

## New data on the distribution and ecology of the mayfly larvae (Insecta: Ephemeroptera) of Serbia (central part of the Balkan Peninsula)

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**Abstract:** This work is a contribution to the knowledge of the Ephemeroptera (Insecta) fauna of the central Balkan Peninsula. A total of 85 mayfly species (31 genera and 12 families) were reported. The largest number of mayfly species was recorded within the Južna and Zapadna Morava river basins, while the lowest taxa richness was observed within the Sava basin. The highest diversity was observed in hilly mountainous regions due to the general environmental requirements of the majority of mayfly taxa, as well as the distribution of anthropogenic stress. Lowland areas have been more exposed to different stress factors that could reduce mayfly taxa richness in comparison to the hilly mountainous region located south of the Danube and Sava rivers. Compared with species richness in neighbouring countries, the expected mayfly diversity for Serbia is certainly higher; it has been estimated that over 100 taxa should occur in different types of aquatic ecosystems.

**Key words:** Ephemeroptera, mayfly larvae, distribution, Serbia

### 1. Introduction

Due to the increase in anthropogenic impact on surface water resources, the main challenge for decision-makers in river management is to restore and maintain the health of river ecosystems (Norris and Thoms, 1999). This is especially true for priority watercourses with rare species and high species richness, where additional effort for their protection is needed (Lock and Goethals, 2011). It is generally recognised that Ephemeroptera larvae are commonly used as water-quality indicators in determining such priority water ecosystems. Mayflies are one of the most diverse groups of macrozoobenthos, occupying a wide variety of niches (Merritt and Cummins, 1996). Their high sensitivity to environmental changes makes them a prime candidate for water quality assessment (Lenat, 1988; Metcalfe, 1989; Kerans and Karr, 1994). Knowledge of mayfly diversity and distribution patterns is the first step for their inclusion in bioassessment programs.

Global species diversity of Ephemeroptera is represented by over 3000 described species distributed in 42 families and 400 genera (Barber-James et al., 2008). Studies presenting the diversity and distribution pattern of lotic systems mainly provide data about mayflies in

Western and Central Europe (Grandi, 1960; Belfiore, 1983, 1994; Belfiore and D'Antonio, 1990, 1991; Belfiore et al., 1992; Zabrcic and Sartori, 1997; Bauernfeind, 2003; Buffagni et al., 2003; Derka, 2003; Haybach and Malzacher, 2003; Ruginis, 2006; Zahrádková et al., 2009). However, the Balkan Peninsula has been a relatively unexplored area, where previous research on the mayfly fauna has been conducted only on particular rivers (Klapalek, 1898, 1906; Živojinović, 1950; Filipović, 1954, 1968, 1969, 1975, 1976, 1979; Ikononov, 1960, 1962; Marković and Janković, 1989; Marković, 1995; Marković and Mitrović-Tutundžić, 1997; Paunović et al., 1997; Marković, 1998; Paunović et al., 1999; Simić and Simić, 1999; Marković and Živić, 2002; Simić and Simić, 2003; Paunović et al., 2006a; Savić et al., 2011).

It seems that the diversity of macroinvertebrates for the central Balkan Peninsula is extremely high because the lotic systems in this area belong to the drainage basins of 3 seas (Black Sea, Adriatic Sea, and Aegean Sea). Due to its central position in the Balkan Peninsula, Serbia is characterised by high diversity concerning overall abiotic factors (diverse climate, petrographic and pedological variety, orographic characteristics) (Radović et al., 1995).

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It has a great heterogeneity of watercourses in terms of elevation stream orders and habitat diversity, which consequently affects the diversity of aquatic insects in the central Balkan Peninsula. For the purpose of better knowledge of the Ephemeroptera community, their diversity and distribution patterns were investigated for 10 drainage basins that encompass the diversity of the central Balkan Peninsula. In addition, we present here the first checklist of Ephemeroptera for the central Balkan Peninsula, with notes on ecology and IUCN Red List categories at a regional level.

## 2. Materials and methods

### 2.1. Study area

The Ephemeroptera review presented in this work covers the territory of the Republic of Serbia. Serbia is a landlocked country located in south-eastern Europe, covering the central part of the Balkan Peninsula and partially covering Central Europe (the southern part of the Pannonian Plain). All surface waters in Serbia belong to the drainage basins of 3 seas: the Black Sea, Adriatic Sea, or Aegean Sea.

Practically the entire territory of Serbia (92%) belongs to the Danube (Black Sea) drainage basin (81,700 km<sup>2</sup>) (Gavrilović and Dukić, 2002). The entire basin is drained by only one river, the Danube, which flows into the Black Sea. All major rivers in Serbia belong to it, including the Sava, Velika, Zapadna and Južna Morava, Kolubara, Timok, and Drina.

The Adriatic Sea drainage basin covers an area of 4732 km<sup>2</sup>, or 5.36% of the territory of Serbia (Gavrilović and Dukić, 2002). It comprises the western half of Kosovo and Metohija, and it is mostly drained by one river, the White Drin, which in Albania meets the Black Drin to create the Drin River, which flows into the Adriatic Sea. However, these rivers have not been investigated recently due to the current political situation in Serbia.

