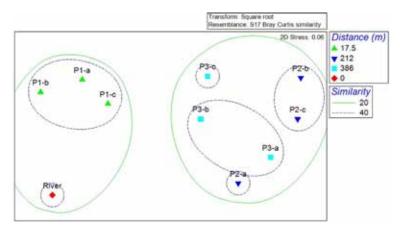
It has been previously shown that the species we recorded both in the river and in the ponds are generalists, inhabiting both lotic and lentic ecosystems. For instance, E. octoculata is usually dominant among leeches in both lotic and lentic habitats (Kubová et al., 2013). The snail R. labiata is adapted to inhabit both lotic and lentic environments by having larger and wider but shorter (globular) shells in lotic versus lentic habitats (Lam & Calow, 1988). Oligochaetes, such as L. hoffmeisteri and T. tubifex inhabit a wide variety of freshwater body types, and the latter one is known to tolerate pollution and can be found in very different conditions (Timm, 1999). Similarly, the biting midges from the genus Culicoides inhabit various aquatic, but also semiaquatic habitats (Mullen & Murphree, 2019).

On the other hand, most of the species we recorded exclusively in the river or in the study



**Fig. 3.** Non-metric multidimensional scaling (NMDS) plot showing spatial patterns of benthic macroinvertebrates from the river and study ponds. P and R denote pond and river respectively. For taxa name abbreviations see Tab. 1

ponds have been previously found to prefer to a certain extent either lotic or lentic aquatic habitats. This is especially true for dragonflies and damselflies (Odonata) whose clear preferences for specific habitat types are mainly determined by the presence of water flow, water regime, vegetation structure, and physicochemical water properties (Corbet, 1999). Species that were recorded in ponds in our study, such as: Coenagrion puella (Linnaeus, 1758), Ischnura elegans (Vander Linder, 1820) (Coenagrionidae), Sympetrum fonscolombii (Selys, 1840) (Libellulidae) and Anax imperator Leach, 1815 (Aeshnidae) typically inhabit standing waters (Rajkov, 2013). In contrast, the species we recorded in the river, such as: Calopterix splendens (Harris, 1780) (Calopterigidae), Gomphus vulgatisimus (Linnaeus,



**Fig. 4.** Non-metric multidimensional scaling (NMDS) plot showing similarities among the study sites based on their benthic macroinvertebrate community structures

1758) and Onychogomphus forcipatus (Linnaeus, 1758) (Gomphidae) almost exclusively inhabit running waters (Rajkov, 2013). Similarly, the mayfly species (Ephemeroptera) that we recorded in the river, such as Baetis rhodani (Pictet, 1843) and Potamanthus luteus (Linnaeus, 1767), are typical lotic species whose larval development is restricted to running waters (Eliot et al., 1988). The similar situation is with caddisflies (Trichoptera) recorded in our study. Namely, Hydropsyche contubernalis McLachlan, 1865, the species we recorded in the river, is regarded as a species of the large rivers (Edington & Hildrew, 1995). On the other hand, the distribution of the species we recorded in the ponds, such as Athripsodes aterrimus (Stephens, 1836), is determined by slow flowing water, fine sediments and presence

of macrophytes, and it is typical for standing waterbodies (Wallace et al., 1990; Previšić et al., 2010).

Although *L. hoffmeisteri* has low dispersal capacity, a possible reason for high abundance of this species both in the river and pond 1 could be its passive dispersion via river floods. It has been shown that an increase in water exchange by flooding events can increase exchange of macroinvertebrates (Siziba et al., 2011), and that passive dispersers can spread by direct water connections in flooded areas (Van Leeuwen et al., 2013). On the other hand, spatial proximity and connectivity of the ponds in our study were not crucial for structuring their benthic macroinvertebrate communities. The observed differences between macroinvertebrate

communities of pond 1 and pond 2, despite their connectedness and spatial proximity, suggest that local factors were more important for structuring benthic macroinvertebrates in the study ponds. For instance, similar depth or water quality could be the reasons for the observed similarity of benthic macroinvertebrate communities between pond 2 and pond 3, even though these ponds are separated by the nearby highway.

Overall, by taking into account that benthic macroinvertebrate communities of the river and all study ponds were statistically significantly different, it may be concluded that river proximity does not affect macroinvertebrate

communities in the study ponds and that Batušinac ponds contribute to overall freshwater biodiversity of the study area by supporting unique macroinvertebrate fauna.

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