

## *Aquatic Macroinvertebrates Diversity in the Upper Stretch of Una River (Una National Park, SW Bosnia and Herzegovina)*

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**Abstract.** The karstic Una River is one of the larger tributaries of the Sava River. Although being a part of Balkan biodiversity hot-spot, aquatic macroinvertebrates of the Una are rarely studied. The aquatic macroinvertebrates sampling was performed in the summer of 2015 throughout the upper river stretch covering the Una and its tributaries situated in the Una National Park (Una NP). A rich community consisting of 130 macroinvertebrate taxa, with dominance of Trichoptera and Ephemeroptera, was detected. A snail *Sadleriana* sp. and endemic gammarid *Echinogammarus acarinatus* were found to be dominant taxa in terms of percentage participation and frequency of occurrence. Mollusca and Crustacea were found to be the most abundant groups, with higher abundance in the Una River than its tributaries. Although a higher overall taxa richness was found in tributaries, calculated biotic indices and evenness pointed to greater diversity in the Una River. Additionally, BMWP and ASPT scores were also higher in the Una River than its tributaries. Calculated Jaccard index value pointed to moderate faunistic similarity between investigated communities. Obtained results suggest that investigated upper stretch of the Una River harbours a high diversity and rather homogenous macroinvertebrate community.

**Key words:** tributaries, benthic community, western Balkans.

### **Introduction**

On global scale aquatic macroinvertebrates are well studied component of aquatic animal diversity. Fairly diverse freshwater habitats in rivers

allow the existence of high variety of endemic, relict or rare macroinvertebrate species. In Europe, the Balkan Peninsula is recognized as one of the main hot-spot of animal diversity (Griffiths et al, 2004; Savic,

2008). It is particularly true in the case of its karstic parts, such as Dinaric region (Western Balkan), which harbours rich communities of aquatic macroinvertebrates (Previsic et al, 2009; Klobucar et al, 2013). The most recognizable macroinvertebrates representatives in Dinaric karstic rivers are aquatic insects (i.e. Matonickin & Pavletic, 1960b; Habdija et al, 1997; Radja & Puljas, 2008; Płóciennik et al, 2016; Savic et al, 2017; Vilenica et al, 2017).

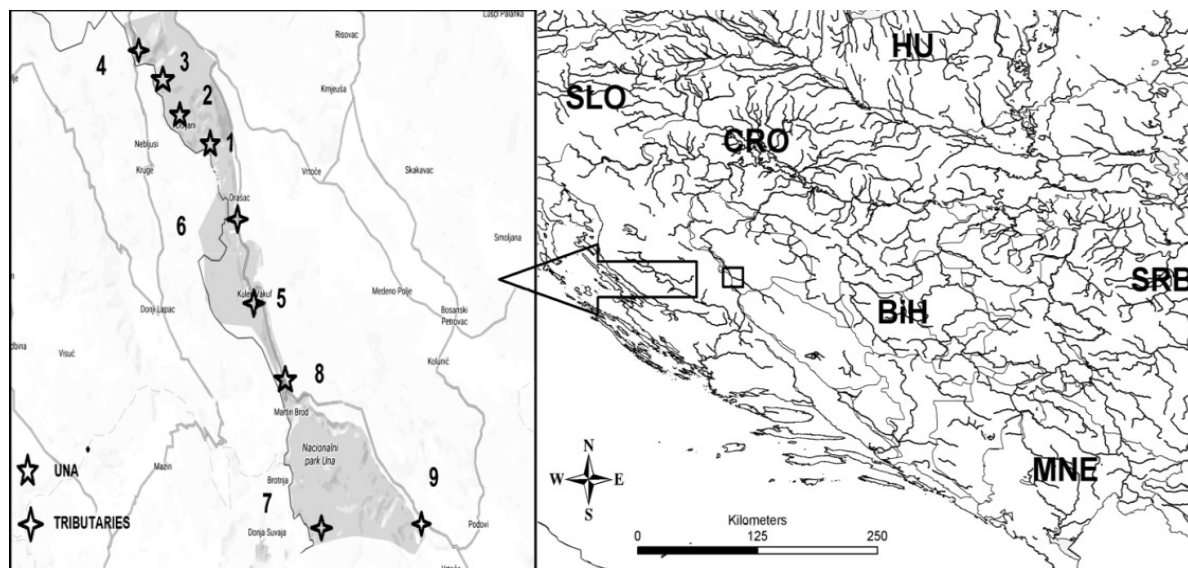
Una is karstic river, one of the largest tributaries of the Sava River. Although a part of biodiversity hot-spot, aquatic macroinvertebrates compositions of the Una, particularly its upper part are rarely studied. To date, apart from study of complete macroinvertebrates communities at few waterfalls (Matonickin & Pavletic, 1960b) researches were conducted with amphipods (Zganec et al, 2010; 2016), water quality assessment (Trozic-Borovac and Skrijelj, 2000; Kerovec et al, 2005; Nogic et al, 2016) and fish-diet (Trozic-Borovac, 2002).