The Aegean Sea basin is the smallest in the area (1926 km<sup>2</sup>, which is approximately 3% of the territory of Serbia), and covers the southern parts of Serbia to the Macedonian and Bulgarian borders. It is drained by 3 rivers: the Lepenac, the Pčinja, and the Dragovištica.

Aquatic ecosystems where the Ephemeroptera species have been found were classified into 1 of 3 drainage areas existing in the territory of Serbia. In addition, the aquatic ecosystems were divided into the following, according to the size of the catchment area (near or above 4000 km<sup>2</sup>): Dunav, Sava, Kolubara, Velika, Zapadna and Južna Morava, Timok, Drina, and rivers belonging to the Aegean Sea basin (Pčinja, Lepenac, and Dragovištica). We followed this classification in presenting the checklist of Ephemeroptera. Furthermore, in this paper, we classified sampling points in accordance with elevation gradient,

expressed as 4 a priori defined categories: 1. lowland (up to 200 m a.s.l.); 2. submontane (200–500 m a.s.l.); 3. mountain (500–800 m a.s.l.); and 4. highland (over 800 m a.s.l.) (SCG ICPDR National Report, 2004; Paunović et al., 2005a, 2005b).

### 2.2. Sampling and data analysis

To construct the checklist and to investigate the distribution and ecology pattern of mayflies, the results of the long-term investigation of Ephemeroptera diversity in Serbia from the informative database BAES ex situ, at <http://baes.pmf.kg.ac.rs> (Simić et al., 2006), were used. BAES ex situ was developed as a result of biodiversity investigation into surface waters in Serbia, within the scope of the project "Ex situ biodiversity protection of aquatic ecosystems of Serbia".

This database presents a new, innovative relational model that, using the appropriate tools, enables monitoring of aquatic species data (macroalgae, macroinvertebrates, and fish) found in surface waters in Serbia from 1860 until now. BAES ex situ consists of 2 types of data: 1) literature data since 1950, and 2) data resulting from our field research during the period 2003–2011. The data for each species are presented as follows: taxonomy of species, ecosystem where this species has been found, year of the particular record, frequency of each record, abundance, and other literature data.

In this study, we have presented the distribution and diversity of the Ephemeroptera community according to data for the period 1950–2011. We used only presence/absence data here, since BAES ex situ has been created from studies with unknown or different sampling methods.

Our field research was conducted during the period 2003–2011. Benthic samples were taken with a 0.0625 m<sup>2</sup> Surber sampler of 250- $\mu$ m mesh or Eckman dredge, depending on the aquatic ecosystem type. Ephemeroptera larvae were separated from the remainder and identified up to the genus or species level with established keys (Ikononov, 1959; Belfiore, 1983; Elliott et al., 1988).

According to Schmedtje and Colling (1996) and AQEM classification (AQEM, 2002), database feeding type, current preference, and temperature range preference were defined for each Ephemeroptera species.

In addition, to determine the level of threat, we used the IUCN 2012 IUCN Red List Categories and Criteria, version 2012.2. Finally, conservation status was estimated for those taxa for which precise and accurate data were available, and whose population status had been revised.

## 3. Results

Based on the analysed data, a total of 85 species distributed into 31 genera and 12 families were recorded in Serbia. The most represented family is Heptageniidae (27), followed by Betidae (24) species. The following families have only 1

species present: Ameletidae, Palingeniidae, Potamanthidae, Siphonuridae, and Polymitarciidae. *Baetis* and *Ecdyonurus* are the most diverse mayfly genera at the species level with 14 and 12 recorded species, respectively. The rest of the genera are represented by fewer species (fewer than 10). During this investigation of lotic systems in Serbia, 2 species were recorded for the first time: *Epeorus yougoslavicus* (Samal, 1935) (Petrović et al., 2006) and *Choroterpes picteti* (Eaton, 1871) (Simić et al., 2005).

The distribution pattern is presented in Table 1. When species richness was estimated by river basins, the Južna Morava (57 taxa) and Zapadna Morava (53) had the highest values. A significantly high number of species was also recorded in the Kolubara (46) and Timok (42) catchments. Species richness decreased in the Danube's small direct tributaries (28 taxa), the Aegean Sea basin (27), Drina basin (23), Velika Morava (20), Danube's main course and flooded zone (8), Sava (8), and Tisa (4).

**Table 1.** Ephemeroptera larvae of the Republic of Serbia: distribution of species within the main river and the elevation class.