Being aware of the gaps in published data we focused our research on the upper river stretch in the area which nowadays is part of the Una national park (located in Bosnia and Herzegovina), aiming to contribute to the knowledge of its aquatic macroinvertebrates fauna and to raise awareness of importance of preserving this Dinaric hot spot. Furthermore, we intend to assess overall aquatic macroinvertebrates diversity (based on biotic indices) and habitat quality (based on BMWP and ASPT scores) between the Una and its studied tributaries.

## Material and methods

### Study area

The upper Una stretch is situated in the south-western part of Bosnia and Herzegovina (Fig. 1). From the 2008, this area is under protection as a national park. Investigated area belongs to inland water ecoregion Dinaric Western Balkan (Illies, 1978).



**Fig. 1.** Investigated localities at the upper Una stretch (the Una NP): 1- Una (Strbacki Buk) N44°39'21.72" E 16° 0'39.53"; 2- Una-(Bjelila) N44°40'11.46" E15°58'49.48"; 3- Una (Loskun) N44°41'39.60" E15°57'29.05"; 4- Una (Martin Brod) N44°29'47.23" E16° 8'0.63"; 5- Djukanov potok N44°42'54.81" E15°56'3.38"; 6. Ostrovica N44°33'32.51" E16° 5'19.71"; 7. Toplica (Klisa) N44°35'45.25" E16° 3'59.78"; 8. Krka (Radjenovici) N44°25'21.34" E16° 8'38.72"; 9. Unac (Bastasi) N 44°23'41.69" E 16°19'39.08".

Situated in Dinarids, a karst mountain range, the Una River water is characterized by high alkalinity and carbonate solidity, harboring favorable conditions for formation of calcareous sinter, although due to lower water temperature this process is slower, excluding parts with better aeration (such as bigger (tectonic) slaps - Martin Brod and Strbacki buk) (Matonickin & Pavletic, 1960a; Bognar 2005).

The investigated upper part of the Una (Una national park - NP) is situated in not so inhabited area. Bottom sediments at sampling localities were characterized by dominance of lithal, mesolithal and psammal. Primary producers consisting of aquatic macrophytes mosses (Bryophyta) and epiphytic algae were common in localities along the Una, and also present in the Ostrovica Stream.

In total samples from 9 localities were taken, of which four were situated on the Una, while five were situated on various Una tributaries (Fig. 1).

Among five tributaries investigated, two were larger (the Unac and the Krka), and three were smaller streams, of which two (Ostrovica and Toplica) in Kulen Vakuf were with partially captivated springs (Dzankic et al., 2006).

#### *Sampling and data analysis*

The sampling was performed by the standard benthological hand-net (25x25 cm, 500 µm mesh size) in the Summer of 2015. One cumulative sample per locality was taken and multi-habitat procedure was applied (Hering, 2004). All samples were preserved with 60-80% ethanol solution and further processed in the laboratory. Identification of macroinvertebrates was done by using appropriate taxonomic keys.

The main physical and chemical parameters (temperature, dissolved oxygen, conductivity, pH) were measured by WTW Multiline IDS field set, while for an additional physical and chemical parameters (nitrates, nitrites, chlorides,

fluorides, total N, total P, Chemical oxygen demand (COD), total dissolved solids (TDS)), a 1L of water was taken at each site, preserved in cold chamber and transported in the laboratory of Biotechnical Faculty, Bihac, where further processing and measurements were done by applying standardized chemical protocols.