River basin											
Species	Danube: small direct tributaries	Danube main course and flooded zone	Sava	Tisa	Kolubara	Velika Morava	Južna Morava	Zapadna Morava	Timok	Drina	Aegean Sea
Ameletidae											
<i>Ameletus inopinatus</i> Eaton, 1887							4	4			
Baetidae											
<i>Acentrella sinaica</i> Bogoescu, 1931							4				
<i>Alainites muticus</i> (Linnaeus, 1758)	2				2		4,2,1	4,3,2	4,3,2	2	
<i>Baetis alpinus</i> (Pictet, 1843)					2	2	4,2	4,3,2	4,3,2	2	2
<i>Baetis fuscatus</i> (Linnaeus, 1761)	2,1	1			2	2,1	4,2,1	4,3,2,1	4,3,2	2	4,2
<i>Baetis buceratus</i> Eaton, 1870			1		1		1	2		1	2
<i>Baetis kozufensis</i> Ikononov, 1962							2				2
<i>Baetis liebenauae</i> Keffermüller, 1974									3		
<i>Baetis lutheri</i> Müller-Liebenau, 1967					2		4	3,2	4,3,2		
<i>Baetis melanonyx</i> (Pictet, 1843)							4,3	4			4
<i>Baetis meridionalis</i> Ikononov, 1954	2						4,2				2
<i>Nigrobaetis niger</i> (Linnaeus, 1761)							4,2	4			
<i>Baetis pavidus</i> Grandi, 1949					2		2		4,2		
<i>Baetis rhodani</i> (Pictet, 1843)	2,1		1	1	2,1	2,1	4,2	4,3,2	4,3,2,1	4,2	3,2
<i>Baetis scambus</i> Eaton, 1870								4			4
<i>Baetis tracheatus</i> Keffermüller and Machel, 1967					2		2		4		
<i>Labiobaetis tricolor</i> Tshernova, 1928	2						3			3	3,2
<i>Baetis vardarensis</i> Ikononov, 1962					3		2	2	3,2		2
<i>Baetis vernus</i> Curtis, 1834					2	2,1	4,2	4,2	4,2,1		
<i>Centroptilum luteolum</i> (Müller, 1776)					1	2,1	4,2	4	2		
<i>Cloeon dipterum</i> (Linnaeus, 1761)	2	1			1	2,1		4,2			

Table 1. (Continued).

Species	Danube: small direct tributaries	Danube main course and flooded zone	Sava	Tisa	Kolubara	Velika Morava	Južna Morava	Zapadna Morava	Timok	Drina	Aegean Sea
<i>Cloeon simile</i> Eaton, 1870						2		4			
<i>Procloeon bifidum</i> (Bengtsson, 1912)							4				
<i>Procloeon macronyx</i> (Kluge and Novikova, 1992)	1	1									
<i>Procloeon pennulatum</i> (Eaton, 1870)									4		
Caenidae											
<i>Brachycercus harrisella</i> Curtis, 1834				1							
<i>Caenis horaria</i> (Linnaeus, 1758)	2,1	1		1	2	1	2	4,2	4,2		
<i>Caenis lactea</i> (Burmeister, 1839)					2						
<i>Caenis macrura</i> Stephens, 1835	2				2		2	4,2	2		2
<i>Caenis luctuosa</i> (Burmeister, 1839)			1		2		3,2		2		
<i>Caenis pseudorivulorum</i> Keffermüller, 1960					2						
<i>Caenis pusilla</i> Navás, 1913		1					2				
<i>Caenis rivulorum</i> Eaton, 1884							2	2			
<i>Caenis robusta</i> Eaton, 1884	1						1				
Heptageniidae											
<i>Ecdyonurus austriacus</i> Kimmins, 1958					1		3,2		3		
<i>Ecdyonurus dispar</i> (Curtis, 1834)	1		1		2		4,2	4,3,2	3,2	3	
<i>Ecdyonurus aurantiacus</i> (Burmeister, 1839)	1						4	4,2	2	3,2	
<i>Ecdyonurus epeorides</i> Demoulin, 1955	1						4,3,2	4	4		4,2
<i>Ecdyonurus forcipula</i> (Pictet, 1843)			1		2,1		1	2	3,2,1		
<i>Ecdyonurus helveticus</i> (Eaton, 1885)							2	4,3	3,2		
<i>Ecdyonurus insignis</i> (Eaton, 1870)	1,2				2	2,1	3,2,1	3,2	3,2,1	3	2
<i>Electrogena quadrilineata</i> Landa, 1970					1						
<i>Ecdyonurus subalpinus</i> Klapálek, 1907										2	
<i>Ecdyonurus submontanus</i> Landa, 1969					3						
<i>Ecdyonurus torrentis</i> Kimmins, 1942					4,3			4	3		3
<i>Ecdyonurus venosus</i> (Fabricius, 1775)	2,1		1		2	2,1	4,2	4,3	4,2		3,2
<i>Ecdyonurus zelleri</i> (Eaton, 1885)								4			
<i>Electrogena affinis</i> (Eaton, 1883)					2			4			
<i>Electrogena lateralis</i> (Curtis, 1834)	1				1			2			
<i>Electrogena macedonica</i> (Ikononov, 1954)	1							2			

Table 1. (Continued).

River basin											
Species	Danube: small direct tributaries	Danube main course and flooded zone	Sava	Tisa	Kolubara	Velika Morava	Južna Morava	Zapadna Morava	Timok	Drina	Aegean Sea
<i>Epeorus yougoslavicus</i> (Samal, 1935)							4	4			
<i>Epeorus assimilis</i> Eaton, 1885					2	3	4,1	4,2,1	4,3	3,2	2
<i>Dacnogenia coeruleans</i> Rostock, 1878					2						
<i>Heptagenia flava</i> Rostock, 1878	1										
<i>Heptagenia longicauda</i> (Stephens, 1835)								4			
<i>Heptagenia sulphurea</i> (Müller, 1776)	1				2,1	1	3,2,1	2			
<i>Kageronia fuscogrisea</i> (Retzius, 1783)					2			2			
<i>Rhithrogena beskidensis</i> Alba-Tercedor and Sowa, 1987							3,2	4	3,2,1	3	2
<i>Rhithrogena germanica</i> Eaton, 1885							2				
<i>Rhithrogena fiorii</i> Grandi, 1953					3				4	4	
<i>Rhithrogena semicolorata</i> (Curtis, 1834)					3,2	3	4,2	4,3,2	4,3	4	4
Ephemeroidea											
Ephemeridae											
<i>Ephemera danica</i> Müller, 1764	1				3,2	2	4,2	4,2	4,3,2	4	
<i>Ephemera glaucops</i> Pictet, 1843							4				
<i>Ephemera hellenica</i> Demoulin, 1955							4,2	4	3		3,2
<i>Ephemera lineata</i> Eaton, 1870					2		2				
<i>Ephemera vulgata</i> Linnaeus, 1843					2		2	3	3	4,3	
Ephemerellidae											
<i>Ephemerella mukronata</i> Bengtsson, 1909					2			4			
<i>Ephemerella notata</i> Eaton, 1887						2,1	4,2,1	4,2	3,2	4	2
<i>Ephemerella ikonovici</i> Puthz, 1971							4,2	4	2	4	2
<i>Serratella ignita</i> (Poda, 1761)	1	1	1		2	2,1	4,2	4,2	4,2	4,2	3,2
<i>Torleya major</i> (Klapálek, 1905)					2		2	3,2	1		2
<i>Choroterpes picteti</i> (Eaton, 1871)								3			
Leptophlebiidae											
<i>Habroleptoides modesta</i> (Hagen, 1864)	1				2		2	4,2	4,2,1	4	1
<i>Habroleptoides confusa</i> Sartori and Jacob, 1986							4	4		3	
<i>Habrophlebia fusca</i> (Curtis, 1834)	1				2	2	2	4,2	4,2		
<i>Habrophlebia lauta</i> McLachlan, 1884	1				2		2	4,2	3,2		
<i>Paraleptophlebia cincta</i> (Retzius, 1783)					2		4,2			4	

**Table 1.** (Continued).

River basin											
Species	Danube: small direct tributaries	Danube main course and flooded zone	Sava	Tisa	Kolubara	Velika Morava	Južna Morava	Zapadna Morava	Timok	Drina	Aegean Sea
<i>Paraleptophlebia lacustris</i> Ikonomov, 1962	1										
<i>Paraleptophlebia submarginata</i> (Stephens, 1835)	1				2	2	4,2	4,2	3	4,3	2
<b>Oligoneuriidae</b>											
<i>Oligoneuriella pallida</i> (Hagen, 1855)					2						2
<i>Oligoneuriella rhenana</i> (Imhoff, 1852)					2	2	4,2	4,2	4,3,2		3,2
<b>Palingeniidae</b>											
<i>Palingenia longicauda</i> Olivier, 1791				1							
<b>Potamanthidae</b>											
<i>Potamanthus luteus</i> (Linnaeus, 1767)	1	1	1			2	3,2	2	3,2,1		
<b>Siphonuridae</b>											
<i>Siphonurus aestivalis</i> Eaton, 1903							1		2		
<b>Polymitarcyidae</b>											
<i>Ephoron virgo</i> (Olivier, 1791)	1	1									
<b>Total number species</b>	<b>28</b>	<b>8</b>	<b>8</b>	<b>4</b>	<b>46</b>	<b>20</b>	<b>57</b>	<b>53</b>	<b>42</b>	<b>23</b>	<b>27</b>

1. Lowland (up to 200 m a.s.l.); 2. submontane (200–500 m a.s.l.); 3. mountain (500–800 m a.s.l.); 4. highland (over 800 m a.s.l.).

Mayfly fauna were recorded in all 4 types of elevation classes (Table 1). Species richness as a function of elevation gradient had the following pattern: 38 for the first class, 62 for the second class, 37 for the third class, and 52 for the fourth class.

The number of species per family per river basin and elevation class is presented in Tables 2a and b. The most diverse families, Heptageniidae and Baetidae, were present with the highest number of species in the Zapadna Morava (19) and Južna Morava (18), respectively. Considering the elevation gradient, Baetidae and Heptageniidae were the most diverse in submontane regions (200–500 m a.s.l.; see Table 2b).

Out of 86 species recorded in this study, 13 species were found at a single site: *Baetis liebenauae* Keffermüller, 1974; *Brachycercus harrisella* Curtis, 1834; *Caenis pseudorivulorum* Keffermüller, 1960; *Electrogena quadrilineata* Landa, 1970; *Ecdyonurus submontanus* Landa, 1969; *Ecdyonurus zelleri* (Eaton, 1885); *Ephemera glaucops* Pictet, 1843; *Heptagenia flava* Rostock, 1878;

*Heptagenia longicauda* (Stephens, 1835); *Choroterpes picteti* (Eaton, 1871); *Rhithrogena germanica* Eaton, 1885; *Paraleptophlebia lacustris* Ikonomov, 1962; *Palingenia longicauda* Olivier, 1791.