To better describe diversity in the Una River and its tributaries a following biotic indices were used: Shannon Wiener index (SWI; Shannon & Weaver, 1949), Evenness (Pielou, 1969; Hill, 1973) and Jaccard similarity coefficient ( $S_j$ ; Jaccard, 1908). The first two were calculated by ASTERICS software (version 3.1; AQEM, 2002), while Jaccard index was calculated manually by using formula  $S_j = a/(a + b + c)$ , where „a“ is number of species common to entire community, „b“ is number of species unique to the first community (Una River) and „c“ is number of species unique to the second community (tributaries). In order to assess water quality a BMWP and ASPT scores (Chester, 1980; Armitage et al, 1983) were calculated by ASTERICS (AQEM, 2002). Kruskal-Wallis nonparametric test (Kruskal & Wallis, 1952) was used to compare differences between calculated indices of the Una and tributaries. This analysis was performed in STATISTICA software ver. 7 (Statsoft, 2006).

#### **Results**

On Table 1 are presented values of some water parameters measured at localities of Una River and its tributaries (Table 1).

A rich community consisting of total of 130 macroinvertebrate taxa, with insects being the most diverse group (Trichoptera and Ephemeroptera with 27 taxa each; Fig. 2a) was recorded.

A higher taxa richness was found in tributaries compared to the Una River itself (108 vs 82 taxa), mainly due to higher number of taxa from groups Trichoptera, Ephemeroptera, Diptera and Odonata. On

the other hand, in the Una River a higher diversity of Mollusca was recorded, while regarding Oligochaeta and Crustacea, no notable difference in number of taxa between the Una River and the tributaries was found.

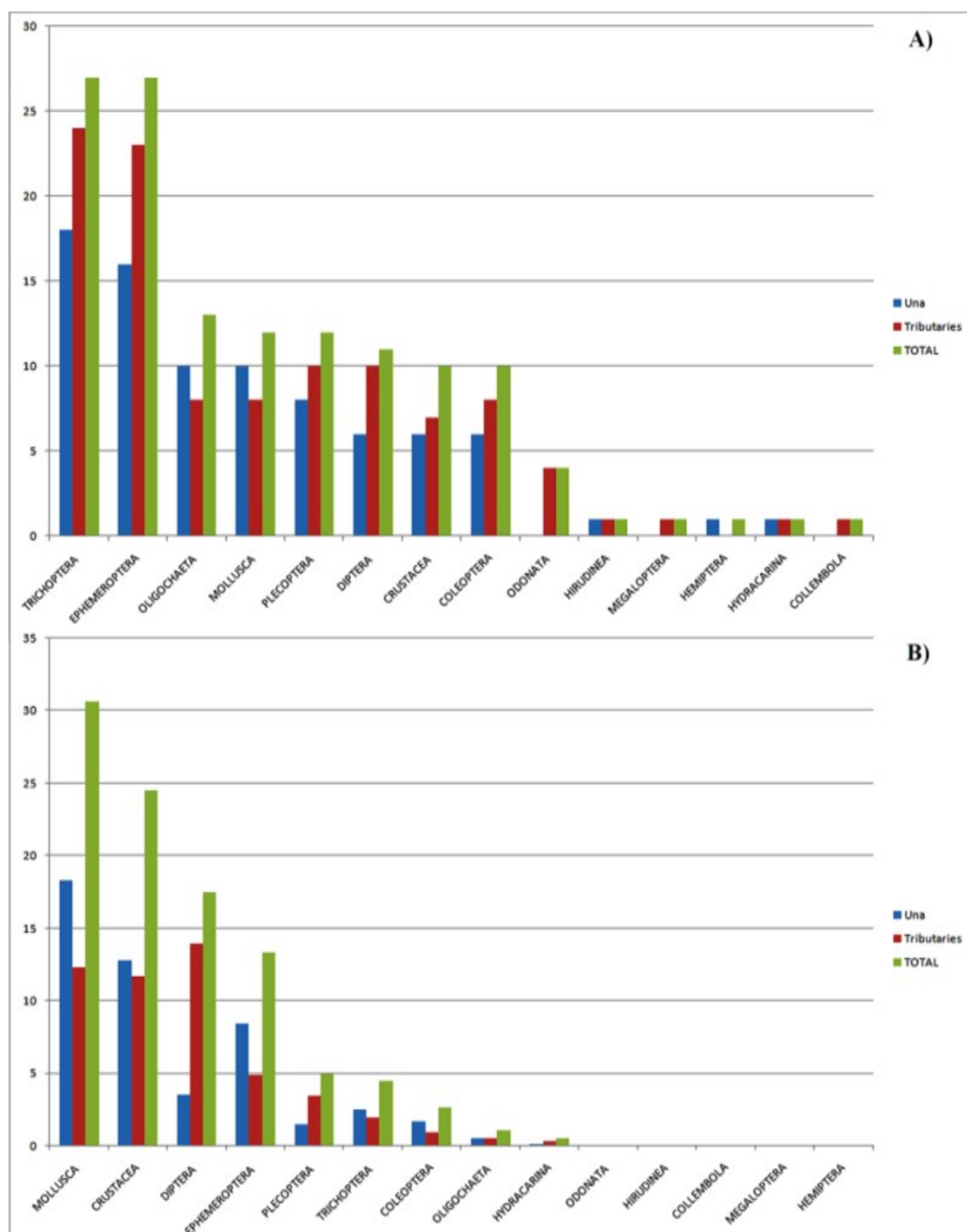
Regarding calculated biotic indices (H and Evenness) a somewhat different situation occurred, with higher values of these indices in the Una River than in its tributaries (Fig. 3a,b). However, the performed nonparametric test (Kruskal-Wallis) showed that found differences were not statistically significant (at the  $p=0.05$  level). The obtained value of the Jaccard index ( $S_j=0.41$ ) pointed to moderate similarity between tested communities (Samples from the Una River and samples from its tributaries. Values of