Information on ecological guilds is presented in Table 3. According to our data, the most common feeding guilds were grazers–scrapers (typically for the genus *Rhithrogena*, and the species *Epeorus assimilis* Eaton, 1885 and *Acentrella sinaica* Bogoescu, 1931) and gatherers–collectors (e.g., Caenidae and Leptophlebiidae). Representative species for other feeding types included some species of family Siphonuridae and Ephemerellidae (shredders), and *Ephemera glaucops* Pictet, 1843 and *Ephemera vulgata* Linnaeus, 1843 as active and *Oligoneuriella rhenana* (Imhoff, 1852) as passive filter feeders.

As for current velocity preferences (Table 3), we considered the following categories: RB (rheobiont, e.g., Baetidae and Heptageniidae); RP (rheophil); RL (rheo- to lithophil, e.g., *Ameletus*, *Baetis*, *Centroptilum*); LR (limno- to rheophil, e.g., *Cloeon dipterum* [Linnaeus, 1761]; *Cloeon simile* Eaton, 1870; *Caenis robusta* Eaton, 1884);

**Table 2a.** Number of species per family per river basin.

River basin											
Families	Danube: small direct tributaries	Danube main course and flooded zone	Sava	Tisa	Kolubara	Velika Morava	Južna Morava	Zapadna Morava	Timok	Drina	Aegean Sea
Ameletidae	-	-	-	-	-	-	1	1	-	-	-
Baetidae	7	3	3	1	12	7	18	14	13	6	10
Caenidae	3	2	1	2	5	1	6	3	3	0	1
Heptageniidae	9	-	3	1	12	7	14	19	13	6	10
Ephemeridae	1	-	-	-	3	1	5	3	3	2	1
Ephemerellidae	1	1	1	-	3	2	4	6	4	3	4
Leptophlebiidae	5	-	-	-	5	2	5	5	4	4	2
Oligoneuriidae	-	-	-	-	2	1	1	1	1	-	2
Palingeniidae	-	-	-	1	-	-	-	-	-	-	-
Potamanthidae	1	1	1	-	-	1	1	1	1	-	-
Siphonuridae	-	-	-	-	-	-	1	-	1	-	-
Polymitarcyidae	1	1	-	-	-	-	-	-	-	-	-

**Table 2b.** Number of species per family per elevation class.

Families												
Elevation classes	Ameletidae	Baetidae	Heptageniidae	Caenidae	Ephemeridae	Ephemerellidae	Leptophlebiidae	Oligoneuriidae	Palingeniidae	Potamanthidae	Siphonuridae	Polymitarcyidae
Lowland (up to 200 m a.s.l.)	-	8	14	5	1	3	5	-	1	1	1	1
Submontane (200–500 m a.s.l.)	-	18	20	7	4	5	5	1	-	1	1	-
Mountain (500–800 m a.s.l.)	-	9	14	-	3	4	3	1	-	1	-	-
Highland (over 800 m a.s.l.)	1	15	15	3	4	4	6	1	-	-	-	-

LP (limnophil, e.g., *Caenis horaria* [Linnaeus, 1758], *Caenis lactea* [Burmeister, 1839]). Considering current preferences, the most dominant were rheophilic (41), rheo- to lithophilic (13), and rheobiontic (11).

Finally, in terms of temperature, out of 86 species, 6 were cold stenothermal (*Ameletus inopinatus* Eaton, 1887; *Baetis alpinus* [Pictet, 1843]; *Caenis robusta*; *Ecdyonurus*

*austriacus* Kimmins, 1958; *Ecdyonurus subalpinus* Klapálek, 1907; *Ecdyonurus zelleri* [Eaton, 1885]), 10 hot stenothermal (some Baetidae, Caenidae, *Cloeon simile* Eaton, 1870, *Ecdyonurus insignis* [Eaton, 1870], *Oligoneuriella rhenana*), and 45 eurytherm, while for the rest of the recorded species (26) temperature preference was unknown (Table 3).

**Table 3.** Ecological features of Ephemeroptera fauna in Serbia: feeding type and current preference (Schmedtje and Colling, 1996), temperature range preference (AQEM 2002), and conservation status (IUCN 2012).