BMWP and ASPT indices (Fig. 3c,d) were higher and more variable in the Una River than its tributaries. In respect to abundances/relative percentage of community, somewhat different picture arises (Fig. 2b). Mollusca and Crustacea were found to be the most abundant groups overall. Diptera, the third group regarding abundances overall, was the most abundant group in the tributaries, followed by the Mollusca and Crustacea.

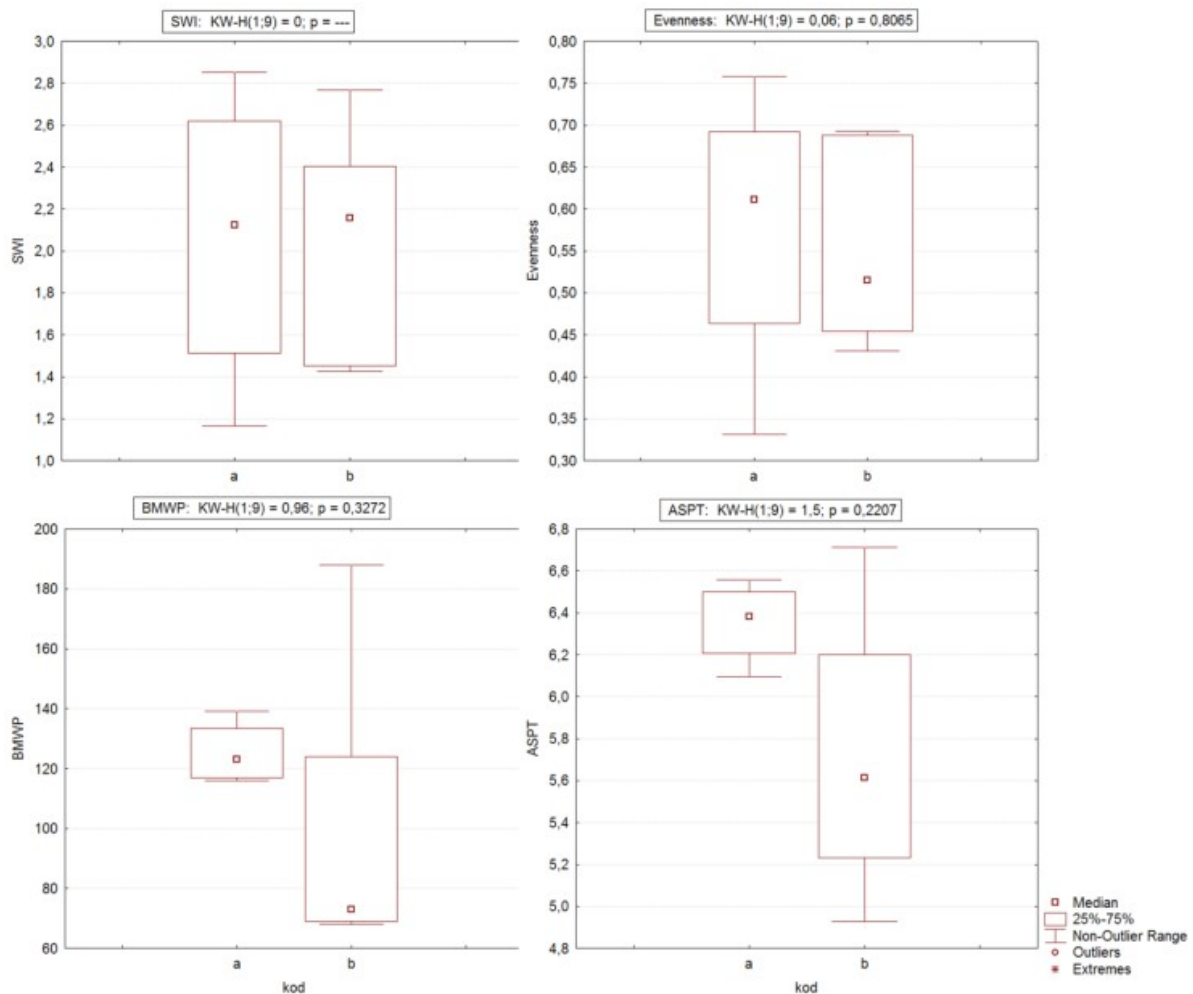
A snail *Sadleriana* sp. was the dominant component of recorded macroinvertebrates community overall. It was omnipresent (found in all samples) and occupied 28% of the overall community recorded during our investigation. The highest abundance was recorded in samples 1 (78% of total community) and 4 (52%).

**Table 1.** Main values of selected physico-chemical parameters.

UNA	MU	Minimum	Maximum	Median	Mean	SD (n-1)	SE of the mean	Median AD
Temperature	°C	10.9000	11.7000	11.1500	11.2250	0.3403	0.1702	0.1500
pH		7.7500	8.0400	7.8200	7.8575	0.1271	0.0636	0.0450
Conductivity	µS	328.0000	412.0000	392.0000	381.0000	36.7967	18.3984	12.5000
COD	mgO <sub>2</sub> /l	3.2000	7.2000	6.4500	5.8250	1.8410	0.9205	0.6500
Total P	mg/l	0.1400	0.4800	0.2200	0.2650	0.1484	0.0742	0.0450
Sulphates	mg/l	26.1530	35.1160	30.9055	30.7700	4.3230	2.1610	3.5120
Nitrates	mg/l	0.8410	2.0360	1.0300	1.2343	0.5419	0.2709	0.0975
Chlorides	mg/l	4.1270	5.2740	4.8835	4.7920	0.5605	0.2803	0.3725
Fluorides	mg/l	0.2570	0.4280	0.3265	0.3345	0.0792	0.0396	0.0570
TRIBUTARIES	MU	Minimum	Maximum	Median	Mean	SD (n-1)	SE of the mean	Median AD
Temperature	°C	9.4000	11.3000	11.2000	10.6000	0.8860	0.3962	0.1000
pH		7.2500	7.9600	7.6400	7.5980	0.2769	0.1238	0.2300
Conductivity	µS	324.0000	386.0000	354.0000	354.4000	21.9727	9.8265	2.0000
COD	mgO <sub>2</sub> /l	2.9000	6.2000	3.3000	3.8000	1.3601	0.6083	0.2000
Total P	mg/l	0.0100	0.3600	0.0310	0.1302	0.1609	0.0720	0.0210
Sulphates	mg/l	21.3050	36.3490	22.7830	26.5270	6.4210	2.8710	1.4780
Nitrates	mg/l	0.1260	1.7640	0.7620	0.687	0.941	0.420	0.6360
Chlorides	mg/l	5.0430	8.0920	6.0340	6.2734	1.1776	0.5266	0.6160
Fluorides	mg/l	0.2130	0.3150	0.2310	0.2408	0.0424	0.0190	0.0170