Species	Feeding type						Current preference <sup>1</sup>	Temperature range preference <sup>2</sup>			IUCN
	Grazers-Scrapers	Shredders	Gatherers-Collectors	Active filter feeders	Passive filter feeders	Predators		COS	WAS	EUT	
<i>Ameletus inopinatus</i>	7	0	3	0	0	0	RL	1			
<i>Acentrella sinaica</i>	9	0	1	0	0	0	RP			1	
<i>Alainites muticus</i>	5	0	5	0	0	0	RP			1	
<i>Baetis alpinus</i>	5	0	5	0	0	0	RB	1			
<i>Baetis fuscatus</i>	5	0	5	0	0	0	RP			1	
<i>Baetis buceratus</i>	5	0	5	0	0	0	RP		1		
<i>Baetis kozufensis</i> <sup>†</sup>											
<i>Baetis liebenauae</i>	4	0	6	0	0	0	RL			1	
<i>Baetis lutheri</i>	5	0	5	0	0	0	RB			1	
<i>Baetis melanonyx</i>	5	0	5	0	0	0	RB			1	EN
<i>Baetis meridionalis</i>	5	0	5	0	0	0			1		
<i>Nigrobaetis niger</i>	4	0	6	0	0	0	RP			1	EN
<i>Baetis pavidus</i>	5	0	5	0	0	0	RP				
<i>Baetis rhodani</i>	5	0	5	0	0	0	RP			1	
<i>Baetis scambus</i>	5	0	5	0	0	0	RP			1	
<i>Baetis tracheatus</i>	4	0	6	0	0	0	RL				
<i>Labiobaetis tricolor</i>	5	0	5	0	0	0			1		
<i>Baetis vardarensis</i>	5	0	5	0	0	0	RB				VU
<i>Baetis vernus</i>	4	0	6	0	0	0	RL			1	
<i>Centroptilum luteolum</i>	6	0	4	0	0	0	RL			1	
<i>Cloeon dipterum</i>	2	0	8	0	0	0	LR			1	
<i>Cloeon simile</i>	2	0	8	0	0	0	LR		1		
<i>Procloeon bifidum</i>	6	0	4	0	0	0	RP				
<i>Procloeon macronyx</i> <sup>†</sup>											
<i>Procloeon pennulatum</i>	6	0	4	0	0	0	RP				
<i>Brachycercus harrisella</i>	+	0	10	0	0	0	RP		1		
<i>Caenis horaria</i>	+	0	10	0	0	0	LP		1		
<i>Caenis lactea</i>	+	0	10	0	0	0	LP				
<i>Caenis macrura</i>	2	0	8	0	0	0	RP			1	
<i>Caenis luctuosa</i>	2	0	8	0	0	0	RP			1	
<i>Caenis pseudorivulorum</i>	3	0	7	0	0	0	RP		1		



Table 3. (Continued).

Species	Feeding type						Temperature range preference <sup>2</sup>				IUCN
	Grazers- Scrapers	Shredders	Gatherers- Collectors	Active filter feeders	Passive filter feeders	Predators	Current preference <sup>1</sup>	COS	WAS	EUT	
<i>Caenis pusilla</i>	2	0	8	0	0	0	RL			1	
<i>Caenis rivulorum</i>	3	0	7	0	0	0	RP			1	
<i>Caenis robusta</i>	+	0	10	0	0	0	LR	1			
<i>Ecdyonurus austriacus</i>	7	0	3	0	0	0	RP	1			
<i>Ecdyonurus dispar</i>	5	0	5	0	0	0	RP			1	
<i>Ecdyonurus aurantiacus</i>	5	0	5	0	0	0	RP				
<i>Ecdyonurus epeorides</i> <sup>*</sup>											
<i>Ecdyonurus forcipula</i> <sup>*</sup>							RP			1	
<i>Ecdyonurus helveticus</i>	5	0	5	0	0	0	RP			1	
<i>Ecdyonurus insignis</i>	6	0	4	0	0	0	RP		1		
<i>Electrogena quadrilineata</i> <sup>*</sup>											
<i>Ecdyonurus subalpinus</i>	5	0	5	0	0	0		1			
<i>Ecdyonurus submontanus</i>	6	0	4	0	0	0	RP			1	
<i>Ecdyonurus torrentis</i>	6	0	4	0	0	0	RP			1	
<i>Ecdyonurus venosus</i>	7	0	3	0	0	0	RP			1	
<i>Ecdyonurus zelleri</i>								1			
<i>Electrogena affinis</i>	5	0	5	0	0	0	RL				
<i>Electrogena lateralis</i>	7	0	3	0	0	0	RP			1	
<i>Electrogena macedonica</i> <sup>*</sup>										1	
<i>Epeorus yougoslavicus</i>							RB			1	CR
<i>Epeorus assimilis</i>	10	0	+	0	0	0	RB			1	
<i>Dacnogenia coeruleans</i>	6	0	4	0	0	0	RP				
<i>Heptagenia flava</i>	6	0	4	0	0	0	RP			1	
<i>Heptagenia longicauda</i>	6	0	4	0	0	0	RP				
<i>Heptagenia sulphurea</i>	6	0	4	0	0	0	RP			1	
<i>Kageronia fuscogrisea</i>	5	0	5	0	0	0	LP			1	
<i>Rhithrogena beskidensis</i>	10	0	+	0	0	0	RB				
<i>Rhithrogena germanica</i>	10	0	+	0	0	0	RB			1	
<i>Rhithrogena fiorii</i> <sup>*</sup>											
<i>Rhithrogena semicolorata</i>	10	0	+	0	0	0	RB			1	
<i>Ephemera danica</i>	+	0	+	8	2	+	RP			1	
<i>Ephemera glaucops</i>	+	0	+	10	0	+	RL			1	

Table 3. (Continued).