**Fig. 2. (a)** Taxa richness and **(b)** percentage participation of main taxonomic groups recorded at the upper Una stretch; x axis - main taxonomic groups, y axis - number of taxa (a) and percentage participation of taxa in the sample (b).



**Fig. 3.** A comparison of values of calculated indices of diversity (SWI), evenness, BMWP and ASPT between the Una River (code a) and its tributaries (code b).

On the other hand, in samples 2 and 6, this gastropod was represented with only 4% and 2% in the community, where in, the highest abundance of the *Echinogammarus accarinatus* (17% of overall community, with frequency of occurrence (F) of 0.89) was observed. Both of these dominant taxa overall were more abundant in the Una (than in tributaries) where they were comprised 35% and 20% of the community (versus 21% and 13% in tributaries), respectively.

Among the Diptera fauna the most dominant was the group Chironomidae (17% of overall community). These dipteres were more abundant in tributaries, particularly in samples 7 and 4, where they consist 58% and 29% of the total community.

The found *Niphargus* spp. group probably consisted of two taxa, one of which could be a hypogean, while the other one based on morphological characters could belong to one of a few epigean niphargids, apart from *Niphargus hrabei*, S. Karaman, 1932 and *Niphargus valachicus*, Dobrea and Manolache, 1933. It should be mentioned that in the same samples with these *Niphargus* specimens, an abundant gammarid population (dominated by *E. accarinatus* and *G. balcanicus*) was recorded. In our research *E. accarinatus* was dominant amphipod and one of the most frequent species. Especially abundant it was in the sample 8 (the Una downstream Martin Brod). Among coleopterans, *Limnius*

*volckmari* (both adults and larvae) was the most numerous/abundant taxon. In the main stream/Una it was much more present and numerous than in tributaries. Other numerous and common beetle found was *Elmis* sp. (possibly *E. bosnica*, but due to lack of taxonomic keys for larvae it could not be identified to species level) and *E. latreillei* were evenly numerous in Una/tributaries. Finally *Esolus angustatus*, one of common coleopterans recorded in this investigation was more abundant in tributaries. Among Oligochaeta naids *Nais* sp./*N. bretscheri* were more abundant in tributaries than in the main course of the Una. Regarding molluscs, *Sadleriana* sp. was found to be dominant taxon in this investigation, as mentioned before. Other molluscs were less numerous, and except *Bythinella* sp. and *Pseudosuccinea columella* were more common in the Una River itself. In case of succinid this could be explained by the fact that they are mostly semiaquatic and commonly found in waters with dense riparian vegetation, as in case of a smaller Una tributaries here. Although mayflies were more abundant in the Una, in tributaries a greater diversity was recorded. Among more common (abundant and frequent) taxa here only *B. rhodani* was evenly numerous and frequent in Una and tributaries. A more common presence of *Ephemerella ignita* in the Una (than in tributaries) was recorded.

### Discussion

High macroinvertebrates taxa richness found (130 taxa) and the dominance of insect could be expected for mountainous rivers in the region (Habdija et al, 1997; Radja & Puljas, 2008). Diversity in karstic rivers/streams is influenced by specificity of habitats and geological (limestone) substrate (Radja & Puljas, 2010). A higher taxa richness found in tributaries was due to higher number of taxa from groups Trichoptera, Ephemeroptera, Diptera and Odonata. Higher abundances of mollusks and crustaceans recorded in the Una, could be expected due to more stable

environmental conditions, which favor these fully aquatic groups. An explanation, mentioned in case of the diversity of groups (regarding its presence in the Una vs tributaries) could be implemented here, as well - Diptera, as insects distinguish shorter aquatic life-phase and thus they should be more adapted to less stable habitat conditions, especially in case of streams running dry (which is common in case of karstic, especially smaller watercourses). An occasionally running dry is common characteristic of some smaller karstic watercourses (Matonickin et al, 1972). In our investigation intermittent watercourses were present (localities 4, 5 and 6), though during our investigation those were not dry, yet with a fairly low water levels (personal communication). Unstable conditions inevitable cause adaptation of its biota (macroinvertebrates included), leading to higher presence of more adapted taxa, such as insects with semiaquatic shorter life cycles and faster colonization abilities (Williams & Feltmate, 1992). Additionally, more pronounced differences in physical and chemical water conditions present in tributaries could influence found macroinvertebrates species, as well (Dzankic et al, 2006). Although, higher diversity (reflecting as taxa richness and H index) was present in tributaries, a higher values of the evenness, along with higher values of BMWP and ASPT indices, pointed to importance of more stable water conditions, which allow more persistent habitat variety and simultaneously more suitable environmental condition for aquatic macroinvertebrates communities in such karstic environment.

In respect to abundances/relative percentage of investigated community, a different picture arised, with Mollusca and Crustacea found to be the most abundant macroinvertebrates. However in tributaries, Diptera (the third most abundant group overall) was the most abundant group, followed by Mollusca and Crustacea. Higher abundances of mollusks and crustaceans recorded in the Una/mainstream, could be

expected due to more stable environmental conditions, which should favor these fully aquatic groups (for example Mollusca). An explanation, mentioned in case of the diversity of groups (regarding its presence in the Una vs tributaries) could be implemented here, as well – Diptera, as not entirely aquatic insects and usually with relatively short aquatic phase should be more adapted to more variable habitat conditions (Williams & Feltmate, 1992), especially in a case of streams running dry, which is common in case of karstic, especially smaller, watercourses (Bonacci et al, 2013).

A snail *Sadleriana* sp. was omnipresent and dominant component of recorded macroinvertebrates community, followed by an endemic gammarid *Echinogammarus acarinatus*. *E. accarinatus* is a rather small (up to 10.5 mm max length) endemic amphipod of western karstic Balkan (Karaman, 1970; Pinkster 1993; Zganec et al, 2010; 2016). Although it is considered as mesohaline species (Stock, 1968) it apparently could well adapt to oligohaline conditions. The species *Gammarus balcanicus* is dominant gammarid in colder Croatian waters (below 20 °C; Zganec, 2009; 2010), but in our investigation although it was present and frequent it was not abundant as previously mentioned *E. accarinatus*.

The finding of *Niphargus* spp. coexisting with rich and abundant gammarid population is particularly interesting. Fisher et al. (2007) analyzed the coexistence of two species (one niphargid and one gammarid) in Slovenia and recorded dominance of gammarids in the mainstream/permanent stream and the presence of niphargids in the fissure system where niphargid species could survive desiccations and gammarid competition. A one endemic gammarid present in the investigated region (*Fontogammarus dalmatinus*) could not be confirmed in our investigation. A recent investigation (Zganec, 2016) states presence of this endemic gammarid species, only in the most upper Una part (spring and

upstream Martin Brod), along with *Echinogammarus accarinatus* which was the most common gammarid in this investigation, so our findings could indicate somewhat wider distribution and more abundant presence of *E. accarinatus* in this region.

The taxa which were not identified to lower/species level, particularly among Diptera and Gastropoda, could hide some interesting and endemic taxa as well.

The investigated upper part of the Una River (Una NP) harbours high aquatic macroinvertebrates diversity, with few rare taxa. A pronounced difference in community structure between tributaries and the Una itself could not be confirmed, although tributaries were more taxa rich, mainly due to the greater diversity of smaller insects such as caddisflies and mayflies. A continued research should shed more light on aquatic macroinvertebrates and its community structure in this protected part of the Una River.

#### **Acknowledgements**

The authors would like to thank Zeljko Mirkovic Grga for valuable help during field work. This study was supported by grant 451-03-9/2021-14/200178 of the Ministry of Education, Science and Technological Development of Serbia.

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Received: 13.04.2021  
Accepted: 28.05.2021