Species	Feeding type						Temperature range preference <sup>2</sup>				
	Grazers- Scrapers	Shredders	Gatherers- Collectors	Active filter feeders	Passive filter feeders	Predators	Current preference <sup>1</sup>	COS	WAS	EUT	IUCN
<i>Ephemera hellenica</i> <sup>*</sup>											
<i>Ephemera lineata</i>	+	0	+	8	2	+	RP				
<i>Ephemera vulgata</i>	+	0	+	10	0	+	RL				
<i>Ephemerella mukronata</i>	6	+	4	0	0	0	RB				
<i>Ephemerella notata</i>	6	+	4	0	0	0	RP				1
<i>Ephemerella ikonovici</i>	5	0	5	0	0	0					
<i>Serratella ignita</i>	5	0	5	0	0	0					1
<i>Torleya major</i>	5	+	5	0	0	0	RP				1
<i>Choroterpes picteti</i>	4	0	6	0	0	0	RP				1
<i>Habroleptoides modesta</i>							RP				1
<i>Habroleptoides confusa</i>	0	0	10	0	0	0	RL				1
<i>Habrophlebia fusca</i>	+	0	10	0	0	0	RL				1
<i>Habrophlebia lauta</i>	+	0	10	0	0	0	RL				1
<i>Paraleptophlebia cincta</i>	+	0	10	0	0	0	RP				1
<i>Paraleptophlebia lacustris</i>											
<i>Paraleptophlebia submarginata</i>	+	0	10	0	0	0	RP				1
<i>Oligoneuriella pallida</i>							RP		1		
<i>Oligoneuriella rhenana</i>	0	0	0	0	10	0	RB				1
<i>Palingenia longicauda</i>	0	0	+	8	2	0	RP				
<i>Potamanthus luteus</i>	0	0	9	1	0	0	RP				1
<i>Siphonurus aestivalis</i>	1	+	9	0	0	+	RL				1
<i>Ephoron virgo</i>	0	0	0	10	0	0	RL		1		

<sup>1</sup>Current preference: RB (rheobiont), RP (rheophile), RL (rheo- to limnophile), LR (limno- to rheophile), LP (limnophile).

<sup>2</sup>Temperature range preference: COS – coldstenotherm, WAS- warmstenotherm, EUT- eurytherm.

<sup>\*</sup>Species for which there are no data for the given characteristics.

According to IUCN criteria (Table 3), 4 species were assessed and classified into 1 of 3 categories. *Epeorus yougoslavicus* (Samal, 1935) has been categorised as critically endangered, since this species has a very small distribution with a restricted area of occupancy (5.5 km<sup>2</sup>). A similar pattern of distribution has been recorded for the threatened species *Baetis melanonyx* (Pictet, 1843) and *Baetis pavidus* Grandi, 1949. With an area of occupancy

less than 150 km<sup>2</sup>, these species have been categorised as endangered. Finally, the vulnerable species *Baetis vardarensis* Ikonov, 1962, distributed in south-eastern Serbia, covers an area of occupancy of 160 km<sup>2</sup>.

#### 4. Discussion

In comparison with neighbouring countries in the Balkan Peninsula, the recorded diversity of Ephemeroptera in

Serbia could be characterised as intermediate to high. The highest number of species was 102, listed in Bulgaria (Vidinova, 2003), followed by Slovenia (75) (Zabrc and Sartori, 1997), Romania (72) (Curtean-Banaduc, 2010), Greece (70) (Bauernfeind, 2003), Macedonia (63) (Smith and Smith, 2003), and Bosnia and Hercegovina (51) (Bauernfeind and Soldán, 2012). In Central European countries, the number of species is generally higher with the following pattern: Slovakia with 140 (Derka, 2003), Czech Republic with 107 (Zahrádková et al., 2009), and Hungary with 91 species (Kovács and Bauernfein, 2003). Finally, with regard to Western Europe, Germany showed the highest diversity with 113 species (Haybach and Malzacher, 2003), and Italy had 110 species (Buffagni et al., 2003).

Concerning the Ephemeroptera fauna of the Danube drainage system, some species from the surrounding countries (Slovenia 20, Czech Republic 18, and Hungary 21 species) have not been found in the lotic system of Serbia. In comparison to other drainage systems in Serbia, the Danube basin has been not completely explored in terms of Ephemeroptera diversity. Therefore, there is a possibility that the species recorded in surrounding countries may be found in Serbia in the future.

The presented pattern of Ephemeroptera taxa richness distribution according to the European countries is not only a consequence of the real number of the species that could be expected within a particular unit (administrative, geographical, or hydrological), but also the result of unequal levels of knowledge about this insect order in different countries. Keeping in mind the Ephemeroptera checklists presented for neighbouring countries, as well as the discussion on the general characteristics of insect distribution in Serbia (Radović et al., 1995), the checklist of Ephemeroptera for Serbia is still not complete.

According to long-term research, diversity hotspots of Ephemeroptera in Serbia are situated in submontane regions that encompass the Južna and Zapadna Morava river basins. In line with expectations, the lowest diversity was recorded in potamal rivers, such as the Danube, Sava, and Velika Morava rivers. The contrast in ecological and hydrological features between lowland lotic systems on the one hand and submontane rivers on the other hand is the main reason behind the observed distribution pattern of mayflies. In addition, the Drina River basin presented an unexpectedly low number of species in our study (23), which was presumably caused by the lower density of the network of study sites. Thus, the investigation on mayfly fauna has to be intensified in order to provide better knowledge on this insect order within the country, primarily to be able to provide information on the level of vulnerability of the taxa, as well as conservation priorities and effective protection measures.

Elevation, as a factor which significantly affects macroinvertebrate fauna, forms an important environmental gradient: longitudinal zonation (Vannote et al., 1980; Helson et al., 2006). To what extent elevation gradient affects the macroinvertebrate community has been well documented by many authors (Allan, 1995; Simić, 1995; Simić and Simić, 1999; Lorenz et al., 2004; Paunović et al., 2003, 2006b). Brittain et al. (2003) pointed out that species richness decreased with increasing elevation gradient. In contrast to the conclusions of Brittain et al. (2003), a different diversity pattern emerged in our study. To be precise, the highest diversity was recorded in submontane regions (200–500 m a.s.l.), with 62 species. In a study of Ephemeroptera fauna in the Czech Republic (Zahrádková et al., 2009), it was claimed that when considering species richness viewed against elevation gradient, submontane areas were the most significant.

Rivers in submontane regions generally provide optimal environmental conditions for the survival of many mayfly species. Midsections of running waters are characterised by the greatest species richness and abundance of mayflies, which has been confirmed not only globally many times (Stanković, 1962; Hynes, 1970; Allan, 1995), but also for the waters in the region of Central Europe (Zahrádková et al., 2009) and Serbian waters (Filipović, 1975, 1979; Simić, 1995; Paunović et al., 2006a, 2006b; Paunović, 2007). A combination of different bottom types typically represented in the streams of hilly and lower mountain areas creates a variety of micro- and mesohabitats, thus contributing to taxa richness. In addition, the natural physical and chemical characteristics of water within hilly mountainous streams are in general favourable for the survival and development of mayfly larvae (Bauernfeind and Soldán, 2012). Moreover, the streams of the submontane region of Serbia are under less anthropogenic influence compared to watercourses at lower elevations due to demographic distribution. Namely, the hilly–mountainous region is characterised with lower population density and consequently lower industrialisation (<http://www.sepa.gov.rs>, Report on the State of the Environment in the Republic of Serbia for Year 2011), and thus the aquatic ecosystems are not under the influence of various stress factors arising from human activity. This greatly contributes to the recorded diversity pattern.

Similar results in this country were presented by Simić (1993, 1995), Marković (1995, 1998), Marković and Mitrović-Tutundžić (1997), Marković et al. (1998), and Paunović et al. (2006a), with the highest diversity observed in submontane regions as well, but with the lowest number of species in highland areas, which was not in accordance with our results. High diversity in watercourses of over 800 m could be explained by the fact that all study sites

are near-pristine to reference, which mainly refers to the Timok and Južna and Zapadna Morava river basins. It is also necessary to emphasise that these river basins have been systematically explored in the past (Simić, 1995), which could be a causative factor for such a high number of recorded species.

Besides the natural characteristics of potamon-type rivers in lowland areas (substrate type, flow velocity, microhabitat diversity, physicochemical features of water and sediment, etc.), the presented distribution is certainly a consequence of the presence of stress factors. Namely, in comparison to hilly and mountainous areas, the lowland part of the country is under higher anthropogenic pressure (ICPDR WFD Roof Report 2004; SCG ICPDR National Report, 2004). Human population density, agricultural activities, and industry are mainly located in lowland areas of the country (CORINE Land Cover, 2006). The organic and nutrient pollution and hydromorphological degradation, as the most prominent factors influencing the aquatic ecosystems in Serbia, were consequently found to be the most intensive in northern, lowland parts of the country (ICPDR WFD Roof Report 2004; SCG ICPDR National Report, 2004).

In comparison with the study of Ephemeroptera fauna in the Czech Republic (Zahrádková et al., 2009), the presence of ecological guilds was similar. Concerning the feeding types, in both studies the majority of species belonged to grazers–scrapers and gatherers–collectors. The coincidence in the results also refers to current velocity preferences with the dominance of rheophilic and rheobiontic species.

Four species classified as critically endangered, endangered, or vulnerable are referred to as threatened (Table 3). Besides these, there is one more species with a small distribution area, *Palingenia longicauda* Olivier,

1791, where more information about extinction risk is needed.

The species *Palingenia longicauda*, as a Ponto-Caspian faunal element, used to be widely distributed in almost all European countries as far west as the Netherlands, and it has been recorded in all large rivers in the east (Danube, Volga, and Dniepr) (Haybach and Haase, 2004), and in the north (Lab, Oder, and Vistula) (Klonwska-Olejnik). In Serbia, *P. longicauda* has been found in only one river, the Tisa (Pil et al., 2009), and in neighbouring Hungary, the species has been declared as endangered. However, threat assessment of this species has not been established yet in Serbia. Since this species is already protected in Europe (Habitats Directive EU 2000; Council of Europe 1979 Bern Convention), it is necessary to estimate the level of threat to this species.

Regarding the aquatic ecosystems of Serbia, an intermediate to high diversity of Ephemeroptera larvae has been recorded in comparison with the other Balkan countries. The highest diversity was observed in submontane regions, while the lowest was detected in potamal rivers. The majority of species in this study belong to grazers–scrapers and gatherers–collectors. Out of the total number of species, 4 were categorised as threatened based on IUCN criteria. In addition, it is observed that *Palingenia longicauda* has a small distribution area. Bearing this in mind, we think that further investigation in Serbia should be focused on the necessity of estimating the level of threat to this species.

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